

Nusret Zencirci · Hakan Ulukan ·
Faheem Shehzad Baloch ·
Shahid Mansoor ·
Awais Rasheed *Editors*

Ancient Wheats

 Springer

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Preface – Acknowledgments

The increased role of wheat's wild relatives and underutilized species for food security, sustainability, and crop improvement has been recognized. Wild and domesticated relatives of wheat from the genera *Aegilops* and *Triticum* can be invaluable resources of genetic diversity for crop improvement. However, because of the limitations with studying these wild relatives, most investigations have concentrated on cultivated forms or landraces. Among these, the ancient wheats (einkorn, emmer, spelt, macha, and vavilovii), both in wild and cultivated forms, have received expanding interest in recent years. Ancient wheats are characterized by “non-threshable grain from the hull” during the traditional threshing processes. Ancient wheats originated from Southeastern Turkey, however, are not commonly grown there today. They exist in small, higher elevation regions throughout Turkey, such as Kastamonu, Samsun, Kars, Sinop, Bolu, Bayburt, Gümüşhane, and Kayseri. Generally, these locations have harsher environmental conditions, which may be a reason for farmer preference with these species.

Even though there are multiple species within each ploidy level of *Triticum* (diploid, tetraploid and hexaploid), the most widely grown ancient wheat is *Triticum monococcum* ssp. *monococcum* L., einkorn wheat (*Siyez* or *IZA* in Turkish). This species is grown today as animal feed, and as human food in some places. Cultivation of ancient wheat dates to 12–14,000 years, and expansion of free-threshing, high-yielding wheat pushed this species to small, mostly marginal lands with poor soils or higher elevations. Ancient wheats are grown today in Turkey, Albania, Austria, Spain, and Germany. In addition to einkorn wheat, emmer (*T. dicoccum* Schrank) and spelt wheat (*T. spelta* L.) are other noteworthy forms of ancient wheat species. Lesser-known ancient wheat species include *T. macha* Dekarp. & Menabde, and *T. vavilovii* Thum. Efforts on wheat breeding since ancient times are mostly concentrated on grain yield. The most important yield component was starch content. Since starch is mostly stored in endosperm tissue, larger endosperm has been a preference by farmer and consumer. With grain yield as the most important trait of interest, gluten, protein, starch contents, and biotic and abiotic stress tolerance/resistance have been additional aims in most breeding programs.

However, phytochemicals and bioactive materials were neglected, due to difficulties in evaluating these traits and adding to breeding programs. However, there is a growing interest, today, in the evaluation of these compounds in wheat's wild and cultured relatives. One of the reasons for this attention is the number of people affected by micronutrient malnutrition. Approximately 3 billion people are affected by some form of micronutrient malnutrition. Zinc, iron, and selenium deficiency affect around 0.5–1 billion people around the world. Finding novel allelic diversity for higher micronutrient content is essential to prevent health issues they cause. Some species/accessions are good on micronutrients but low on starch content, which is partly due to larger seed size on domesticated species. Like starch content, there is a considerable variation in dietary fibers, phenols, terpenes, alkaloids, carotenoid, and vitamin contents (B and E). Different parts of the world have different nutrient deficiencies or toxicities (e.g., zinc deficiency in Turkey, or vitamin A deficiency in Eastern Asia). Phenols, terpenes, and alkaloids are essential compounds for our immunity and well-being.

Ancient wheats were widely grown in ancient times. One obvious reason for the loss of interest in the years was the additional processing of the grain to remove the hull prior to consumption and the higher grain yield of common (*Triticum aestivum* L.) and durum wheat (*Triticum turgidum* ssp. *durum*). Other reasons have generally been the end-user preferences such as bread-making quality, pasta-making quality (common and durum wheats have generally strong gluten compared to ancient wheats), and larger seed size (16.7–33.4 g einkorn, 28.9–41.2 g bread wheat, and 46.0–58.4 g durum wheat) in wheat species. Technological advances allowed end-users to search for alternative food solutions. Two wheat species getting public attention in Turkey are einkorn “Siyez-IZA” and emmer “Gernik” wheat. Both species are generally grown across the western Black Sea region and are preferred for bulgur taste, texture, color, fiber content, and other nutritional values. Einkorn wheat tends to have a higher protein content than common wheat. However, einkorn grain yield is lower than common wheat, so the advantage of protein content brings disadvantage of grain yield.

Starch content of einkorn, emmer, spelt, common, and durum wheat is similar (all between 60.6–72.9%); however, amylose (a source of resistant starch) and amylopectin rates in these species may vary. The lower glycemic index is recommended for a healthier diet and spelt wheat may have tenfold more resistant starch. Wheat, when consumed as whole-grain bread, is the biggest source of vitamin B complex. It provides between 10–40% of B1, B2, B3, B6, and B9 vitamins. Phytochemicals and antioxidants such as ferulic acid, tocopherols, phytosterols, and carotenoids can be found at high amounts in einkorn wheat when compared with common wheat. Einkorn wheat has lower values for β -glucan than other wheat species. Also, rheological analysis for above species suggests that common and durum wheat are better qualified as bread and pasta, respectively. But this does not eliminate einkorn and emmer from bread making. Einkorn flour is suggested as a good source for “bazlama,” a sort of flat bread of Turkey. Ancient wheat has various end-use options, including bread, pasta, erişte (Turkish home-style noodle), bulgur, and cookie/biscuit. Even if einkorn has high-quality traits, it may not be consumers' preferred

product, because of darker color of its pasta and bulgur. Einkorn wheat flour produces smaller loaf volume, compared to common wheat flour. Given that many of the results highlighted here are from only limited observations, there remains a need for in-depth evolutions of einkorn, emmer, and spelt wheat and their certain mixes for bread, pasta, cookie, and other consumer products. Phytochemical, antioxidant, and vitamin contents of ancient wheat should be evaluated distinctively and included in breeding objectives for these species.

Global wheat production needs to increase by 50% by 2050 to feed our growing populations. Consumer preferences extend while a country develops, from classical consumer to more selective consumer that expects healthier, high-quality foods. Einkorn, once an animal feed, has become a high-end product. Einkorn products such as cookies, bulgur, flour, and bread are selling at higher prices than common wheat products. Future projections suggest an even bigger trade value for these species, especially for einkorn and emmer. Growing trade potential of these crops can not only help gain interest for breeding purposes but also help germplasm resource protection.

This book **Ancient Wheat** is an **extended version** of Hulled Wheat with some new chapters included to cover current issues better in ancient wheat.

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Bolu, Turkey

Nusret Zencirci

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Chapter 1

Introduction



Kürşad Özbek and Faheem Shehzad Baloch

1.1 Introduction

Plant genetic resources constitute the basis of worldwide agricultural development and assure our safeguarded future. There is a continual daily trend in the loss of the existing crop diversities, which were caused by a variety of reasons. It is unfortunate that genetic resources are, too, exposed to a serious level of erosion. Man should, therefore, give the increased attention to preserve the genetic diversity harbored by genetic resources. Plant genetic resources, which have unfortunately been neglected and underutilized, doubtlessly gain the desired interest today. This will hopefully safeguard the future of mankind. Thanks to the advanced technology, man has much realized the potential of these genetic resources. The world with a shrunk area for man must feed the fast-rising populations. For this purpose, it is vital that primary importance must be attached to this genetic diversity legacy so that our future generations can survive.

Turkey is situated on such a geography that creates a unique atmosphere for plant genetic resources. With 11,707 taxa and 3035 endemic species (Güner et al., 2012), Turkey has an important place in the world. The discovery of a very new species once every 5 days gives an idea about the plant biodiversity in the flora of the country, with a major part of endemic species, local varieties, and the crop wild relatives that have directed agriculture worldwide. Among them, wheat emerges of utmost importance for the world history, economy, and culture. Wheat is a species with around 25,000 cultivated cultivars of bread and durum wheats, which occupy the highest share. Bread wheat (*Triticum aestivum* L.) makes up the 95% of the wheat

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consumed in the world today. The rest 5% is the hard pasta or semolina wheats (*T. turgidum* ssp. *durum* Desf.).

Bread and durum wheats were cultivated from the start of the evolution from the wild wheat. Ancient einkorn (*Triticum monococcum* ssp. *monococcum* L.) is the most primitive form of wheat, cultivated in the same way, though not widespread in production. *Siyez*, -and IZA- a Turkish word for einkorn, is referred to both emmer and einkorn in different parts of Turkey (Karagöz, 1996; Ertug, 2004; Giuliani et al., 2009) and is being cultivated favorably for its palate taste. On the other hand, einkorn is generally susceptible to leaf diseases and is not responsive to fertilizer applications (Özberk et al., 2005). Because of having long plant height, it remains laying down on the ground.

Ancient wheat botanically refers to *Triticum* populations, which grow wild or cultivated, and has grains inseparable from the hull through classical threshing. Ancient wheats (einkorn, emmer, spelt, macha, and vavilovii) are known to be the most ancient cultivated species. These crops had been widely planted as man's basic food in their own regions across the world for hundreds of years. With the start of utilization of high-yielding wheat cultivars with easier threshing, hulled wheats, however, came to the verge of their production fields over time. It is pleasing nowadays that ancient wheat has gained popularity and a substantial recognition again simply because of social, cultural, economic, or scientific reasons. Ancient wheat, which formerly used to be cultivated only by low-income farmers, was the food of the poor. As a result of its significant awareness, it has now been preferred much more than ever especially by discerning consumers, and its products are top-placed among those expensive ones in the markets. Even though this is the case at present, there appears to be a further need to take efforts to raise more public awareness for an increased production of this niche product in the markets. Since einkorn is planted by small-scale producers in particular, any negative fluctuations in the prices may adversely affect them and in turn lead to the point of extinction.

Einkorn is the species domesticated from wild wheats, einkorn (*T. monococcum* ssp. *monococcum*), or *T. boeoticum* Boiss. Einkorn also showed distribution in mixture with the cultivated emmer (*Triticum dicoccum* Schrank.) in the early ages. It is suggested that einkorn was domesticated for the first time in [Karacadag](#) in Southeast Anatolia (Kimber & Sears, 1983). First cultivated form around 10,000 years ago, einkorn is also regarded as the ancestor of the modern wheat and the simplest wheat with its chromosomes of $2n = 14$, which places einkorn in the class of crusted wheats. This species was first called "Ziz," a Hittite word, which was cultivated by the Hittites and Phrygians, whereas the name later turned to "Siyez/IZA" and sometimes "kaplica." Today, its harvest is still being carried out in North Anatolia, Balkan countries, Germany, Switzerland, Spain, and Italy. Siyez owns a structure that differs from *T. dicoccum* Schrank. (in Turkish; Gernik or Kavılca) variety, called *emmer* in English.

The most important distinguishing feature of ancient/hulled wheat, as the name implies, is that it bears a hulled structure inseparable from the grain with the standard threshing operation. The thick and sturdy glume provides a perfect protection

to the grains during harvest under field and storage conditions, even if it appears to be a problem during the processing.

Because of many similar reasons, einkorn flour and its other characteristics show some differences from bread and durum wheats. That is, the glume is not the only feature that distinguishes ancient wheat from that of the other wheats species, studied as a separate group.

1.2 History of Wheat

Wheat is the primary agricultural product that played a crucial role in the development of civilizations throughout history. Compared to the other products for a food-stuff, wheat has ever been found exclusive in agriculture, and whenever its harvest resulted in lower yields, people had to cope with the starvation. It is assumed that wheat spread worldwide from the region known as the Fertile Crescent (Baloch et al., 2017). Many studies supported the idea that Southeast Anatolia is the birthplace of wheat and the source area of its gene (Lupton, 1987; Salamini et al., 2008). Wheat was cultivated for the first time in this region as well (Heun et al., 1997).

Such agricultural products of crucial importance as wheat forced man to shift from the nomadic hunter-gatherer lifestyle, which had been traced for thousands of years, to the settled-producer life, because the process of sowing and harvesting required long periods of settlement in one region. The Fertile Crescent, which embraces today's Iran, Iraq, Turkey, Syria, Lebanon, Israel, and Palestine, is the region where mankind first started the settled life and agriculture. The most apparent reason for this is that the region is the gene center of many wild cereals, especially of wheat and barley, and that man had become aware of the easy availability of those small and nutritious wheat and barley grains and started to use them. Before getting involved in agriculture, man had just collected wild grains of wheat and barley from nature to consume for food, and over time, they learned how to plant and harvest these species. Certain historical studies show that wheat agriculture later turned to be an indispensably traditional part of human life and shaped the social values throughout history (Nesbitt & Samuel, 1996).

Wheat samples were found in the archaeological excavations carried out in Turkey (Table 1.1). According to the recent findings, the first cultivation of wheat took place about 12,000 years ago (Hirst, 2017). Ancient wheat species, einkorn (*T. monococcum* ssp. *monococcum*), emmer (*T. dicoccum* Schrank.), and spelt (*Triticum spelta* L.), were the oldest domesticated cereals along with barley, pea, lentil, chickpea, bitter vetch, and flax, as some archaeo-botanic excavations indicated (Perrino et al., 1996; Zohary & Hopf, 2000; Colledge & Conolly, 2007; Zohary & Weiss, 2011). For example, excavations at Çatalhöyük indicated that their storage was fulfilled with utmost care under so special conditions than the other species (Helbaek, 1964; Fairbairn & Martinoli, 2005; Bogaard et al., 2012).

According to archaeological and genetic studies, it was found that the Karacadağ area in the southeast of Turkey is the origin and agricultural start of two of the eight

Table 1.1 Cereal residues found in some excavation sites in Turkey with their dates (Harlan 1995)

Approximate date (BC)	Location of excavation	Plant residues
7200–6500	Çayönü	Wild einkorn, emmer, and barley; cultivated einkorn, emmer, peas, lentils, vetch, and flax
6750	Hacılar	Wild einkorn, cultivated emmer
6500	Can Hasan	Wild einkorn; cultivated einkorn, emmer, wheat, barley (2 rows), lentils, and vetch
6000–5000	Çatal Höyük	Cultivated einkorn, emmer, wheat, barley (bare), peas, and vetch
6000–5000	Erbaba	Cultivated einkorn, emmer, wheat, barley (2 rows and bare), peas, lentils, and vetch

species: three cereals (einkorn wheat, emmer wheat, and barley), four legumes (lentils, peas, chickpeas, and bitter vetch), and one oil and fiber crop (flax) (Özkan et al., 2002; Andeden et al., 2013). Recent studies on the samples of wild and primitive (einkorn) wheat showed that diploid einkorn wheat agriculture started in Karacadağ area (Heun et al., 1997). Though it was cultivated a long time ago, however, its production has dropped to a great extent in our time. Emmer, a tetraploid species, was also domesticated in ancient times, but now its production is much rather lower. Its primitive cultivation started 7000–9500 years ago when farmers selected tetraploid wheats from its wild relatives (*T. dicocoides* Korn.) for a mainly production during the Bronze Age. In all the regions where einkorn is produced, emmer had been the most sprawling one among the ancient wheat species until agricultural mechanization started. *T. spelta* (spelt) is, on the other hand, a hexaploid species of *T. aestivum* origin, with quite a limited production (Perrino et al., 1996).

The initiation of wheat farming affected the life of mankind from all aspects. Wheat species first planted for production purposes in ancient times has as well recorded an evolution because of man's selection of seeds for farming. In the first farming villages, people planted two species of ancient wheats (*T. monococcum* ssp. *monococcum*/*T. dicocum* Schrank.), which owned glumes like the wild wheat and harbor more brittle color than today's wheat cultivars but with bigger grains. With the improvements in agriculture in later historical ages, farmers started to have a compulsory habit of sorting out the larger wheat grains, which could be more advantageous for their next planting time. This process each time assured the next year's harvest, resulting over time in the emergence of two taller wheat species with big easily threshed grain. They are durum (*Triticum durum* Desf.) and bread wheats (*T. aestivum*) of today.

In some high plateaus of Turkey, Italy, and Greece, it is observed that einkorn and emmer are planted just for farmers' self-sufficiency purposes on a restricted field. In some regions of the world, there are other wheat species or subspecies in limited acreages grown, suitable for the region's local climate and soil conditions. In addition, other species such as spelt (*T. spelta*) in Europe used to be planted on a large-scale area in the past. It is not available in satisfactory quantities today. Advanced technology enlightened the evolution the wheat has gone through and its

origins much better than ever today. Our experience and knowledge in these fields have so far indicated that Anatolia offers the potential to unearth invaluable data for man to further raise the productivity and quality of wheat for the next generations.

1.3 Other Ancient Wheat Varieties

Hexaploid wheat with a chromosome number of $2n = 42$ is the bread wheat group. The rachis in the group lengthens especially in speltoid forms. This group comprises sphaerococcum subspecies with the shortest rachis. The rachis is salient all along the middle section of the length of the outer glume from the top. The lower section of the outer glume is wider than those in the other groups. For this reason, grain pullout among the group's bare granular cultivars is an issue of concern. The group's cultivated hulled cultivars are spelta, macha, and vavilovii (Chao et al., 1989; Kam-Morgan et al., 1989).

Many studies have been carried out up to the present on these ancient wheat species. Common bunt resistance was observed frequently in *Triticum vavilovii* Thum., *Triticum timopheevii* Zhuk., *Triticum dicoccum* Schrank., and *Triticum macha* Dekapr. & Menabde., but rarely in *Triticum spelta*. All accessions of *T. macha* and the few accessions of *T. vavilovii* and *T. karamyshevii* were vulnerable to rust (Borgen, 2010). *Triticum macha* seemed a good donor plant (Buerstmayr et al., 2011) against *Fusarium* head blight.

1.3.1 *Triticum macha* Dekapr. & Menabde

Ancient hexaploid, *T. macha*, was first identified in 1929. It was widely planted in the Caucasus until the 1930s. It has a stable crop yield under different climatic conditions and is known to have a strong resistance to various diseases (Barisashvili & Gorgidze, 1979).

The ear morphology of *T. macha*, a late popping variety, is quite different from that of bread wheat. With a dense, squarehead, non-compact spike phenotype, *T. macha* possesses a fragile rachis and hairy and waxy glumes. It is only partly free blending. In comparison to *T. spelta*, *T. macha* does not develop a speltoid ear shape, which is illustrated as a spear-shaped spike with elongated rachis. The European ancient *T. macha* bears the q allele, whereas the Iranian hulled carries Q (Simons et al., 2006). Macha possesses a good intraspecific diversity. Macha's high protein content grain makes it high quality. Ten to twelve varieties in macha significantly show difference for spike morphology and yield (Borgen, 2010).

1.3.2 *Triticum spelta* L.

Spelt varieties can be either winter or spring habit. Spikes are very lax, long, and round or almost square in cross section. They have no awns or harbored short awns. The rachis is brittle. After the threshing procedure, the base of the rachis internodes across the back of the spikelets remains attached to the base of the spikelets. The spikelets bear double kernels, and they are closely appressed to the rachis. The glumes are tightly clinging and have wide, square shoulders. The beaks are short and obtuse. Following the threshing, the kernels are left encircled in the glumes. They are pale red, laterally compressed, with an acute tip. The tuck is narrow and shallow (Briggle & Reitz, 1963).

Triticum spelta does not need any special condition to grow as it has high adaptation ability and it bears an alternative to the other wheat species with its hulled structure. In high plateaus, it can be more productive than *T. aestivum* (Ruegger & Winzeler, 1993). Even with less fertilizer application, it likely yields better crops. Its mineral contents are higher than *T. aestivum* (Moudrý & Dvořáček, 1999). It is advisable, as well, for areas with water shortage (Háp, 1995).

In addition, *T. spelta* has high values of nutrients such as saccharides, proteins, fiber, lipids, vitamins, and minerals (Bognar & Kellermann, 1993; Abdel-Aal et al., 1995). Especially its protein rate (16–17%) is higher than *T. aestivum* (Smolková et al., 2000), which means that products made from hulled wheat are not recommended for people suffering from celiac disease (Kasarda & D'Olivio, 1999). The content of gluten is higher in *T. spelta* in comparison to *T. aestivum* (Moudrý & Vlasák, 1996; Stehno et al., 1998). The flour obtained from *T. spelta* is favorable because of its bread-like smell as being an exclusive characteristic in bread production and good volume, flavor, and long-lasting freshness and softness (Bojčanská & Franěáková, 2002).

1.3.3 *Triticum vavilovii* Thum.

Being a hexaploid variety, *T. vavilovii* has elongated rachillae that give the ear a branched appearance (Singh et al., 1957). It was discovered as a rare admixture of *T. aestivum* near Lake Van, eastern Turkey in 1929 (Zhukovsky, 1933). Its close link is *T. aestivum*, apart from the two closely related genes controlling branching and the hulled character, and it can be considered that *T. vavilovii* is another local, which recently evolved from wheat that hardly rates species status (Padulosi et al., 1995).

It is of winter habit and midseason. The straw is thick and very strong. Spikes are mid-dense to lax, with short awns. They are alike to *T. spelta*. *Triticum vavilovii* spikes show a branched appearance. This characteristic is unique and does not occur in other *Triticum* species or subspecies. The rachis is brittle but more resistant to crushing than *T. spelta* or *T. macha*. The glumes are coarse and wide with a square shoulder (Briggle & Reitz, 1963).

1.4 Ancient Wheat Production

In European countries, ancient wheat production currently seems to have stuck into some marginal fields with low fertility soil. It is basically produced for feeding animal as well as for bakeries such as pasta and biscuit production and similar other kinds of products in Albania, Austria, Germany, Greece, Italy, Switzerland, Spain, and Eastern Europe (Maggioni, 2000; Buerli, 2006).

Ancient wheats can also be observed in Turkey's marginal crop areas. Einkorn and emmer adapt good to mountainous areas with harsh environments. Einkorn cultivation turned to be a traditional process for the communities who planted it for thousands of years. Its consumption by inhabitants has presently been widespread in the form of bread and "*bulgur*" (cracked and parboiled grains) as well as live-stock feed in Turkey. Since wheat and bulgur production are still being carried out with traditional methods to a large scale, there appears no need for fertilizing and spraying any pesticide nor much irrigation, which, additionally, creates positive influences for the environment. Hence, einkorn is not a cereal that requires high inputs, with its production supporting the farmer's economy as well as showing environment-friendly aspects. Because the farmers' income level in these regions is not enough to make a living, they need to plant einkorn just for subsistence. After all, there has been a rise in the acreage of sowing for commercial purposes in recent years.

In Turkey, the major einkorn planting zones are İhsangazi, Seydiler, and Devrekani districts of Kastamonu Province in West Black Sea Region. Other provinces are Bolu, Sinop, Bilecik, Düzce, and Samsun. Though many farmers in Kastamonu have a rising habit of applying fertilizers and pesticide for better crop yield in quantity and quality, traditional methods are still overwhelmingly applied in most areas.

The West Black Sea Region is quite mountainous and covered with dense forests. In the region, hulled wheat is planted at elevations from 600 to 1200 m. Farmers presently are involved on a large scale in planting only emmer or a mix of einkorn and emmer. Farmers tend to mix up einkorn and emmer during the sowing process, as was the case in the historical past (Jones & Halstead, 1995). Harsh climatic conditions, such as heavy snowfall, rough mountainous terrain, and steep cultivation areas in the region, make transportation and communications difficult, and this creates a complete isolation in the region at certain times of the year. This, on the other hand, has helped einkorn wheat remain conserved in this region.

Ancient wheat agriculture is frequently performed as a small-scale production activity solely through household efforts mainly due to the limited availability of arable land for the crop. Furthermore, the production of ancient wheat species necessitates more labor as compared to the modern varieties, which forces farmers to collaborate during the harvest, threshing, and bulgur-making seasons. That is why ancient wheat agriculture has over time turned out to be a ceremonial part in the social life.

Einkorn and emmer consumptions in the regions generally differentiate, although both are to a great extent utilized as bulgur and flour for human consumption as well as animal feed. Their production records have a downward trend day by day. There is a publicly rising awareness of the advantages of this ancient crop (e.g., high fiber, low carbohydrate content) in terms of nutritional and flavor benefits against the modern cultural varieties. Discerning consumers especially residing in metropolitan cities have had an appreciation of this difference for their concerns over health improvement (Ünal, 2009; Giuliani et al., 2009). Meanwhile, the number of traditional stone mills that were once operated to grind wheat to make bulgur, the basic foodstuff for most of the people in the region, is declining each day, and the production by those mills can no longer meet today's rising demand. On the other hand, bulgur consumption statistics show a steadily upward trend with an average of 12 kg/year per person in the country (Ünal & Sacilik, 2011). Bulgur, regarded publicly as an exclusive food in the region, has played an important role in shaping the social life as well. However, it is due to certain socioeconomic reasons and the widespread use of modern farming methods and migration from rural areas to urban areas that ancient wheat has been replaced by modern wheat cultivars. Moreover, a treasure of traditions and knowledge gained by man as a result of life-long occupation with that species, as transmitted through generations, has been faced with the danger of disappearance.

Farmers conduct traditional methods for storing hulled wheat to meet their seed requirements and for other purposes. The storage of hulled wheat is carried out in wooden boxes in houses suitable for short-term storage, enough to meet annual needs only, or in depots called “*ambar*” in larger dimensions (with an average 500 kg capacity) in 2-store wooden buildings.

1.5 Utilization

Ancient wheat species have been cultivated by mankind for thousands of years. Even if the overall area where hulled wheat is sown has declined to some extent today, it is still grown in marginal areas because of its hardness and primary place in local food traditions and for its use as animal feed. At the same time, it is found special by today's niche consumers for its nutritional value and flavor.

Among the hulled wheat species, hulled wheat is the one that is found the most favorable by people in Turkey. Siyez wheat is utilized for a variety of purposes, and it has attracted the attention of even science and industry very recently. It also carries traditional significance for residential life in the regions where it is grown. Turkey's first presidium application that Siyez was selected for was made in Kastamonu, where it is the most plenty harvested today.

People consume Siyez mostly in the form of bulgur, the main traditional food in the country. Made of hulled wheat, which bears a hulled structure with single-grained spikelets, bulgur is obtained through parboiling, drying, and finally crushing the grains in a completely traditional manner in the old-type stone mills. Hulled

bulgur has a marketing potential in Kastamonu and its environments where the production figures are the highest of all. The local people who plant hulled still make use of it as animal feed, nonetheless: A major portion of the production (80%) is used to feed animals, while the rest of the product (20%) is usually converted into bulgur in stone mills (Ünal, 2009).

There is a growing public appeal toward utilization of hulled bulgur. Those who taste hulled bulgur mostly out of einkorn once are likely to be addicted to it because of its unique taste. As a delicacy, it is advisable to try distinctive flavors made of Siyez, for example, *Siyez pilaf* (with tomato sauce or yogurt), *Siyez dolması* (grape leaves stuffed with emmer bulgur), or *Siyez soup*. Moreover, loaves of bread made of Siyez flour have recently started to be sold in the markets upon this growing demand.

Emmer, which was consumed especially as bread making in Turkey in the past, was almost disappearing in the Near East and Europe during the second half of the twentieth century. It is today used by farmers with low-income level for feeding their livestock at an elevation of between 1000 and 2000 m above sea level in the environments of Kastamonu and Sinop (Özkan et al., 2011). Having been harvested mostly in the Near East formerly, *T. spelta*, another hulled wheat species, is nowadays grown in some European countries only, especially in Germany and Switzerland, for production of bakeries like a variety of pasta products, muesli, and flakes (Laghetti et al., 1999). Especially common in the Caucasus region, *T. macha* is preferred because of its high protein content (Borgen, 2010; Barisashvili & Gorgidze, 1979).

Hulled wheats provide significantly high values of beneficial herbaceous chemicals and antioxidants for human health (Köksel et al., 2008; Serpen et al., 2008). Furthermore, einkorn and emmer, with their differing values of starch properties from the other extensively sown wheat species, are of interest to the food industry. It is also known that einkorn wheat supplies such a low nutritive and glycemic index that makes one feel satiated. Einkorn, generally consumed as bulgur, is a valuable nutrient with high protein, phosphorus, antioxidant, and digestible fiber contents.

According to the other studies conducted on the potential areas of utilization of kaplica wheats, it was found that the crackers (Kütük et al., 2008), biscuits (Öztürk et al., 2008), and sorts of pasta (Gümüş et al., 2008), which are made from these wheats, yield very similar to those standard samples and that they could be grown for these purposes (Akıncı & Yıldırım, 2009).

Einkorn wheat is habitually processed to produce whole wheat flour and bulgur for pilaf wherever it is grown. Food companies operating in accordance with international regulations and at specified production standards utilize this precious cereal to produce many other products such as pasta, ravioli, and cookie.

Einkorn wheat flour has already proven its potential to replace white flour in all the fields where the latter is utilized classically, and hence, it has attained the level of becoming one of the basic components particularly in the scope of einkorn bread preparation.

Another popular product derived from einkorn wheat is *Siyez bulgur*, which has been at a premium. It helps einkorn wheat be further promoted on the market

nationwide. Einkorn bulgur, with such other fields as pilaf, kısır, and meatballs, can be used safely as a substitute for the dishes that contain rice in Turkish dining today. Being satisfactory with respect to taste and nutritional values, this species is worthy of being accredited, which makes it exclusive compared to the most popular einkorn wheat products.

“*Erişte*” (homemade noodles), derived from einkorn flour as well, is regarded as one of the most favorable tastes of Turkish culinary culture. *Erişte* can be stored for a long time because it is prepared entirely free from moisture. Made of 100% einkorn flour, einkorn noodle is dried up in a natural manner, and with the addition of eggs that raises its level of tastiness, einkorn noodle enhances its potential of being in demand as one of the most palatable einkorn selections.

Apart from the varieties of food mentioned above, einkorn flour also finds its worthy place in the production process of world flavors. Thanks to einkorn flour ingredients, a new sense of cookery has emerged, and fruitful results have been obtained in the preparation of acknowledged Italian origin pasta assortments, ranging from *orecchiette*, *fusilli*, *penne*, and *tagliatelle* to traditional *Siyez ravioli*, *Siyez cheese stuffed pasta*, etc. *Siyez* flour, which has a fast-cooking feature, a sweet aroma, and a more darkening color when kneaded, enables these pasta varieties to replace white noodles.

There are dozens of varieties ranging from the pastry product *Siyez rusk*, *Siyez leaf rusk*, and *Siyez-added classical French baguette* to sour fermented *Siyez bread*, sesame oil *Siyez muffin*, and *Siyez bread with walnut*, sun-flowered *bagel*, and *Siyez cookies*, all of which ever have the einkorn maintain its roots.

Consequently, it seems certain that the ancient wheat species would apparently find its deserving prestigious place all over the world, relying on scientific research into, and correct findings on, its differences from the white and other sorts of flour, as well as with the ever-growing support from regional administrative units, NGOs, and relevant global organizations’ focus on local products, producers’ rising awareness of the matter, and the multiplied effect that arises from globally efficient companies’ closer attention to this species, all leading the relevant options to diffuse into a wider geographical area across the world.

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Chapter 2

Domestication and Evolution of Ancient Wheats



Özlem Özbek

2.1 Introduction

Wheat is one of the most prominent crop plants, discovered approximately 10,000 years ago, and has great importance as a major food source in human diets worldwide. In the Paleolithic era, humans used to hunt wild animals and gather the leaves, fruits, seeds, and roots of wild plants to feed. The climate conditions became milder and favored plant diversity in the natural environment. This factor might have enriched and diversified the plant food resources for human food and led to an increase in population size that brought about the requirement for an increase in food resources, because the collection of only wild plant food resources was not enough to supply the increased populations' needs. Therefore, in the late Paleolithic and early Neolithic eras, humans started to collect the seeds of wild emmer wheat for food, and some were left for the next year's planting. After the plants ripened, they were harvested, or they were harvested before maturation, as wild emmer wheat has a brittle rachis, which ensures independent seed dispersal and self-burying of the seeds into the soil, thus giving rise to the first form of primitive agriculture. This was the huge revolution in human lifestyle, which arose in the Levantine region and then spread to the Fertile Crescent, Europe, and Africa; it was thought that the transition from a hunter-gatherer lifestyle to a sedentary lifestyle started with agriculture.

Charred wheat grains, reaps made with silex attached to animal horns, stones used for grinding (millstone-like) processes, and cereal storage wells were found in archaeological remains in Diyarbakır Çay Önü (7000 BC), Konya Çatal Höyük (6800–5700 BC), and Burdur Hacılar (6750–6500 BC) in Anatolia (Kunter, 2011)

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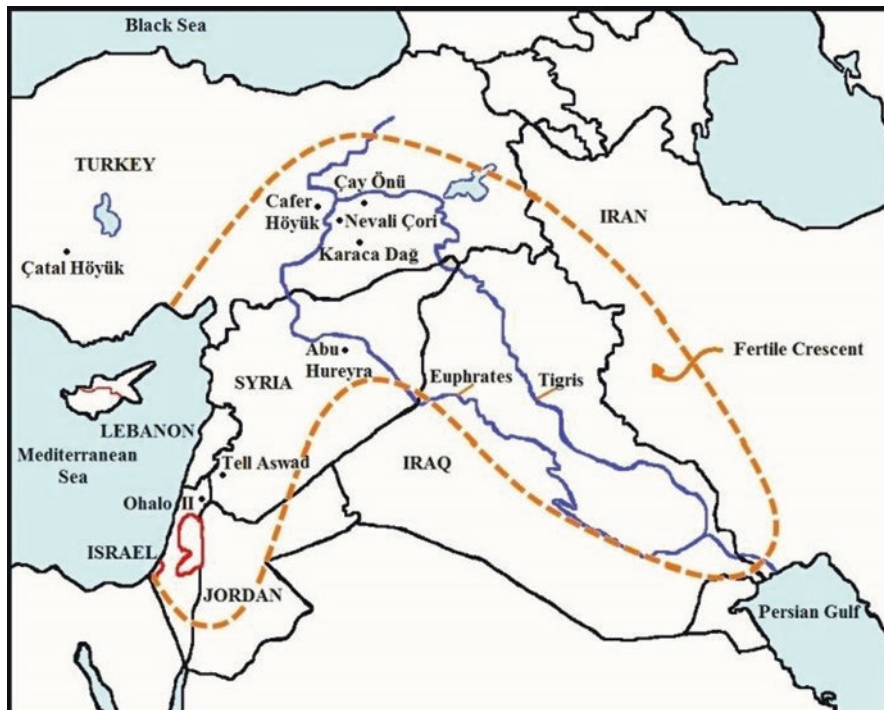


Fig. 2.1 The map is representing the domestication geography of wheat and excavated archaeological sites, where wheat grains were found, in the Fertile Crescent (orange dashed region) represented on the Near East

(Fig. 2.1). These findings suggested that people were harvesting and grinding cereal grains and stored them as food resources. Most probably, they cooked the ground cereal flours as food or bread-like food in ash.

According to the archaeological remains, in 4000 BC, people cooked bread in bakeries in Babylon. It was thought that the ancient Egyptians used the first leavened dough to make bread. There is even a legend about a bread made with leavened dough. Once upon a time (around 2600 BC) in ancient Egypt, slaves were charged with the task of making bread, and 1 day, the slaves had fallen asleep and the fire went out; the dough was left in a warm environment, and it fermented and became sourdough. When the sourdough was cooked, they noticed that the bread was softer, more delicious, and more easily digested than flatbread. After that, they started to cook bread leavened by fermentation. Therefore, it is thought that fermented bread spread out from Egypt and Israel (Kapucu, 2015). The ancient Greeks learnt how to make leavened bread from Egyptians and Jews. In 312 BC, there were 254 bakeries controlled by governments in Rome, and there were bread standards (e.g., bread weight and price). Turks knew how to cook flatbread (pitta), a thin leaflike bread, sac bread, and a thin pitta-like (lavaş) bread, from non-fermented dough; these breads had a long life and kept fresh when the people had a nomadic lifestyle.

Bread wheat is used to produce many products, such as cookies and bread, while durum wheat is used to make items such as macaroni, couscous, bulgur, and home-made macaroni. In the beginning, wheat was ground between stones and emulsified with water and then cooked on heated stones or in ash.

2.2 Evolution of Wheat

There are many theories, proposed by different research groups, about the origin of wheat. Solms-Laubach (1899) thought that the cultivation of all wheats started in Central Asia and then migrated westward, while Much (1908) proposed that many cereals, especially wheat and barley, were cultivated in Europe and domesticated there (Feldman & Millet, 2001). Körnicke saw a spike segment of two-grained wheat with brittle rachis found by Kotschy in Rashayya on the north slope of Mt. Hermon in 1855, in the National Museum, Vienna, in 1873. Körnicke claimed that the two-grained wild progenitor of wheat originated in this area. Then, he made expeditions to find it in this area, but he did not succeed. Aaronsohn made an expedition to this area in 1904, but he could not find this wheat too. Aaronsohn made a second expedition in 1906, and he found a single plant at Rosh Pinna in Eastern Galilee, Israel. After that, Aaronsohn saw this wild wheat on Mt. Hermon and other sites in Israel, Jordan, Lebanon, and Syria (Aaronsohn, 1910). After this great discovery, it was assumed that the two-grained wild wheat (*Triticum dicoccoides*) was the progenitor of most cultivated wheats, and this enabled a search for the origin and evolution of wheat by cytogenetic methods (cited in Feldman & Millet, 2001).

Wheat belongs to the subfamily Pooideae, which diverged 20 million years ago (MYA) from the family Poaceae (grasses), which evolved 50–70 MYA (Peng et al., 2011). The tribe Triticeae started to diverge from its progenitor approximately 35 MYA and the *Triticum* genus diverged from its progenitor about 11 MYA. *Triticum* and *Aegilops* taxa evolved from a common ancestor, with seven basic chromosome numbers, about 3 MYA (Gustafson et al., 2009).

When an organism has more than a diploid complement of chromosomes, the situation is called polyploidy, which is the basic driving evolutionary force in the evolution of wheat and many crop plants (Stebbins, 1950, 1971). There are two types of polyploidy; hybridization between two species with different genome formulae, called allopolyploidy, a good example for the wheat speciation based on which. The other type of polyploidy is whole-genome duplication, in which the existing organism may have at least three copies of the same (or a near identical) genome. The new organism existing as a result of either type of polyploidization will have multivalent pairs or, in autopolyploidy, random bivalent pairs between homologous chromosomes (Renny-Byfield & Wendel, 2014). Polyploidy contributes to polyploid plant species being able to adapt to diverse environmental conditions by buffering the effect of their genomes and may provide phenotypic divergence (Soltis et al., 2009).

In the *Triticum* genus, there are two wild diploid species with genome A, *Triticum monococcum* ssp. *aegilopoides* ($2n = 2x = 14, AA$) and *Triticum urartu* ($2n = 2x = 14, A^uA^u$) (Fig. 2.2), which diverged from its progenitor about 2.6 MYA (Fig. 2.2). All these diploid wheats, except for *Triticum monococcum* ssp. *monococcum* ($2n = 2x = 14, A^m A^m$) (Fig. 2.3), with non-brittle rachis (*br*), which arose as a result of a mutation in the brittle rachis (*Br*) gene of *Triticum monococcum* ssp. *aegilopoides*, are wild type. In the *Aegilops* genus, there are six diploid wild wheats, also called sytopsis: *Aegilops tauschii* ($2n = 2x = 14, DD$), *Aegilops bicornis* ($2n = 2x = 14, S^bS^b$), *Aegilops searsii* ($2n = 2x = 14, S^sS^s$), *Aegilops longissima* ($2n = 2x = 14, S'S'$),

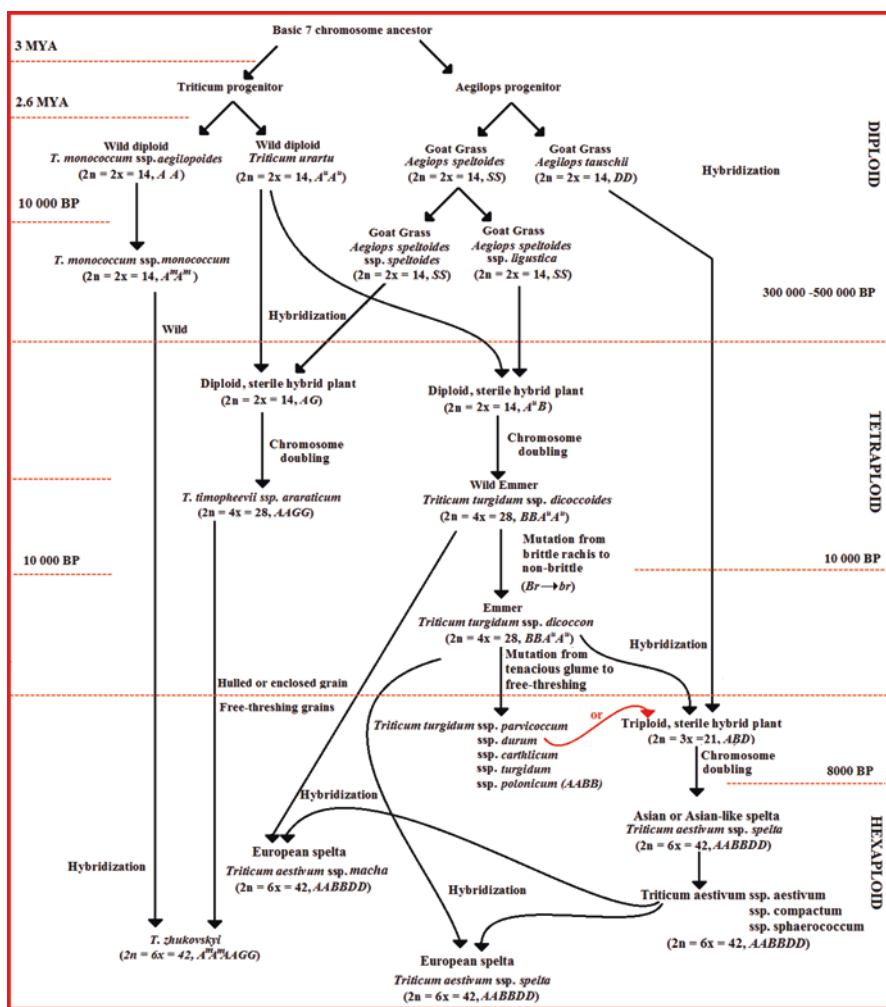


Fig. 2.2 Evolutionary lineage of wheat. (The figure is modified from Faris (2014) and Peng et al. (2011))



Fig. 2.3 *Triticum monococcum* spike morphology. (Photo taken by Ö. Özbek in 2017)



Fig. 2.4 Wild emmer wheat (*Triticum dicoccoides*) in the natural growing field at Golan Height in Israel. (Photo taken by Ö. Özbek in 2005)

Aegilops sharonensis ($2n = 2x = 14, S^hS^h$), and *Aegilops speltoides* ($2n = 2x = 14, BB/GG$), which diverged about 2.6 MYA (Feldman & Millet, 2001; Faris, 2014; Peng et al., 2011).

The first tetraploid wild wheats *Triticum araraticum* ($2n = 4x = 28, A^uA^uGG$) and *Triticum turgidum* ssp. *dicoccoides* ($2n = 4x = 28, BBA^uA^u$) (Figs. 2.4 and 2.5) existed about 300,000–500,000 years before the present (BP), formed by the



Fig. 2.5 Wild wheat (*Triticum dicoccoides*) in its natural field in Israel. (Photo taken by Ö. Özbek in 2005)

hybridization of *Triticum urartu* ($2n = 2x = 14$, A^uA^u) with *Ae. speltoides* ssp. *speltoides* ($2n = 2x = 14$, GG), and with *Aegilops speltoides* ssp. *ligustica* ($2n = 2x = 14$, SS) (Fig. 2.2), respectively. The resultant hybrids were most probably sterile owing to the incomplete meiosis of non-homologous chromosome pairs of different genomes (A and S/B genome pairs, A and G genome pairs). After a chromosome doubling process by autopolyploidization, new species of fertile wild emmer wheat (*Triticum dicoccoides*, $2n = 4x = 28$, BBA^uA^u) and *Triticum araraticum* ($2n = 4x = 28$, A^uA^uGG) were formed (Feldman & Millet, 2001; Faris, 2014; Peng et al., 2011).

A mutation event in the genes controlling tenacious glume in *T. dicoccum* led to the evolution of naked tetraploid wheat ($2n = 4x = 28$, $AABB$), ssp. *parvicoccum*, ssp. *durum*, and ssp. *carthlicum* around 7500–8000 BC.

A spontaneous hybridization occurred between emmer wheat (*Triticum turgidum* ssp. *dicoccon*, $2n = 4x = 28$, BBA^uA^u) and *Aegilops tauschii* ($2n = 2x = 14$, DD), when it arrived at the growing area of *Ae. tauschii* after having spread from the Fertile Crescent to different regions of the world. This hybridization event produced a new form of wheat, hexaploid wheat (*Triticum aestivum* ssp. *spelta*, $2n = 6x = 42$, $AABBDD$) (Fig. 2.6), considered to be Asian spelt wheat, about 8000 BP. *Triticum monococcum* ssp. *monococcum* contributed the A^m genome of *Triticum zhukovskyi* ($2n = 6x = 42$, $A^m A^m AAGG$), another hexaploid wheat form, the A and G genomes of which, contributed by the wild progenitors of *Triticum timopheevii* ($2n = 4x = 28$,



Fig. 2.6 Bread wheat field and its spike morphology. (Photos taken by Ö. Özbek in 2017)

AAGG), resulted from hybridization between *T. urartu* and *Ae. speltoides* ssp. *speltoides* (Faris, 2014; Feldman & Millet, 2001; Charme, 2011).

2.3 Domestication of Wheat

Archaeological researches at archaeological remains have produced fruitful results that shed light on the domestication of several crop plants and early forms of agriculture in the Fertile Crescent, and this has helped us to date the domestication time of these species. Thus, researchers have suggested different scenarios about when and where the domestication of these crop plants took place and how and when the first forms of agriculture started. Many of these scenarios share the idea that the domestication of eight crop plant species [wheats: diploid einkorn wheat (*Triticum monococcum*), tetraploid emmer wheat (*Triticum dicoccon*), and barley (*Hordeum vulgare*); two pulses: lentil (*Lens culinaris*) and pea (*Pisum sativum*); and flax (*Linum usitatissimum*), bitter vetch (*Vicia ervilia*), and chickpea (*Cicer arietinum*) (Lev-Yadun et al., 2000; Terence et al., 2008; Weiss & Zohary, 2011)] took place in the Fertile Crescent. However, agriculture developed independently in several regions of the world at around the same time. Other than the Fertile Crescent region, the regions are Mesoamerica, where maize was first grown, rice was first grown in the Yangtze region of Southeast Asia, and potato, peanut, and manioc were first grown in South America (Terence et al., 2008; Harlan, 1998; Feldman & Kislev, 2007).

2.4 Genetics of Domestication in Wheat

The domestication of wheat was accelerated by evolution. Domestication is the process of cultivation toward the plant's adaptation to a new anthropized environment following anatomical, morphological, and genetic changes, which also modified yield and yield components (Brown, 2010; Nesbitt, 2002; Peng et al., 2011). Cereals, particularly wheat, were among the first crop plants to have been domesticated in the Middle East and then spread out to the world. The genetic changes in the transition from wild to domesticated forms include three major genes—brittle rachis (*Br*), tenacious glume (*Tg*), and non-free-threshing character (*q*)—found in wild forms (Faris, 2014).

The domestication process caused a domestication syndrome, which can be defined as the genetic modifications in some genes in wild forms to a human-dependent state owing to human needs (Peleg et al., 2005). The factors driving the domestication syndrome are important not only from an evolutionary perspective, but also they have important economic and social conclusions. Therefore, much attention should be paid to the analysis of the traits related to the domestication syndrome.

2.4.1 Brittle Rachis (*Br*)

The evolutionary significance of brittle rachis (*Br*) is its adaptive value, because that is responsible for the free shattering of seeds, not dependent on humans for seed dispersal and self-sowing into the soil (Zohary & Hopf, 2000) in the wild form, whereas loss of seeds was seen before this mutation. In Triticeae, the spikelets can disarticulate or shatter at maturity owing to brittle rachis, and this enabled seed dispersion and self-planting (Nesbitt, 2002; Zohary & Hopf, 2000) independently in wild forms such as wild emmer wheat [*Triticum turgidum* ssp. *dicocoides* (Körn. Ex Asch. and Graebner) Thell., $2n = 4x = 28$, *BBAuAu*], but in cultivated emmer wheat (*Triticum turgidum* ssp. *dicoccon*, $2n = 4x = 28$, *BBAuAu*), the seeds stay intact, with non-brittle rachis—dependent on humans for seed dispersion, which is the major distinguishing feature between them—until the plants are harvested. The genes *brittle rachis* 2 (*Br-A2*) and *brittle rachis* 3 (*Br-A3*) controlling rachis fragility were characterized as dominant and were located on the short arms of chromosomes 3A and 3B, respectively, in wheat (Cao et al., 1997; Chen et al., 1998; Watanabe & Ikebata, 2000; Watanabe et al., 2002; Salamini et al., 2002; Watanabe, 2005; Li & Gill, 2006; Nalam et al., 2006; Gill et al., 2007; Peng et al., 2011).

A mutation in the *Br* locus led to a recessive non-brittle locus *br* in cultivated forms that occurred independently in both diploid and tetraploid wild wheats (Gill et al., 2007). The mutation existed in wild einkorn wheat *Triticum monococcum* ssp. *aegilopoides*, giving rise to domesticated *Triticum monococcum* ssp. *monococcum* (Fig. 2.3), while the same mutation event took place in tetraploid wild emmer wheat

(*Triticum turgidum* ssp. *dicoccoides*) and *Triticum timophevii* ssp. *araraticum* (AAGG); the *br* locus resulted in domesticated tetraploid emmer wheat (*Triticum turgidum* ssp. *dicoccon*) (Fig. 2.7) and *Triticum timophevii* ssp. *timophevii* (AAGG), respectively. In both cases, the species are very similar to each other in phenotype, but the latter one has a tough rachis (Faris, 2014). Today all wild wheat forms, both diploid and tetraploid, have a brittle rachis, while domesticated forms have a tough (non-brittle) rachis (Gill et al., 2007).

Disarticulation due to brittle rachis resulted in two basic types of disarticulation in wheats. When the whole spike is broken at the base and the whole spike is dispersed, this is called spike-type disarticulation, controlled by the *Br1* gene, as observed in *Ae. speltoides* var. *speltoides* (Chen et al., 1998). The second type of disarticulation is the spikelet type, which is subdivided into two other disarticulation types depending on the breakage point of the spikelets. In wedge-type (W-type) disarticulation, the spikelet is broken at a point between the upper side of the junction of the rachis and spikelet base attached to a rachis fragment, and it is controlled by *Br1^{3A}* (3AS), *Br1^{3B}* (3BS), and *Br1^{3D}* (3DS) genes (Li & Gill, 2006), as observed in species possessing A, B, G, S, and T genomes (Faris, 2014; Kimber & Feldman, 1987; Li & Gill, 2006), while non-allelic recessive genes or complementary recessive genes on 5A and 7A control W-type disarticulation in *T. monococcum* (Fig. 2.8) (Sharma & Waynes, 1980). Peng et al. (2003, 2004) reported a gene (*Br42A*) (2AL) related to rachis brittleness in *T. turgidum* ssp. *dicoccon* (Fig. 2.9).

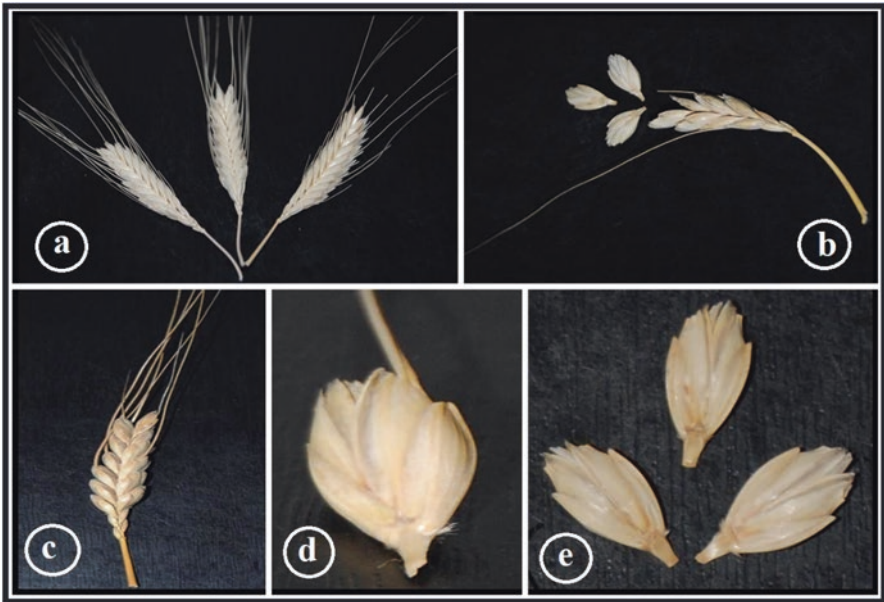


Fig. 2.7 *Triticum dicoccon* spike morphology (a, b, and c) and wedge-type spikelet (d and e). (Photo taken by Ö. Özbek in 2017)



Fig. 2.8 Wedge-type disarticulation of *T. monococcum* spikelet. (Photo taken by Ö. Özbek in 2017)



Fig. 2.9 Wedge-type disarticulation in *T. dicoccum* spikelet. (Photos taken by Ö. Özbek in 2017)

In barrel-type (B-type) disarticulation, the spikelet is detached from the rachis junction point, having a flat-like scar at its base, and it is controlled by the *Br2^{3D}* (3DL) gene, as observed in species containing the *D* genome, such as *Ae. tauschii* (Faris, 2014; Gill et al., 2007; Li & Gill, 2006).

2.4.2 Tenacious Glume (Tg)

There needs to be a mechanism or a structure to protect wheat seeds from harsh environmental conditions to ensure their survival until self-planting occurs during the period of natural seed dispersal. That structure is called a tenacious glume; it is tough and tightly encases the seed, and it is not threshed from the seed in wild wheats; it occurs in what is called hulled wheats, such as ssp. *aegilopoides* and ssp. *dicoccoides* and some domesticated forms, ssp. *monococcum* (Fig. 2.10) and ssp. *dicoccum* (Fig. 2.11). The characteristics of tenacious glume were defined for the first time by Kerber and Dyck (1969); the structure is controlled by an incompletely dominant gene, *Tg1^{2D}*, and it was mapped and validated on chromosome 2DS by Jantasuriyarat et al. (2004) in *T. aestivum* ssp. *aestivum*. Simonetti et al. (1999) reported another gene, *Tg2^{2B}*, mapped on chromosome 2BS in *T. turgidum* ssp. *parvicoccum*, a subspecies which is not known for certain—possibly it might have been extinct and it might have been *parvicoccum* or *durum* or may be another tetraploid subspecies (Faris, 2014). On the other hand, a domesticated einkorn wheat, *T.*



Fig. 2.10 *T. monococcum* spike and hulled grains. (Photos taken by Ö. Özbek in 2017)



Fig. 2.11 *Triticum dicoccum* spike and hulled grains. (Photo taken by Ö. Özbek in 2017)

sinskajae, has a free-threshing soft glume (*sog*) trait controlled by a single recessive gene located on $2A^m$ (Faris, 2014; Sood, 2009; Taenzler et al., 2002).

The genes $Tg1^{2D}$ and $Tg2^{2B}$ controlling tenacity possibly might be homeologous, or another possibility is that $Tg2^{2B}$ and *Sog* on $2A^m$ are homeologous genes (Faris, 2014). Free-threshing or naked grain has a shorter dormancy period, enhancing the possibility of germination and growth under favorable conditions. Domestication started 10,000 years ago and resulted in fully domesticated wheat, *Triticum turgidum*, and *Triticum aestivum*, through rachis and free-threshing naked seed. It is not clear whether early farmers through the collection of wheat plants, which had naked seeds that were then resown, by the farmers drove domestication.

2.4.3 Free Threshing (Q Factor)

Following the domestication process, polyploid wheat species existed, having non-brittle rachis and a free-threshing nature. These transitions gave rise to the development of the two most common modern wheats, bread wheat (*Triticum aestivum*, $2n = 6x = 42$ AABBDD) and durum wheat (*Triticum turgidum*, $2n = 4x = 28$ AABB). The free-threshing nature in the phenotype of wheat controlled by two genes was gained through changes from tenacious and tightly enveloped glume to softened and easily separable hulls (Faris, 2014). The latter change was driven by mutations that

occurred in the genes; *Tg* controls the tenacious glume character and is mapped on chromosome 2B of the *BB* genome, and *q* controls non-free threshing and is mapped on the long arm of chromosome 5A of the *AA* genome. After mutations, the alleles *tg* and *Q* control free threshing; they are partially recessive and partially dominant, respectively (Peng et al., 2011).

The genotype of a free-threshing tetraploid (*AABB*) genome should be *tgtg*-^{2B}*QQ*^{5A}. On the other hand, a second mutation could have occurred in the *DD* genome of bread wheat, contributed by *Ae. tauschii*. If this has not occurred, the *Tg* locus gained from *Ae. tauschii* prohibits free threshing in bread wheat (*AABBDD*), which has the *tgtg*^{2B}*tgtg*^{2D}*QQ*^{5A} genotype (Sang, 2009). *Tg* is epistatic to (Gill et al., 2007) or has a dominant effect over the *Q* allele under hulled or non-free-threshing conditions, while the plants with *br* and *tg* alleles, but lack of *Q*, also have a non-free-threshing character. In addition to this latter character, these plants have a semi-brittle rachis, a speltoid spike, and a tenacious glume and are taller; they flower earlier and display differences in yield compared with plants that have the *Q*^{5A} allele (Faris, 2014). These consequences infer the pleiotropic effects of *Q*^{5A} on other traits: squarehead spike, rachis fragility, glume toughness, glume shape, and plant vigor, which are important in wheat domestication and in agronomy (Faris, 2014; Muramatsu, 1963; Simons et al., 2006). According to previous studies, it was confirmed that homoeologous *q* loci appeared on chromosomes 5B and 5D (Simons et al., 2006; Zhang et al., 2011). The high conservation of *Q/q* gene sequences also described all ploidy levels in the *A*, *B*, *S*, and *D* genomes. It was thought that duplication of the *q* gene in a diploid ancestor genome of diploid wheats occurred before they separated, about 5.8 MYA.

In the evolution of diploid wheat, one copy of the gene was conserved in *A* genome progenitor, while the other copy was lost in *B*, *S*, and *D* genomes. The *q*^{5D} allele in *Ae. tauschii* and hexaploid wheat is still present and helps to suppress the speltoid syndrome, but to a lesser degree compared with *Q*^{5D}. During allopolyploidy formation, the *q*^{5B} allele changed to a pseudogene, but it is still involved in the regulation of domestication-related traits; finally, *q*^{5S} is present in *Ae. speltoides*, also still functional and with the whole sequence conserved. Consequently, the mutant *Q*^{5A} allele is the major effective allele in the domestication of wheat, with the contributions of *q*^{5B} and *q*^{5D} (Faris, 2014). Peleg et al. (2005) reported qualitative trait loci related to glume toughness on chromosomes 4A, 4B, and 7B, in addition to the major tough glume genes *Tg* and *sog* on chromosome 2 and the *Q* gene on chromosome 5AL in emmer wheat.

2.5 Domestication Geography of Hulled Wheats

The origin of domestication of plants and the origin of agriculture may not be the same. But agriculture helped the spread of domesticated wheats westward around the Mediterranean; across North Africa and southern Europe; northward across the Balkans to western Europe, the British Isles, Scandinavia, and Russia; and from the

Ethiopian Plateau to India (Harlan, 1966). Archaeological remains have contributed to our understanding of the domestication origins or geography of crop plants; here we concentrate on hulled wheats (einkorn and emmer wheat) particularly. Many investigations have been done on botanical remains in Anatolia, which is part of the domestication area considered as a cradle of agriculture. It is thought that this domestication process took place about 10,000 BP in the Fertile Crescent. The first domesticated wheat variety was the einkorn wheat, *Triticum monococcum* ssp. *monococcum*, which has a diploid AA genome derived from the wild einkorn wheat *Triticum boeoticum*; both were found together, considered as a mark of domestication, at an archaeological site that was characterized as being about 10,500 calibrated years before the present (cal BP) (Gopher et al., 2002; Zohary & Hopf, 2000). Although *Triticum monococcum* ssp. *monococcum* became abundant at about 9500 cal BP (Nesbitt & Samuel, 1995) and then spread to Cyprus, Greece, and the Balkans in 8000 cal BP (Van Zeist, 1981; Renfrew, 1979a, b, cited in Salamini et al., 2002), later spreading to the countries of the former Yugoslavia, Bulgaria, and Hungary, its cultivation started to decrease in the Bronze Age (Salamini et al., 2002), and today, it is regarded as a relic crop and cultivated in a very limited area in the world, including a few provinces in Turkey, Italy, and the former Yugoslavia, primarily as animal fodder. Recently in Turkey, popular interest in organic farming and its healthy end products has led to the reinstatement of the farming of domesticated einkorn wheat (Morgounov et al., 2016).

Earlier studies indicated that *Triticum monococcum* ssp. *boeoticum* and *Triticum monococcum* ssp. *urartu* displayed differences in both morphology (Dorofeev et al., 1979; Gandilian, 1972) and biochemical and molecular markers (Dvořák et al., 1988; Jaaska, 1974; Johnson, 1975). Valkoun et al. (1998) reported that two wild diploid wheats (*Triticum monococcum* ssp. *boeoticum* and *Triticum monococcum* ssp. *urartu*) differed in their geographical distribution, with ssp. *urartu* existing in the area extending from Transcaucasia through eastern Anatolia and the Fertile Crescent to Jordan and Hauran and the Jebel Al Arab region of southern Syria, including the Syrio-Palestinian region (Johnson, 1975), while ssp. *boeoticum* existed mainly in southern Turkey, where it was most probably domesticated (Heun et al., 1997) extending as well across Turkey from Transcaucasia to Greece (Johnson & Dhaliwal, 1976). The ssp. *urartu*, an A genome donor of tetraploid and hexaploid wheats, exhibits a distribution similar to that of *Triticum turgidum* ssp. *dicoccoides*. Heun et al. (1997) used amplified fragment length polymorphism (AFLP) fragment analysis to investigate the site of einkorn domestication. They concluded that Karacadağ *Triticum monococcum* ssp. *boeoticum* lines could have been the progenitor species of cultivated einkorn domesticated about 10,000 years ago in the Karacadağ mountain region. Thus, Karacadağ mountain is most probably the site of einkorn domestication, a finding that was also supported by findings from early settlements near Karacadağ mountain, covering Cafer Höyük (De Moulins, 2000), Çay Önü (Van Zeist & de Roller, 1991/1992), Nevalı Çori (Pasternak, 1998), and Abu Hureyra (De Moulins, 2000; Hillman, 2000; Salamini et al., 2002) (Fig. 2.11).

Then genetic diversity was investigated using 151 AFLP loci and 18 haplotype loci in *Triticum monococcum* (einkorn) germplasm by Kilian et al. (2007). The

natural genetic differentiation was determined among the wild einkorn wheats, and the wild einkorn (*Triticum monococcum* ssp. *boeoticum*) underwent genetic differentiation prior to domestication. The larger nucleotide diversity was observed within *Triticum monococcum* ssp. *monococcum* than within *Triticum monococcum* ssp. *boeoticum*, and *T. urartu* was genetically diverged than others. *Triticum monococcum* ssp. *aegilopoides* was not found in the Karacadağ mountain and Kartal–Karadağ region (Kilian et al., 2007). Einkorn wheat replaced with tetraploid and hexaploid wheats in the last 5000 years and neglected by modern breeders, cultivated until the early Bronze Age (Bar-Yosef, 1998). *Triticum turgidum* ssp. *dicoccoides* emerged by the hybridization of two wild diploid wheats, *Triticum urartu* ($2n = 2x = 14 AA$), considered as the paternal A genome donor, and *Aegilops speltoides* ($2n = 2x = 14 SS/BB$), considered as the maternal parent B genome donor, around Mount Hermon and the Jordan River in the Fertile Crescent about 360,000 years ago (Dvořák & Akhunov, 2005; Feldman & Kislev, 2007; Özkan et al., 2011). This inference was made owing to the great morphological, phenological, biochemical, and molecular variation observed in wild emmer populations from southeastern Turkey, northern Iraq, southwestern Iran, Israel, and Jordan (Nevo & Beiles, 1989; Ozbek et al., 2007a, b; Özkan et al., 2002).

The geographical distribution of wild emmer has been described in other research (Harlan & Zohary, 1966; Johnson, 1975; Özkan et al., 2005, 2011; Valkoun et al., 1998; Zohary & Hopf, 2000). Its major distribution area is placed in the Fertile Crescent, including the central southeastern part of Turkey, the eastern part of the mountainous area of Iraq, western Iran, and the Jordan Valley (Harlan & Zohary, 1966; Zohary, 1973), where the habitat was characterized by Nevo et al. (2002). The population structure and distribution in Israel was described as an archipelago in which populations were isolated or semi-isolated, showing an island model (Nevo & Beiles, 1989; Ozbek et al., 2007a). This characteristic might be associated with the soil type; where wild emmer wheat grows, the area is not covered continuously by the same type of soil. Wild wheats grow on basalt bedrock or decalcified alluvial soils, and much of the area is unsuitable for wild wheats and rye, owing to the presence of Eocene and Paleocene chalk limestone (Willcox, 2005). The distribution of wild emmer wheat is scarce throughout Iraq and Iran, and the density of another tetraploid wheat, *Triticum araraticum* Jakubz. ($2n = 4x = 28, AAGG$), displays dominance (Johnson, 1975) in those areas. In southeastern Turkey, a dense wild emmer wheat population grows on the slopes of the Karacadağ mountain (Şanlı Urfa and Diyarbakır provinces) and the Kartal–Karadağ mountain range region (Harlan & Zohary, 1966; Özkan et al., 2011). Wild emmer wheat is distributed mainly in primary habitats from Karacadağ mountains to Israel, ranging in altitude from 100 to 150 m below sea level up to 1600–1800 m above sea level (Aaronsohn, 1909; Nevo et al., 2002; Schweinfurth, 1908), and occasionally, it is distributed in secondary habitats, as in Turkey (Özkan et al., 2005, 2011).

Grains of wild emmer wheat were found for the first time at Ohalo II, a permanent site of Epipaleolithic settlement 19,000 BP (Fig. 2.2); this find showed that hunter-gatherer humans collected and used wild emmer wheat seeds (Feldman & Kislev, 2007). When the gene responsible for brittle rachis (*Br*), also called the domestication

gene, had undergone mutation in wild tetraploids, the cultivated tetraploid forms, *Triticum araraticum* ($2n = 4x = 28$, AAGG) and emmer wheat (*Triticum turgidum* ssp. *dicoccon*, $2n = 4x = 28$, BBA^uA^u) were formed. The only difference between wild emmer wheat and cultivated emmer wheat is non-brittle rachis (*br*).

Nevali Çori (NÇ) is a prehistoric settlement dated from the early pre-Pottery Neolithic B era to the early Bronze Age (Fig. 2.2) (Hauptmann, 1984, 1987, 1988, 1997). Pasternak (1998) reported some botanical remains, including 26,792 items determined as wheat, from NÇ. Although there were some difficulties about the identification of one-seeded and two-seeded grains as *T. boeoticum* or *T. dicocoides* in his findings, he revealed that 4 of 30 larger types of grains were obviously two seeded. These four types could have been identified as *T. dicoccon* and the others as *T. monococcon*, while 23 terminal spikelets of 500 complete spikelet forks were an indication of the existence of tetraploid wheat. Domesticated emmer wheat, *T. dicoccon* ($2n = 4x = 28$ BBAA), is a hulled wheat type, while *T. parvicoccon* ($2n = 4x = 28$ BBAA), the naked form, assumed to be an extinct tetraploid form (Faris, 2014), had small grain (Kislev, 1980); both have a BBAA genome formula, as is common for tetraploid wheats. Although domesticated emmer wheat was the most important cereal for cultivation in the Fertile Crescent, einkorn wheat was cultivated until the early Bronze Age (Bar-Yosef, 1998), and today, only traditional farmers are growing einkorn wheat on a limited scale in Ethiopia, Iran, Italy, Transcaucasia, Turkey, and the Balkans. Einkorn wheat was found at early Neolithic sites (such as Tell Aswad, ~10,800 BP; Van Zeist & Bakker-heeres, 1982; Özkan et al., 2005). In the Fertile Crescent, emmer wheat (*Triticum turgidum* ssp. *dicoccon* [Schrank] Thell.) was the first domesticated tetraploid wheat, and its subspecies were identified as follows: (1) ssp. *maroccanum* Flaksb. (Moroccan emmer); (2) ssp. *abyssinicum* Vav. (Ethiopian emmer); (3) ssp. *europaeum* Vav. = ssp. *dicoccon* (European emmer); and (4) ssp. *asiaticum* Vav. (Eastern emmer) (Gökgöl, 1955; Dorofeev et al., 1979; Szabó & Hammer, 1996; Teklu et al., 2007; cited in Özkan et al., 2011). Previous studies (Heun et al., 1997; Lev-Yadun et al., 2000) had described a small core area in southeastern Turkey where several cereal crops and some other crop plant progenitors were distributed, and the origin of domestication of cereal crops was reported to have existed in that area.

Özkan et al. (2002) investigated the core area to determine whether it was the place where additional founder crops of the Fertile Crescent also originated; they used 204 AFLP loci in 43 domesticated lines and 99 wild emmer (Turkey, 22; Israel, 37; Jordan, 8; Lebanon, 13; Syria, 18; and Iran, 1) populations of tetraploid wheats from their primary habitats. These researchers found that 15 of 19 Karacadağ lines were closely related to domesticated emmer wheat; as well, a line from a secondary habitat from İzmir was also closely related to cultivated emmer. Overall, the results indicated that there were two genetically distinct different wild emmer taxa: the central-eastern one, which includes Turkey and Iran, and the western one, which includes Israel, Jordan, Lebanon, and Syria. The results were consistent with previous studies (Abbo et al., 2002; Gopher et al., 2002; Heun et al., 1997; Lev-Yadun et al., 2000), suggesting that a small core area located in southeastern Turkey near the Tigris and Euphrates Rivers was the place of origin of tetraploid wheat

domestication. This place also covers the area for the domestication of the wild progenitors of other founder crops of pea, chickpea, lentils, and einkorn.

Özkan et al. (2005) investigated the domestication geography of tetraploid wheats using the same method (AFLP) as in their previous study (Özkan et al., 2002), but using a new set of wild emmer lines; 69 accessions from the collections of Mori et al. (2003), 7 lines from Dr. B. Gill, and 12 lines from Dr. A. Karagöz. *Triticum dicoccoides* was domesticated in the same core area previously described by Özkan et al. (2002), which was confirmed, and the domestication origin was considered as monophyletic. However, Salamini et al. (2004) suggested that the domestication origin of tetraploid wheats might have been possibly diphyletic, with two major phylogenetic tree topologies taking place by *T. durum* (free threshing) and *T. dicoccum* (hulled) lines. According to AFLP markers, two genetically very different taxa of *T. dicoccoides*—one with its primary habitat in Israel, Jordan, Lebanon, and Syria, named the western colony, and the other in Turkey, Iran, and Iraq, named the central-eastern colony (Sachs, 1953; Harlan & Zohary, 1966; Rao & Smith, 1968; cited in Özkan et al., 2005).

Cultivated emmer wheat, a domesticated form of hulled wheat, arose in the Fertile Crescent, and advances in knowledge and agricultural technology led to its spreading west, east, north, and south, giving rise to agriculture in the area. Examples of these sites are ca. 9000–7600 cal BP, Dhali Agridhi, Cyprus (Stewart, 1974); ca. 9000–8050 cal BP, Franchthi Cave (Hansen, 1992); ca. 8650–8200 cal BP, Sesklo, Greece (Hopf, 1962; Kroll, 1981); and ca. 8650–8400 cal BP, Knossos, Crete (Sarpaki, 2009). Cultivated emmer wheat continued to spread around 4500 BP, and it arrived in the Balkan countries. Emmer wheat findings in Egypt date to ca. 7500–6650 cal BP in Fayum and 1325 BC in Tutankhamun's tomb (Caton-Thompson & Gardner, 1934; Täckholm, 1976; Germer, 1989; Hepper, 1990; Wetterstrom, 1993; De Vartavan, 2010). Toward the east, the botanical remains found at Neolithic sites in Caucasia and Transcaucasia were considered to be from ca. 7950–7150 cal BP at Aratashen and Aknashen, Armenia, and emmer wheat was one of the main crop elements (Wasylikowa et al., 1991; Hovsepian & Willcox, 2008; cited in Zohary et al., 2012).

2.6 Conclusion

The wild wheat was first found in the Fertile Crescent region, the cradle of agriculture, and then evolution and domestication of wheat followed. The hybridization events by allopolyploidization between the species belong to genus *Triticum* and *Aegilops* taxa, evolved from a common ancestor about 3 MYA, giving rise to the wild tetraploid wheats and hexaploid spelt wheat species. The mutations existed in three major genes, which are brittle rachis (*Br*), tenacious glume (*Tg*), and non-free-threshing character (*q*) found in wild forms that led to the development of wheats with non-brittle rachis or with both non-brittle rachis and free-threshing modern wheats. Today, modern wheat varieties are distributed throughout the world, and it became a staple food and consumed as primary food and calorie source in the world.

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Chapter 3

Origin, Taxonomy, and Distribution of Ancient Wheats in Turkey



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3.1 Introduction

Wheat (*Triticum* spp.) holds a major component of global food security providing food and energy to one-third of the global population (FAO, 2016). Cultivated wheats and its close wild ancestors belong to the genus *Triticum* L., a member of the tribe Triticeae, which contains about 300 species (Baloch et al., 2017). Modern bread and durum wheats are genuine breeding hybrids of their wild grass species ancestor, still growing in the Fertile Crescent (Baloch et al., 2016). Lev-Yadun et al. (2000) proposed southeastern Turkey and northern Syria as the “core area” for the origins of agriculture within the Fertile Crescent. This was based on the proposition

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that wild einkorn and wild emmer from this area are genetically more closely related to the domesticated crop plants than elsewhere (Alsaleh et al., 2016; Yediay et al., 2010). Ancient wheat ancestors are einkorn (*Triticum monococcum*), emmer (*T. turgidum* ssp. *dicoccum*), and spelt (*T. aestivum* L. ssp. *spelta*), which are diploid, tetraploid, and hexaploid wheats, respectively, and are also known as “ancient wheats (also termed as farro).” Today’s modern bread and durum wheats have replaced these ancient wheats mainly due to the high yield and ease in threshing and milling. However, ancient wheats are not only a promising source of many important traits related with biotic and abiotic stresses for modern wheat improvement (Longin & Reif, 2014; Mujeeb-Kazi et al., 2013), but they are also attracting renewed interest for cultivation due to the global efforts in enhancing food diversity (Longin & Wurschum, 2016). Besides low grain yield, grains of ancient wheats are tightly enclosed by tough glumes and require special dehulling procedures to separate the chaff from the grain. Likewise, wild wheats have brittle ears that disarticulate at maturity into individual spikelets. Each spikelet, with the wedge-shaped rachis internode at its base, constitutes an arrow-like device that inserts the seed into the ground (Zohary & Hopf, 2000). By contrast, cultivated wheats have non-brittle ears that stay intact after maturation, depending on humans, for reaping, threshing, and sowing (Nevo et al., 2002).

Historically wild einkorn and emmer have driven the Neolithic revolution in agriculture and have been major food crops for thousands of years. Einkorn wheat was cultivated for centuries in the Middle East, Central Asia, Europe, and North Africa under different names. Today, it is a relic crop, grown in isolated, marginal lands of Turkey, Caucasus region, Europe, and Morocco (Zaharieva & Monneveux, 2014). Emmer was widely cultivated in antiquity, particularly in Egypt, and until recently started to cultivate in a large range of countries under different names (Zaharieva et al., 2010). Spelt was one of the major cereals of the Alamannians in southern Germany, Austria, and Switzerland between the twelfth and nineteenth century (Miedaner & Longin, 2016) and was also grown in tens of thousands of hectares in the United States.

The exploitation of ancient species is seen as a key factor to further drive genetic improvements in plant breeding (Baloch et al., 2015). Several strategies have been proposed, from the introgression of single genes (Saintenac et al., 2013) to breeding strategies aimed at improving quantitatively inherited traits such as grain yield (Longin & Reif, 2014). However, it has been well elaborated to extend these efforts to a more holistic and sustainable use of the available ancient species. Longin and Wurschum (2016) proposed that these ancient wheats can be reintroduced as crops by creating markets for specialty products, along with the discovery of traditional recipes and customs. This concept allows to not only increase the biological diversity in our agroecosystems but also enrich our food diversity. These ancient species will hardly help to feed the growing world population as they are often low yielding and not adapted to modern agricultural practices.

We have discussed 13 different species of ancient wheat in this chapter, out of which nine are cultivated and three are wild (Table 3.1). It is now consensus that family Poaceae (grasses) originated 50–70 million years ago (Mya) and the

Table 3.1 Botanical names (according to Dorofeev & Migushova, 1979) and genetic features of ancient wheats

Common name	Ploidy	Scientific name	Wild/ cultivated	Genome	
Einkorn	Diploid	<i>T. urartu</i> Thum. ex Gandil.	Wild	A ^u	Armenia, East of Turkey
		<i>T. boeoticum</i> Boiss.	Wild	A ^b	SE Europe, Asia Transcaucasia
		<i>T. monococcum</i> L.	Cultivated	A ^m	Turkey, Italy, Germany (locally)
Emmer	Tetraploid	<i>T. araraticum</i> Jakubz.	Wild	AG	Southeast and East Turkey, North Iran, North Iraq, Transcaucasia
		<i>T. dicoccoides</i> (Körn. ex Aschers. et Gräbn.) Schweinf.	Wild	A ^a B	Southeast and East Turkey, Syria, Israel, Palestine, North Iraq and Iran
		<i>T. dicoccum</i> (Schränk.) Schübl.	Cultivated	A ^a B	Northern Turkey, the Balkan Peninsula, Italy, Spain, Switzerland, Australia, Czech Republic
		<i>T. ispahanicum</i> Heslot	Cultivated	AB	İsfahan-Iran
		<i>T. palaeocolchicum</i> Menabde	Cultivated	AB	Georgia
		<i>T. timopheevii</i> (Zhuk.) Zhuk.	Cultivated	A ^a B	Georgia
Spelt	Hexaploid	<i>T. spelta</i> L.	Cultivated	A ^a BD	Europe, Iran
		<i>T. vavilovii</i> (Thum.) Jakubz.	Cultivated	A ^a BD	Armenia, Georgia
		<i>T. macha</i> Dekapr. et Menabde	Cultivated	A ^a BD	Georgia
		<i>T. zhukovskyi</i> Menabde et Ericzjan	Cultivated	A ^b A ^b G	Georgia

subfamily Pooideae is 25 Mya. Today, einkorn is a relic crop, although it is still sporadically grown in western Turkey, Balkan, Germany, Switzerland, Spain, and Caucasia (Nesbitt & Samuel, 1996). Prominent in Neolithic agriculture, its importance declined gradually since the Bronze Age, competitively replaced by free-threshing wheat. Einkorn is a small plant with a relatively small yield, yet it survives on poor soils where other wheats fail (Zohary & Hopf, 2000). While nutritious, einkorn produces bread with poor rising qualities. It is primarily used as porridge and also as animal feed. Wild einkorn (*T. boeoticum* included as a subspecies of *T. monococcum*, AA, Ab, Ab) is fully fertile with cultivated *T. monococcum*, and they are morphologically similar except for the brittle ears of wild einkorn (Zohary & Hopf, 2000). *Triticum boeoticum* is widespread in western Asia and southern

Balkans (Harlan & Zohary, 1966). It ranges primarily in the northern, cooler, and rainier regions of the Near East Fertile Crescent (map 1 in Zohary & Hopf, 2000), in oak and steppic widespread ecologies, and secondarily in edges of cultivations and roadsides. It prefers cooler climates and does not penetrate drier and warmer Israel but prevails in Turkey in the Karacadağ Mountains where cultivated einkorn may have evolved. Flaksberger (1935) and Vavilov (1935) divided the species in three and six eco-geographical groups, respectively. The three groups, considered by Flaksberger (1935), were *heothinum* Flaksb. (Turkey, Transcaucasia and Crimea), *alemanum* Flaksb. (Germany, North Switzerland), and *ibericum* Flaksb. (Spain, southern France, and Morocco). Knupffer (2009) reported a number of 5367 accessions of einkorn conserved in 54 gene banks. Jing et al. (2009) analyzed the diversity among 16 einkorn accessions using diversity array technology (DArT) markers. An accession (PI 355520) of Iranian origin was found to be distantly related to other accessions including most of the accessions of European origin. Interestingly, this accession was previously found to produce fertile F1 hybrids with hexaploid wheat (Cox et al., 1991) and to be a valuable source of resistance to leaf rust (Jacobs et al., 1996; Heun et al., 1997). Ancient emmer wheat, *T. turgidum* L. ssp. *dicoccum* (Schrank.) Thell. (traditionally designated as *T. dicoccum* Schuebl.), in which the threshing products are individual spikelets, represents the primitive situation in cultivated *T. turgidum* wheat.

As elaborated above, the einkorn and emmer wheats consist of both wild and domesticated species, whereas spelt wheats contain only domesticated species.

3.2 Taxonomy of Wheat and Distribution of Ancient Wheats

3.2.1 Taxonomy of the Wheat

The genera *Triticum* and *Aegilops*, called the wheat group, belong to the Triticeae tribe of the Poaceae family. The genus *Triticum* is divided into three groups according to the number of chromosomes: diploid ($2n = 14$), tetraploid ($2n = 28$), and hexaploid ($2n = 42$) (Feldman et al., 2001). Today, it is necessary to refer to tetraploid and hexaploid wheat species together with the number and set of chromosomes they contain. Tetraploid species consist of $2n = 28$, AABB genomes, and hexaploid species comprise $2n = 42$, AABBDD genomes, and each chromosome set is observed in a different wild and primitive wheat species (Nesbitt et al., 2001).

The classification of ancient wheats is shown in Table 3.1, and the taxonomic classification and origin of modern-day bread and durum wheats and role of various ancient wheats in their origin are briefly illustrated in Fig. 3.1. The characteristics of the diploid, tetraploid, and hexaploid wheat species are briefly explained below.

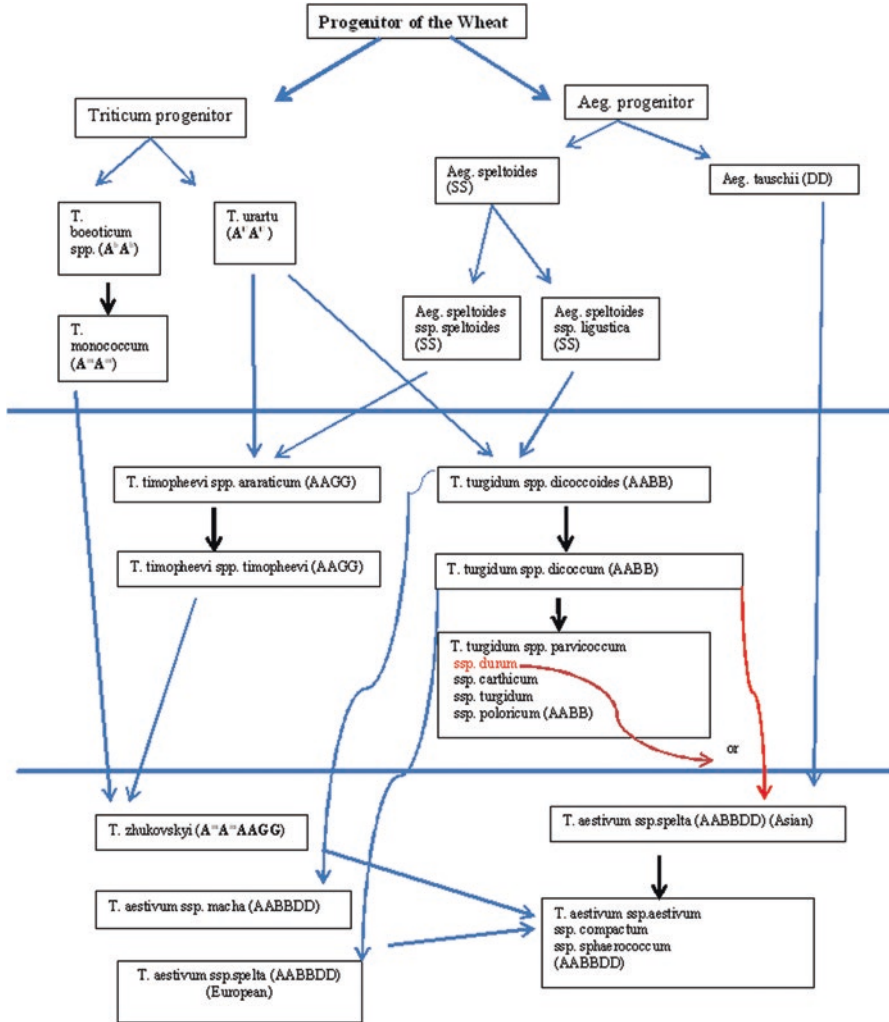


Fig. 3.1 Taxonomic classification and origin of tetraploid and hexaploid wheat

3.2.2 Diploid Wheat

Triticum urartu, *T. boeoticum* (wild einkorn wheat), *T. speltoides*, and *T. tauschii* are included in diploid wild ancient wheat, while *T. monococcum* is a cultivated einkorn in diploid ancient wheat. *Triticum boeoticum* has spike with brittle rachis, while *T. monococcum* has a non-brittle rachis in spike, which are called wild and domesticated einkorn, respectively. *Triticum boeoticum* (A^uA^u) and *T. urartu* (A^uA^u) are included in the wild wheat group and have a chromosome structure of 2n = 14, AA, while another diploid wheat is *T. monococcum* (A^mA^m), which contains the 2n = 14,

AA chromosome set, but is a domesticated form. In the literature, *T. monococcum* is considered to be the domesticated descendent of *T. boeoticum*. *Triticum urartu* ($2n = 14$, AA) is a non-cultivated species but is regarded as the progenitor of the A genome for both tetraploid and hexaploid wheat species (Dvorak et al., 1993). Other wild diploid wheat species include *Aegilops speltoides* ($2n = 14$; BB), which is the progenitor of the B genome of hexaploid and tetraploid wheat, and *A. tauschii* ($2n = 14$; DD), which is the progenitor of the D genome of hexaploid wheat species (Rodriquez et al., 2000).

3.2.3 Tetraploid Wheat

Wild tetraploid group *T. dicoccoides* and *T. araraticum* are accepted as wild emmer ancient wheat group and their domesticated form is a *T. dicoccum*, which was named tetraploid cultivated ancient wheat. *Triticum isphanicum*, *T. palaecolchicum*, and *T. timopheevii* are the other ancient tetraploid and domesticated form of the wheat. *Triticum dicoccoides* having a *breakable spike* feature and *T. dicoccum* with non-breakable spike are called wild emmer and domesticated emmer wheat, respectively. *Triticum dicoccoides* is the first form of tetraploid ancient wild wheat species ($2n = 28$, AABB), which is considered to be the predecessor of the domesticated species *T. dicoccum* (cultivated emmer wheat). Many researchers have suggested that the cultivated species *T. durum* derived from *T. dicoccum* (Kilian et al., 2007; Özkan et al., 2011; Zhang et al., 2002).

3.2.4 Hexaploid Wheat

The hexaploid ancient and also domesticated form species *T. spelta*, *T. vavilovii*, *T. macha*, and *T. zhukovskyi* are called as a spelt group, but in this group, *T. spelta* is the most common grown species. Hexaploid species ($2n = 42$, A^aA^bBBDD) have been reported to emerge from the hybridization between the *T. dicoccoides* ($2n = 28$, AABB) and *Aegilops tauschii* ($2n = 14$, DD) species and the spontaneous duplication of their chromosomes. *Triticum spelta* ($2n = 42$, AABBDD), the first domesticated hexaploid wheat, is described as ancient wheat and also ancestor of bread wheat. No wild hexaploid wheat species has been found in nature (Doebley et al., 2006).

3.3 Origin and Domestication of Ancient Wheats

In the abovementioned wild and domesticated ancient wheat species, *T. monococcum*, *T. dicoccum*, and *T. spelta* are the most important because of their potential traits for organic agricultural production and being resistant to marginal climatic and soil conditions (Castagna et al., 1995). During domestication of the wheat and

subsequent crop improvement under domestication by natural and unnatural selection, several morphological and physiological traits of wild progenitors were changed to meet human needs (Feldman & Kislev, 2007). The first and important domestication character in wheats was non-brittle rachis, which is associated with the loss of kernel dispersal mechanisms. It is strongly believed that transition was occurred from shattering wild einkorn ancient wheats (*T. boeoticum*) and wild emmer wheat (*T. dicoccoides*) to non-shattering and hard threshing diploid einkorn wheat (*T. monococcum*) and *T. dicoccum* (tetraploid emmer wheat), respectively, during domestication (Sood et al., 2009). Subsequently, several traits such as glumes toughness, grain number per spike, size, shape and weight of spike, seed dormancy, disease and pest resistance, and adaptation and productivity in different climatic and soil conditions were appeared appealing to farmers (Abbo et al., 2014; Peng et al., 2003).

Emmer wheat (*T. dicoccum*) and einkorn wheat (*T. monococcum*) were the first cultivated wheat species by humans. Domestication from the wild form of these species dates back to 10.000 years ago, and it is believed that they were originated and domesticated in Tigris–Euphrates areas of the Fertile Crescent. Also, it is believed that gathering of wild emmer wheat started 19.000 years ago, and this stage is regarded as the first phase of wheat cultivation. In the second stage, grains of domesticated forms of einkorn and emmer wheat were grown by humans. The farmers tried to select wheats that has non-brittle spikes following efficiently harvest grain without the spike shattering. The last domestication stage was the selection of free-threshing traits. This process led to replacement of the ancient wheats with free-threshing bread and durum wheat even if this process was so slow. Derivation of *T. monococcum* from *T. boeoticum* and *T. dicoccum* from *T. dicoccoides* and the hybridization between *T. urartu* (donor of the A genome) and *T. speltoides* (the progenitor of the B genome) resulted in the formation of a tetraploid species *T. dicoccoides* followed by *T. dicoccum*, which was then hybridized with a diploid species *Aegilops tauschii* to form hexaploid wheat species that indicated the existence of a close relationship between modern, domesticated, and wild wheat species and the inclusion of wild species in the evolution of wheat and underlined the importance of discussing the natural growing areas of these species. Arceobotanic, cytogenetic, and molecular studies based on DNA fingerprinting have shown the evidences suggesting that domestication of einkorn and emmer wheat took place in mountains of Karacadağ located in the southeastern region of Turkey (Mori et al., 1995; Özkan et al., 2011). Einkorn was the first widely grown cereal with its wide distribution around Transcaucasia, Middle East, southwestern Europe, the Balkans, and Mediterranean areas.

3.4 Distribution of Ancient Wheat Species

Triticum boeoticum, *T. urartu*, and *T. speltoides* are diploid wild ancient wheat. The northern and eastern regions of the Fertile Crescent are the major distribution areas of the *T. speltoides*. *Triticum urartu* is distributed in Armenia and eastern Turkey.

Near East, Central Anatolian region of Turkey, and northern and eastern parts of the Fertile Crescent are the primary habitats of *T. boeoticum* (Harlan & Zohary, 1966). Although the difference between *T. boeoticum* and *T. urartu* had been reported by several researchers (Takumi et al., 1993; Ciaffi et al., 2000; Sasanuma et al., 2002), many taxonomists suggest these two taxa as a single biological species (Sharma & Waines, 1981).

Hybridization between *T. urartu* ($2n = 14$, AA) and *T. speltoides* ($2n = 14$, BB) and chromosome number doubling resulted in tetraploid wild ancient wheat *T. dicoccoides* ($2n = 28$, AABB), and it is also called as a wild emmer wheat. Southeastern and eastern Turkey, Syria, Israel, Palestine, northern Iraq, and Iran are the major distribution areas of the *T. dicoccoides*. The domesticated form of ancient wheat specie *T. dicoccum* was derived from *T. dicoccoides* (domesticated emmer wheat). *Triticum dicoccum* is cultivated in limited areas of northern Turkey, the Balkan Peninsula, Italy, Spain, Switzerland, Australia, and Czech Republic.

The other ancient wheat species *T. monococcum* (domesticated form of einkorn wheat) derived from wild diploid wheat *T. boeoticum*, while the hexaploid ancient wheat species *T. spelta* derived from hybridization between *T. dicoccoides* and *Aegilops tauschii* (Colledge & Conolly, 2010). *Triticum monococcum* today is locally grown in some parts of northern (Kastamonu, Samsun, Sinop, Bolu, Tokat, and Bayburt provinces) and eastern parts of Turkey (Kars province) and in limited areas of Italy and Germany, and *T. spelta* in Europe and Iran (Zohary & Hopf, 2000; Zaharieva & Monneveux, 2014).

Triticum vavilovii is a hexaploid domesticated ancient wheat that was discovered first time near the Van Lake of Turkey (Zhukovsky, 1933). It has elongated rachillae and is morphologically similar to *T. aestivum*. Hexaploid ancient wheat *T. zhukovskii* carries G genome and is closely related to *T. timopheevii*, and it is grown in western Georgia (Jakubziner, 1958; Johnson, 1968).

Some other tetraploid wild and domesticated ancient wheat species are *T. araraticum* and *T. timopheevii*. *Triticum araraticum* is a tetraploid wild type of ancient wheat, morphologically close to *T. dicoccoides* and naturally grown in Tunceli, Şırnak, and Hakkari provinces of southeastern and eastern of Turkey (Aktaş, 2007; Özkan et al., 2011). Cytogenetic study indicated that *T. timopheevii* is more likely to have evolved from *T. araraticum*. The lack of natural habitat of the *T. araraticum* and limited distribution of *T. timopheevii* may explain why *T. timopheevii* is secondarily domesticated across northern and eastern regions of the Fertile Crescent. Although archeological records for *T. timopheevii* suggested that it domesticated in the Bronze Age onwards in Georgia, some researchers doubt that it can be distinguished from *T. dicoccum*.

Two other domesticated tetraploid types of ancient wheat have been identified, *T. isphanicum* and *T. palaeocolchicum*, and these are closely related to *T. dicoccum*. *Triticum isphanicum* is long glumed tetraploid ancient wheat, and it is similar to *T. polonicum* because of its free-threshing trait and is grown in Isfahan province of Iran. *Triticum palaeocolchicum*, which has compacted ear and zigzag rachis, is grown in limited areas of Georgia, and most of the taxonomists suggest that it is the

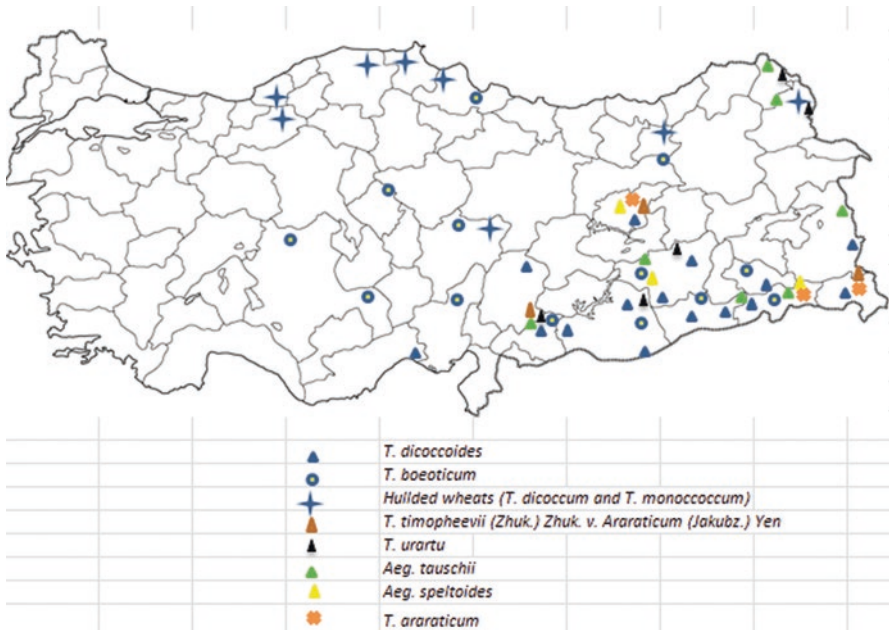


Fig. 3.2 Distribution of wild and domesticated wheat in Turkey

subspecies of *T. dicoccum*. Some brief detail about the distribution of wild and domesticated ancient wheats is briefly illustrated in Fig. 3.2.

The abovementioned origins of the diploid, tetraploid, and hexaploid groups indicated the existence of a close relationship between modern, domesticated, and wild wheat species and the inclusion of wild species in the evolution of modern-day tetraploid and hexaploid wheats and underlined the importance of discussing the natural growing areas of these species.

3.5 Distribution of Wild Ancient Wheats in Turkey

It has been reported by many researchers that the first cultivation of wheat took place in the Fertile Crescent primarily covering the southeastern Anatolia region of Turkey and extending to Iraq, Iran, and Syria and from there to other parts of the world (Dvorak & Luo, 2001; Blatter et al., 2002; Heun et al., 1997). Many researchers who conducted research on the evolution of wheat in the southeastern Anatolia region of Turkey found that wheat was first domesticated in Karacadağ mountain (Heun et al., 1997; Purugganan & Fuller, 2009; Feldman et al., 2001). We believe that Karacadağ area requires more examination and should have more considerations for domestication and distribution of wheat. Karacadağ is not simply a volcanic mountain; it is an 8000 km² area with a dimension of 120 km covered by basalt

stones located in the triangle of the Diyarbakır, Sanlıurfa, and Mardin provinces in the southeastern Anatolia region of Turkey. Entering the Karacadağ area, the altitude from the sea starts at 550 m, which, after a short distance, increases to 1500–2000 m (Aktaş, 2007; Özkan et al., 2011). In the remainder of the region, the altitude declines and increases. These short-distance changes in altitude result in an increase in the genetic diversity in terms of wild wheat species. We believe that Karacadağ is a natural laboratory of wild wheats. Karacadağ is the natural growing area of 35 wild wheat species and can be considered to have the highest global genetic diversity particularly for the *T. dicoccoides*, *T. araraticum*, *T. boeoticum*, and *T. speltoides* species (Aktaş, 2007; Kilian et al., 2009).

Other areas of wheat evolution and cultivation in the southeastern region of Turkey such as Cayonu and Nevala Cori are part of the Karacadağ area, which are also topographically similar to this area and are intensely covered by dark-colored basalt-volcanic stones. In this sense, southeastern Turkey and the Karacadağ area have played a significant role in wheat spreading to other parts of the world (Özkan et al., 2011). In Karacadağ region, the basalt stones that naturally protect wild wheat are removed to prepare area for agricultural use, extensive farming is undertaken, and herbicides are used (Ertekin, 2002; Kaya, 2006). This situation restricts the habitat of wild wheat and threatens genetic diversity. These areas need to be taken under in situ protection.

In Turkey, in addition to the Karacadağ area, *T. dicoccoides*, *T. boeoticum*, *T. araraticum*, *T. speltoides*, and *T. urartu* species are naturally grown in Siirt, Sirnak, and Hakkari provinces of southeastern Turkey located in the north of Iraq and in Mardin and Gaziantep provinces of Turkey located in the north of Syria, in Karadag, which is the extension of the Amanos mountain range and in Tunceli in the east of Turkey. *Triticum urartu* can be found in Ardahan and Kars provinces of the eastern region of Turkey (Aktaş 2007). However, there is limited data on the distribution of wild wheat species in these areas. The central Anatolia region of Turkey also has a rich habitat in terms of *T. boeoticum* and *Aegilops* species.

Comprehensive studies on the comparison of genetic diversity between wild wheat species that grow in Turkey with those in Iraq, Iran, Syria, and Israel have the potential to further elucidate the evolution of wheat and its distribution to other parts of the world. Figure 3.2 presents the distribution of diploid and tetraploid wild ancient wheat species in Turkey.

3.6 Domesticated Ancient Wheats in Turkey

The major ancient wheat species consist of *T. dicoccum* (domesticated emmer wheat) and *T. monococcum* (domesticated einkorn wheat) derived from the wild wheat species *T. dicoccoides* and *T. boeoticum*, respectively. Since ancient wheat species are tightly attached to their glumes, their threshing process is more difficult than that of bread wheat and durum wheat (*T. durum*) but easier than that of wild wheat species. Archaeological excavations have proven that ancient wheats, einkorn

(*T. monococcum* spp.) and emmer (*T. dicoccum*) wheat species were first domesticated 10,000 years ago in Cayonu and Gobeklitepe, which are part of Karacadağ mountain (Zohary & Hopf, 2000; Colledge & Conolly, 2010). Furthermore, *T. dicoccum*, *T. boeoticum*, *T. monococcum*, and barley samples were found in archaeological excavations performed in Catalhoyuk (Konya), Asıklı Hoyuk, Hacılar, and Erbaba areas located in the central Anatolia region of Turkey, and these areas have been shown to be significant in terms of the domestication and distribution of wheat (Bilgiç et al., 2016). The distribution of domesticated ancient wheat in Turkey is reflected in Fig. 3.2.

Since ancient wheats are resistant to poor soil structure and extremely hot or cold weather conditions, they can be cultivated in marginal areas of Turkey. Today, ancient wheats are grown in northern Turkey, the Balkan Peninsula, Italy, Spain, Switzerland, Australia, Czech Republic, and the restricted areas of Germany (Brandolini et al., 2011; Arzani & Ashraf, 2017). Ancient wheats appeal to consumers because of their high nutritional value, and they are also of interest to organic farming enterprises since they offer a sustainable production capability with its resistance to extreme climatic conditions.

In Turkey, ancient wheats are intensively cultivated by local small farmers in the provinces of Kastamonu, Samsun, Kars, Sinop, Bolu, Bayburt, Gumushane, and Kayseri (Aslan et al., 2016). In these areas, the einkorn group wheat species are known as “Siyez” and the emmer group is called “Gracer” and “Gernik.” Although no detailed study has been undertaken, based on the data from 2015, it is estimated that the total sowing area of ancient wheats in Turkey is approximately 14,000 ha. Ancient wheats are usually cultivated in rural areas with limited irrigation facilities, a cold climate, and small fragmented plots of agricultural land. Despite being the first domestication area of ancient wheats, the ancient wheats are not cultivated in the southeastern Anatolian region of Turkey. However, it is still possible to find small amounts of these species mixed with landraces cultivated in the region. The fact that despite being domesticated in the southeastern and central Anatolia regions, ancient wheats are only grown to a limited extent in these areas, which is an interesting issue that is open for discussion and investigation. Figure 3.2 presents the planting area of domesticated ancient wheats in Turkey by province.

3.7 Genetic Factors for Non-domestication in Ancient Wheats

The transformation of ancient wheat to non-brittleness and nakedness in modern wheat is largely underpinned by the “squarehead” *Q* locus located on chromosome 5A (Luo et al., 2000). Some other genes like controlling brittle rachis (*Br1* and *Br2*) and tough glume (*Tg* and *Sog*) are also responsible for spike-related traits and were the major forces driving domestication. A series of mutations have been described in literature from these genes of ancient varieties in all three ploidy members (Feldman

et al., 1995). Gill et al. (2007) cloned the *Q* gene, unraveling the structural and functional nature of the free-threshing trait and other early domestication events.

It was suggested in a series of experiments that free-threshing condition in modern bread wheat was due to two events: the appearance of the free-threshing gene, *q*, on 5A chromosome and the mutation from *Tg* to *tg* in the gene responsible for the tenacious glumes trait on chromosome 2D (reviewed in Faris et al., 2005). All the wild diploid and tetraploid progenitor species have the allele combination *BrBr/TgTg/qq*; however, hexaploid wheat species have different allele combinations. The transformation of brittle spike into tough spike is due to independent mutations, which led to the domestication of *Triticum monococcum* ssp. *monococcum*, *T. timopheevii* ssp. *timopheevii*, and *T. turgidum* ssp. *dicoccum*. Because the hexaploid wheat species originated under domestication, they shared these mutations from their respective progenitor species. Although mutations at the shattering and tough glume loci led to the domestication of a wheat that produced a decent harvest, it still had a wild-type speltoid spike and plant habit, and the seed was hulled. The master switch was the mutation at *q* to *Q*, which produced the modern robust plant habit with squareheaded and free-threshing spike. For this reason, *Q* has been referred to as a super-gene, and a 100-year-long history of research on this gene was reviewed by Faris et al. (2005). The mutation from *q* to *Q* occurred only once and, most likely, in a plant similar to the tetraploid wheat *T. turgidum* ssp. *dicoccum*, which has a tough spike and a soft glume phenotype with a speltoid spike (*br1br1tgqgq*).

The molecular genetic basis of the *Q* gene revealed that it encodes an AP2-like transcription factor (Simons et al., 2006). Allelic variations at 5A *Q* gene have been well elaborated by research; however, less is known about the *q* alleles on other homeologous chromosomes 5B (*5Bq*) and 5D (*5Dq*). Zhang et al. (2011) thoroughly investigated the organization, evolution, and function of the *Q/q* homeoalleles in bread wheat and its progenitors. It was observed that *Q/q* gene sequences are highly conserved within and among the A, B, and D genomes of bread wheat, the A and B genomes of tetraploid wheat, and the A, S, and D genomes of the diploid progenitors. However, the intergenic regions of the *Q/q* locus were highly divergent among homeologous genomes. Duplication of the *q* gene 5.8 Mya was likely followed by selective loss of one of the copies from the A genome progenitor and the other copy from the B, D, and S genomes. A recent V329-to-I mutation in the A lineage is correlated with the *Q* phenotype. The 5B*q* homeoalleles became a pseudogene after allotetraploidization. The transcript analysis showed that the alleles on homeologous chromosomes were coregulated in a complex manner. But combined phenotypic and expression analysis indicated that *Q* gene on 5A chromosome played a distinct role in conferring domestication-related traits, and *5Dq* contributes directly and *5Bq* indirectly to suppression of the speltoid phenotype. The evolution of the *Q/q* loci in polyploid wheat resulted in the hyperfunctionalization of 5A*Q*, pseudogenization of 5B*q*, and subfunctionalization of 5D*q*, all contributing to the domestication traits.

3.8 Recent Genetic Studies in Ancient Wheats

In the post-genomic era, rapid advances in biotechnological techniques should be better targeted to mine genetic diversity as part of pre-breeding and be more closely integrated with conventional breeding programs in order to achieve better and faster breeding outcomes. The effective genotyping tools are supposed to cover complete genome and should be high throughput and cost-effective. In the case of other Triticeae species, Ren et al. (2013a) analyzed genetic diversity in wild emmer wheat from Israel and Turkey and found genetic diversity to be correlated with ecological factors. Similarly, Ren et al. (2013b) evaluated genetic diversity in 150 worldwide collections of durum wheats and concluded that South America, North America, and Europe possessed the richest genetic diversity in durum wheat. Spelt wheat (*T. spelta* L.) now attracting the world's attention is being re-established in several parts of the world (Longin & Wurschum, 2016), which was also used for genome-wide association studies (GWAS) for agronomic and disease resistance traits using 15K SNP array (Würschum et al., 2017). Rapp et al. (2017) evaluated the agronomic and quality traits in 30 spelt varieties over several locations and concluded that sedimentation values along with aromatic bread flavor could be manipulated and improved in spelt wheat, and interdisciplinary research is needed to develop faster methods for evaluation of these traits. Similarly, Longin and Wurschum (2014) evaluated 150 old and new spelt varieties, and a higher genetic variability was estimated for agronomic, quality, and disease resistance traits in both spelt groups. They recommended pedigree selection with early generation screening on plant height and lodging for spelt wheat improvement. Conclusively, reintroduction of ancient crops like ancient wheats (especially spelt wheat) could be valuable as a high-value crop for farmers, millers, and bakers and would also fulfil the need of diversification in our food crops. A more holistic approach is needed to successfully implement this, which could be possible by interdisciplinary cooperation between breeding, agronomy, product quality, human nutrition, and socio-economics.

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Chapter 4

Genetic Diversity in Ancient Wheats



Özlem Özbek

4.1 Introduction

The term biodiversity or biological diversity started to be used in the 1980s. Biological diversity was recognized as an important critical issue for evolution and maintaining life-sustaining systems of biosphere at Convention on Biological Diversity (CBD) conference on “*Environment and Development*” held in Rio de Janeiro in June 1992 by the United Nations. In this convention, the term “*biological diversity*” was defined as “*the variability among living organisms from all sources including, inter alia, terrestrial, marine and other aquatic ecosystems and the ecological complexes of which they are part: this includes diversity within species, between species and of ecosystems*” (Secretariat of the Convention on Biological Diversity, 2005).

Biodiversity is the bases of the life on earth. Human activities such as inappropriate usage of natural resources, the change in habitats and climate, and distribution of pathogenic, exotic, and domestic plants and animals caused the dramatic reduction in biodiversity, which is observed in ecosystems worldwide in terms of taxonomic, phylogenetic, genetic, and functional diversity. As a result, it turns out a vitally important ecological problem in the world now (Naeem et al., 2012).

Biodiversity displays organization from the simplest level to more complex levels such as genes, individuals, populations, species, communities, ecosystems, and biomes in ecological systems (Norse et al., 1986; Barker et al., 2010). Biological diversity is divided into three levels: ecological (community) diversity, species (species numbers) diversity, and genetic (within or between species) diversity (Harper & Hawkworth, 1994).

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Ecological Diversity

Ecological diversity is the largest scale of the biodiversity and the ecosystems, in which it contains both species diversity and genetic diversity. Ecological diversity refers to the variation in both terrestrial and aquatic ecosystems. The variations in desert, forest, grassland, wetland, and ocean ecosystems could be examples for ecological diversity at a global scale (Cunningham, 2017). Ecosystems are open systems and conducting energy between the biological communities and their physical environment (Plischoff and Luebert in CONAMA 2008).

Species (Species Numbers) Diversity

Species diversity is the number of species in a community or in a sample, and usually, it can be used interchangeably for richness (Whittaker et al., 2001; Vellend & Geber, 2005). Species diversity may be the same in two different communities, but the evenness of abundance across species may be different. Therefore, species diversity represents both richness and equitability of a sample (Whittaker et al., 2001).

Genetic (Within or Between Species) Diversity

Genetic diversity is the differences (variations) between either in coding or non-coding DNA sequences (or chromosomes) of distinct individuals within a population or between populations of a species or interspecies (Vellend & Geber, 2005; Rubenstein et al., 2005; Ellegren & Galtier, 2016), and it is known as polymorphism. Species diversity and genetic diversity have positive relationships (Vellend & Geber, 2005).

Genetic diversity is an important issue to describe population structure and to determine the destiny of populations for their future. Evolution is mainly proceeding based on the genetic diversity in populations. In addition, mutation, genetic drift, immigration, and natural selection are the driving forces of evolution as well as genetic diversity in a population (Vellend & Geber, 2005). A mutation produces new alleles, which have undergone the effects of selection process, and the survived and well-adapted new alleles contribute to expand the genetic diversity in a population or populations of a species. Genetic drift and immigration affect genetic diversity in addition to mutation and speciation according to the neutral theory (Kimura, 1983; Vellend & Geber, 2005) as well as non-neutral diversity (Lenormand, 2002; Mouquet et al., 2004; Vellend, 2005; Vellend & Geber, 2005). The influence of natural selection on genetic diversity is a complex process, which is affected by several factors such as spatial and temporal heterogeneity in the environment (Vellend & Geber, 2005), population type (panmictic or captive), and population size (Lacy, 1987). Spatial and temporal heterogeneity in the environment could be of two general types: exogenous heterogeneity and endogenous heterogeneity. The components of soil or climate conditions in the environment may vary in different localities, which are considered as exogenous heterogeneity, while endogenous heterogeneity is the characteristics specific to species or genotypes that provide advantage on others to be selected in the environment (Vellend & Geber, 2005).

Crop plants are important resources as food supply for human populations in the world. Therefore, anything that influences the crop plants will influence directly

human populations particularly. Founder crops domesticated about 10,000 years ago in the Fertile Crescent. Traditional farmers have been developing landraces or local varieties since domestication started. Thus, crop plants restored the diversified gene pools for the traits, which increase their adaptability to challenging biotic and abiotic environmental stress factors under the domestication period. Crop genetic diversity is crucially important as genetic resources for the development of high-yielding modern varieties with resistance to changing environmental conditions (Di Falco & Perrings, 2003). Crop genetic diversity ensures the maintenance of productivity and sustainability in agro-ecosystems, for example, reduction in crop genetic diversity leads to an increase in crop pests and pathogen populations, which will have opportunities to spread out in more areas and result in low yield (Sumner et al., 1981; Di Falco & Perrings, 2003).

Although many crop plants existed in nature, only a few crop plants such as wheat, rice, corn, oats, tomato, and potatoes are the major food sources for the global food supply. A dramatic reduction in genetic diversity due to several reasons such as preferring high-yielding varieties instead of local or landraces varieties, global climatic changes in the ecosystems, and destruction of natural habitats by anthropic activities imply pressure that makes the agriculture vulnerable and probably puts food resources under a risk (Di Falco & Perrings, 2003). Therefore, genetic diversity in wild crop plant relatives and farmers' varieties or landraces should be evaluated by using proper tools such as genetic markers and then be managed to exploit for improvement of cultivated varieties in breeding programs.

4.2 Evaluation of Genetic Diversity in Turkish Ancient Wheat Germplasm Using Genetic Markers

Genetic diversity is a matter of priority for continuation of life itself; therefore, it is very important in every aspect of life. A decrease in genetic diversity of a population or a species may lead to genetic drift, genetic bottleneck, or even extinction. In other words, the genetic diversity determines the fate of a population or a species. For efficient management of genetic resources, assessment of genetic diversity found in crop plants is of primary importance. Crop breeding for the development of high-yielding and resistant new varieties by using the germplasms of wild or primitive (landraces) relatives is a fundamental process to supply food for increasing human population on the world. The level of genetic diversity within or between individuals of a population or a species is also important for maintenance of ecological balance in an ecosystem or between ecosystems. Therefore, assessment of genetic diversity is of first-rate importance for all these cases (Mondini et al., 2009). The development of the techniques used for evaluation of genetic diversity follows the order of morphological markers, biochemical markers, and molecular markers.

4.2.1 Morphological Markers

Carl Linneaus used morphological markers to determine systematic relationships in plants for the first time in the eighteenth century (Schulman, 2007). Gregor J. Mendel crossed the individual common edible pea plants with opposite phenotypic characters such as flower (purple or white), seed (yellow or green), and pod (yellow or green) colors, seed (round or wrinkled) and pod (inflated or constricted) shapes, and flower position (axil or terminal) and scored their progenies according to their phenotypes (Schulman, 2007; Agarwal et al., 2008; Mondini et al., 2009). The fruitful results of these studies produced the basic principles of heredity known as Mendel's laws in the nineteenth century. Alfred H. Sturtevent (1913), a brilliant scientist, constructed the first linkage map by using six morphological traits (called "factor") in fruit fly (*Drosophila melanogaster*) (Andersen & Lübberstedt, 2003; Schlotterer, 2004; Agarwal et al., 2008; Mondini et al., 2009). Since then, morphological markers are still in use, but not frequent as much as before, because morphological markers are restricted in number and affected by environmental conditions due to the plasticity of the traits controlling them and pleiotropic gene effects of some other genes on morphological traits (Andersen & Lübberstedt, 2003) or epistatic interactions. Morphological markers have low polymorphism, and they could not have power to discriminate species. A good example for morphological diversity is the variation observed in spike color of *Triticum turgidum* ssp. *dicoccoides*: yellow, green, and black (Fig. 4.1).

4.2.2 Biochemical Markers

The limitations in morphological markers let the scientists look for more specific marker systems. Protein markers are the second generation in marker systems. Proteins are separated by electrophoresis, and band patterns of different individuals are compared according to their mobilities and intensities. The differences are indication of different alleles at a protein locus or polymorphism in the locus of interest. Seed storage proteins and isoenzymes are the most frequently used biochemical



Fig. 4.1 Spike color polymorphism in wild emmer, *Triticum turgidum* ssp. *Dicoccoides*, in Israel: yellow, green, and black color. (Photos taken by Ö. Özbek in 2005)

markers. Particularly isoenzyme markers are used in breeding programs and applied genetics. Isoelectric focusing is the technique used for resolution of isoenzymes. After electrophoresis, different alleles of an isoenzyme locus are identified with staining based on the catalytic activity of the enzyme (Tanksley, 1983). An example of *aminopeptidase-2* isoelectric focusing gel electrophoresis band patterns of *Triticum turgidum* ssp. *dicoccum* populations from Sinop–Durağan Sarıkadı, Sinop–Durağan Çandağı village, and Sinop–Durağan Köseli village is given in Fig. 4.2. The technique is easy and inexpensive, but expressions of band patterns are affected by environmental conditions (Schulman, 2007). Genetic variation in seed storage proteins of crop plants was used in breeding programs to improve the bread-making and pasta quality characteristics of modern wheat varieties. Some gliadin loci have linkages with some traits, for example, *Gli-1* loci linked with genes for resistance to fungal diseases (powdery mildew, *Pm3* locus; leaf rust, *Lr 10* locus; yellow rust, *Yr10* locus) and with genes controlling morphological characters of the plant such as glume hairiness (*Hg1* locus) and glume color (*Rg1* locus) (Pogna et al., 1994). The protein marker systems are limited in number and have low polymorphism; therefore, they were replaced by the development of molecular markers in time.

4.2.3 Molecular Markers

Morphological and biochemical markers have the limitations in numbers, and expression of low polymorphism level is displaying the genetic variation indirectly; however, the genes control those morphological and biochemical markers, which are affected by environmental conditions, and their expression may change based on the stage of development. A new genetic marker system displaying the variation directly on the DNA sequence of organisms has been evolved and called molecular markers, which can be defined as a DNA fragment or sequence found at a specific region on DNA. DNA markers might be associated with a gene or a visible phenotype or might not be associated (Agarwal et al., 2008). Molecular markers do not display any biological activity, as they are not normal genes, but they are localized permanently at specific positions in the genome. The standard law of inheritance

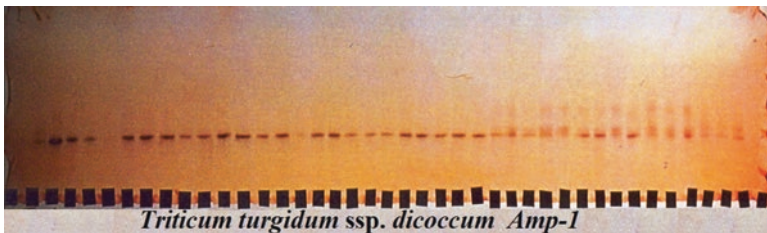


Fig. 4.2 *Aminopeptidase-2* isoelectric focusing gel electrophoresis band patterns of *Triticum turgidum* ssp. *dicoccum* populations from Sinop–Durağan Sarıkadı, Sinop–Durağan Çandağı village, and Sinop–Durağan Köseli village. (Source: Özbek, 1998)

from one generation to the next generation (Semagn et al., 2006) transmits them. In general, there are three types of molecular markers with different properties (Gupta et al., 1999; Agarwal et al., 2008).

- (i) Hybridization-based DNA markers: Restriction fragment length polymorphisms (RFLP) (Botstein et al., 1980) and oligonucleotide fingerprinting are examples for this system. In this marker system, DNA is digested with restriction enzymes; the restriction fragments are separated by using sub-marine agarose gel electrophoresis and blotted on a positively charged nylon membrane. Then, using different locus-specific probes up to 8–10 labelled with radioactive isotopes such as ^{32}P , the blotted membrane can be hybridized several times. RFLP markers are reliable and reproducible and may produce multiple loci per single marker and show codominant inheritance. Although it has several advantageous, it is a time-consuming and very expensive method; besides, the materials used are toxic for human health and environment. Therefore, today they have limited use for specific cases.
- (ii) PCR-based DNA markers: Technique is based on PCR amplification. These types of molecular markers are further divided into two groups. (i) Arbitrarily primed PCR-based techniques or sequence nonspecific techniques, in which there is no need to have prior sequence information and markers display dominant inheritance. Randomly amplified polymorphic DNAs (RAPDs) (Williams et al., 1990), amplified fragment length polymorphisms (AFLPs) (Vos et al., 1995), cleaved amplified polymorphic sequences (CAPS), and amplicon length polymorphisms (ALPs) are examples for this marker system. (ii) Sequence-targeted PCR-based techniques, in which the prior sequence information is necessary. Simple sequence repeats (SSRs) or microsatellites, sequence-tagged sites (STS), and inter-simple sequence repeat amplification (ISA) are examples for this marker, and they show codominant inheritance and produce multiple loci per marker.
- (iii) DNA chip and sequencing-based DNA markers: Advanced sequencing technologies made amenable millions of single nucleotide polymorphisms (SNPs), which is a single nucleotide polymorphism in the genomes of individuals in a population, in plant genomes (Patel et al., 2015). SNP markers are spread out in the genome and present abundantly, for example, maize has 1 SNP per 60–120 bp (Ching et al., 2002), while humans have an estimated 1 SNP per 1,000 bp (Sachidanandam et al., 2001; Agarwal et al., 2008).

4.2.4 Genetic Variation in Turkish Ancient Wheat Revealed by Genetic Markers

Turkish tetraploid ancient wheat (ssp. *dicoccum*, emmer) populations were characterized in terms of three isoenzyme [*endopeptidase-1* (*Ep-1*), *aminopeptidase-1* (*Amp-1*), and *aminopeptidase-2* (*Amp-2*)] systems, by isoelectric focusing gel

electrophoresis (Özbek et al., 2013). An example of *aminopeptidase-2* isoelectric focusing gel electrophoresis band patterns of *Triticum turgidum* ssp. *dicoccum* populations from Sinop–Merkez Eymir village and Çorum–Osmancık–Çampınar village is given in Fig. 4.3. Although wheat is a selfing plant and rarely outcrossing (less than 1%), researchers detected a considerably high level of genetic diversity ($H_e = 0.23$). It was noted that eco-geographical variables had significant effects on isoenzyme genetic diversity according to statistical analysis in this study. *Triticum turgidum* ssp. *dicoccum* (emmer wheat) has some important traits such as resistance to powdery mildew (Jakubziner, 1969), leaf diseases and common bunt (Corazza et al., 1986), yellow rust (Damania & Srivastava, 1990), and fusarium head blight or scab (Oliver et al., 2008). Those traits can be exploitable in durum and bread wheat breeding programs. McMillin et al. (1986) reported first time that there was an association between the genes coding *Ep-1* placed on homeoallelic series of Triticeae group 7 chromosomes and a gene conferring resistance to eyespot disease, an agronomic trait, for which *Ep-1* locus is an important marker (cited in Koebner, 1987).

Several types of molecular markers are extensively used in tribe Triticeae to investigate molecular genetic diversity (Vierling & Nuguyen, 1992; Hegde et al., 2000; Fahima et al., 2002; Barcaccia et al., 2002; Figliuolo & Perrino, 2004; Pagnotta et al., 2005; Teklu et al., 2006; Teklu et al., 2007; Terzi et al., 2007; Salunkhe et al., 2013) to determine the taxonomic relationships within the tribe Triticeae (Castagna et al., 1994); to map the genetic linkage between molecular markers and desirable traits such as drought resistance, disease resistance, and high yield (Kojima et al., 1998; Mondini et al., 2014); to assess the relationships between agronomic and quality traits and molecular markers in germplasms of wild wheats and primitive wheats (landraces) for marker-assisted selection in breeding programs in cereal crop genetics (Pagnotta et al., 2009); and to characterize genetic structures and genetic differentiation within and between populations in the same species (Özbek et al., 2007a, b).

Cenkçi et al. (2008) investigated phylogenetic relationships between Turkish *Triticum* and *Aegilops* species using RAPD markers. In that study, they compared the differentiation in the genomes of wild *Triticum* species (*T. monococcum* and *T. boeoticum*), *Aegilops* species [*Ae. tauschii*, *Ae. biuncialis* (UM), *Ae. crassa* 4X (DM), *Ae. crassa* 6X (DDM), *Ae. cylindrica* (CD), *Ae. geniculata* (UM), *Ae.*

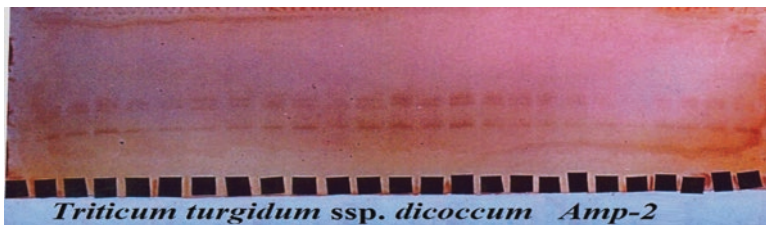


Fig. 4.3 *Aminopeptidase-2* isoelectric focusing gel electrophoresis band patterns of *Triticum turgidum* ssp. *dicoccum* populations from Sinop–Merkez Eymir village and Çorum–Osmancık–Çampınar village (Source: Özbek, 1998)

tauschii (D) *Ae. triuncialis* (UC) and *Ae. umbellulata* (U)], cultivated tetraploid wheat cultivars (durum wheat) [*T. turgidum* cv. Diyarbakır-81, *T. turgidum* cv. Ege-88, *T. turgidum* cv. Fırat-93, and *T. turgidum* cv. Kunduru-1149], and cultivated hexaploid wheat cultivars (common or bread wheat) [*T. aestivum* (BAD) cultivars (*Basribey-95*, *Bezostaya-1*, *Çukurova-86*, *Gerek-79*, *Gönen-98*, *Kasifbey-95*)]. The researchers reported that RAPD markers differentiated the species analyzed, and the results indicated that wild *Triticum* species and *Aegilops* species clustered together in a major group, but in different sub-clusters, while cultivated durum and bread wheat cultivars clustered in the second major group but in different sub-clusters.

Özbek and Demir (2019) investigated genetic diversity among the nine emmer wheat [*Triticum turgidum* L. ssp. *dicoccon* (Schrank) Thell.] landrace populations, grown in Turkey by SSR technique. Nine SSR primer sets produced 100% polymorphic 497 alleles, which ranged between 57 bp and 376 bp. The mean number of allele per locus (n_a), effective number of allele (n_{ea}), and value of genetic diversity (H_e) were determined as 40.89, 13, and 0.9, respectively. The genetic differentiation (F_{ST}) and gene flow (N_m) between populations were calculated as 0.15 and 1.41, respectively. The A genome showed higher genetic diversity estimates than the B genome. The SSR loci at telomeric and sub-telomeric regions showed lower genetic diversity than other regions on the chromosomes. The *x-gwm-312* is the marker of salinity tolerance gene in wheat. It was also used to screen polymorphism in emmer wheat populations, and it displayed the highest polymorphism ($H_e = 0.97$) among the SSR markers used for analysis. The high level of polymorphism observed for *x-gwm-312* marker might be an indication of potential salinity tolerance in Turkish emmer wheat samples stored in the gene bank. Salinity is one of the major problems in many wheat cultivation lands in the world. Turkish emmer wheat landraces having a high level of genetic diversity could be exploited to improve new wheat varieties adaptable to different climatic environmental conditions including salinity stressed environmental conditions in breeding programs. SSR marker system determined efficiently the genetic diversity among the emmer wheat populations, and it was successful to differentiate the different populations from each other.

The genetic diversity and population genetic structure in six *Triticum monococcum* L. ssp. *monococcum* landrace populations and two bread wheat (*Triticum aestivum* L.) populations from Bolu and Kastamonu provinces by using ISSR markers (Özbek & Zencirci, 2021). *Triticum monococcum* L. populations showed a higher genetic diversity ($h = 0.20$) than *Triticum aestivum* L. populations ($h = 0.14$). The high-yielding modern wheat varieties *Triticum monococcum* L. wheats lost competition with the high-yielding modern wheat varieties in agricultural production and are in danger of extinction today. The conservation of this important genetic resource as ex situ and in situ is an urgent necessity. For landraces, production on farm is a kind of in situ conservation, which will keep the dynamic evolution process and is compatible with the current ecological conditions. Einkorn wheat is also suitable for organic agriculture and healthy food in terms of food content.

4.3 Variation in Quality Traits

4.3.1 Vernalization

Environmental signals are received at one period of development and kept in mind until late periods to promote the growth and development in many organisms, and vernalization is one of these processes (Song et al., 2012). Vernalization is the prolonged exposure to low temperature, which is the requirement for transition to flowering in the reproductive success of plants (Schwabe, 1986; Michaels & Amasino, 2000; Finnegan, 1989; Song et al., 2012). It is critical process for some plants such as cereals and canola. There are three major genes, *VRN1*, *VRN2*, and *VRN3*, controlling the vernalization requirement in wheat as in barley (*Hordeum vulgare*) (Pugsley, 1970; Takahashi & Yasuda, 1971; Dubcovsky et al., 1998; Yan et al., 2006; Distelfeld et al., 2009). Mutations in *VRN1* gene prevent flowering and fail reproductive process in diploid wheat (*Triticum monococcum*) (Shitsukawa et al., 2007). Geographical variation in heading characters among wheat landraces and its application for adaptability (Kato & Yakoyama, 1992) and variation in vernalization and photoperiod responses in tetraploid wheat (*T. turgidum* ssp. *dicoccoides*) ecotypes (Kushnir & Halloran, 1982) and geographical variation in heading traits in wild emmer wheat (*T. turgidum* ssp. *dicoccoides*) and variation in vernalization response and ecological differentiation (Kato et al., 1997) were investigated.

4.3.2 Micronutrients

Micronutrients, which required trace amount intake, are crucially important for optimal metabolism in tissues. There are two kinds of micronutrients: inorganic micronutrients (trace elements or minerals) and organic micronutrients (fat-soluble and water-soluble vitamins). Many of the micronutrients are not produced in our bodies; therefore, we need to intake with our normal dietary food supplies such as fruits and vegetables. Inorganic micronutrients are functional as cofactors of enzymes, which are biological catalysts and accelerate chemical reactions such as zinc, which is the cofactor of more than 100 enzymes. Organic micronutrients are functional as coenzymes in the metabolism playing an intermediary role, for example, riboflavin and niacin take part in the electron transport chain. Nutrient deficiency or some metabolic disorders may cause micronutrient deficiency that leads to severe diseases or health problems. Some diseases such as anorexia or diarrhea may cause depletion of micronutrients. Socio-economic status of populations determines also nutrition quality. Low income is not sufficient to afford the costs for fruits and vegetables, and the result is inadequate nutrition (Finch et al., 1998; Hoare et al., 2004) and may cause a break in the development and healthy metabolism due to micronutrient deficiency. Bread is the most common foodstuff in every part of the world, particularly in developing countries with low income (Shenkin, 2006).

Cereals are the main source of calorie intake and used commonly for bread making since ancient times. In developing countries, nutrition is usually based on cereal foods; nevertheless, cereals contain less in both concentration and bioavailability of iron (Fe), and zinc (Zn). However, in developing countries, cereals are the primary source of iron (Fe) and zinc (Zn) (Welch & Graham, 1999). Thus, increasing the micronutrient concentration and bioavailability in cereals is one of the primary research interests.

Turkey is one of the countries that is experiencing zinc deficiency problem for both in crop plants (Cakmak et al., 1996, 1999) and human beings (Cavdar et al., 1983). Based on the studies about the zinc availability in cultivation areas indicated, Turkey had the lowest zinc concentration (Sillanpää, 1982, 1990), and 49.8% of the cultivation areas are recognized as zinc deficient (Lindsay & Norvell, 1978; Eyupoglu et al., 1994; Çakmak, 2008).

Ozkan et al. (2007) investigated micronutrients [copper (Cu), iron (Fe), manganese (Mn), and zinc (Zn)] in 54 accessions of einkorn wheat (*T. monococcum*). They reported that they found a great genotypic variation in content of micronutrients particularly in the concentration of zinc among the tested accessions, and they recommended that this variation could be used in breeding programs. Cakmak et al. (2004) determined the variation in Zn and Fe concentration and content in 825 tetraploid wild wheat accessions collected from Turkey and other parts of the Fertile Crescent (e.g., Iran, Iraq, Lebanon, Syria, Israel, and Jordan). They reported that spp. *dicoccoides* accessions having a high concentration of Zn than Fe were candidates as genetic resources for improvement of cultivated modern wheat varieties. Wild wheats and primitive wheats (landraces) have a higher Zn and Fe concentration and high variability compared to modern wheat varieties, and they could be used as donor parental lines in breeding programs (Cakmak et al., 1999; Cakmak et al., 2000; Monasterio & Graham, 2000).

4.3.3 *Wheat Storage Proteins*

Proteins are major food components in human and animal diet; therefore, they are also among the primary research interests since the eighteenth century, with the first protein research made by Beccari (1745) on wheat gluten. American protein chemist T.B. Osborne classified the storage proteins systematically based on the extraction and solubility of proteins in different mediums such as albumins dissolved in water, globulins dissolved in dilute aqueous solutions of salts, prolamins dissolved in alcohol/water mixtures, and glutelins dissolved in dilute acids or alkalis. Gliadins and glutenins are endosperm or seed storage proteins, also known as prolamins that provide amino acids for embryo in germinating seeds (Ciaffi et al., 1993). Seed storage proteins occupy 80% of the total proteins found in wheat grain. The germplasm of wild wheat and primitive wheats (landraces) was assessed with the storage proteins (gliadin and glutenin) to screen their relationship with technological properties (Ciaffi et al., 1991, 1992).

(i) Gliadins

Gliadins are alcohol soluble and a heterogeneous mixture of monomeric proteins, but disulfide bonds between the chains of some gliadins are observed, with the molecular size of which ranges between 28 and 70 kDa, and form 40% of storage proteins (Ojaghi & Akhundova, 2010). Gliadins are resolved at acid polyacrylamide gel electrophoresis (PAGE) (pH 3.1) up to 25 components, which are classified as alpha (α), beta (β), gamma (γ), and omega (ω) in the order of decreasing mobility on the gel (Bushuk & Zillman, 1978). Low lysine (0.5 mol %) content in gliadins causes a decrease in nutritional quality of wheat (Pogna et al., 1994).

Nineteen Turkish cultivated emmer wheat [*Triticum turgidum* L. ssp. *dicoccon* (Schrank) Thell.] populations were investigated in terms of gliadin polymorphism using the aluminum lactate-polyacrylamide gel electrophoresis (A-PAGE) method by Özbek et al. (2011). Landrace populations displayed considerably high amounts of genetic variation ($H_e = 0.92$) within and among the populations. Populations displayed 27 alleles (or different band patterns), and 10 patterns were unique to populations C, D, H, K, L, M, and N in the study (Fig. 4.4). Some gliadin subunits have association with dough quality such as the α -45 and ω -35 gliadins, which were determined in 13 and 18 of the landrace populations, respectively. The strong positive correlation ($r_p = 0.510$; $p = 0.026$ at $<0.05\%$) between the gene diversity estimates and latitude was calculated by statistical analysis of Pearson's correlation.

Keskin et al. (2015) investigated 10 Turkish cultivated einkorn wheats (*Triticum monococcum* ssp. *monococcum*) landrace populations to determine the polymorphism in seed endosperm proteins (gliadins and glutenins) using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) and A-PAGE (Fig. 4.5), respectively. The genetic diversity ($H_e = 0.65$) observed in gliadins was higher than the genetic diversity ($H_e = 0.65$) observed in high-molecular-weight (HMW) glutenin subunits. The interest in healthy food, produced through organic farming, is an increasing demand on the world recently. The einkorn wheat has a great potential for organic farming; therefore, the farming of einkorn wheat should be supported, and their germplasm should be maintained as important genetic resources for future plant breeding programs.

Alsaleh et al. (2016) investigated phenotypic and genotypic intra-diversity among Anatolian durum wheat "Kundurur" landraces. In the study, they analyzed polymorphism in gluten proteins. They observed high polymorphism in gliadin and glutenin banding patterns. They reported that many of the landraces studied are in danger of disappearing from the local farmers' fields; their study demonstrates the importance of maintaining and conserving these precious genetic resources.

(ii) Glutenins

Glutenins are classified as high-molecular-weight (HMW) and low-molecular-weight (LMW) glutenin subunits (Payne & Lawrence, 1983) according to their molecular sizes, and both of which have polymeric protein structures due to cross-linkages by disulfide bonds and HMW glutenins and have high glycine content.

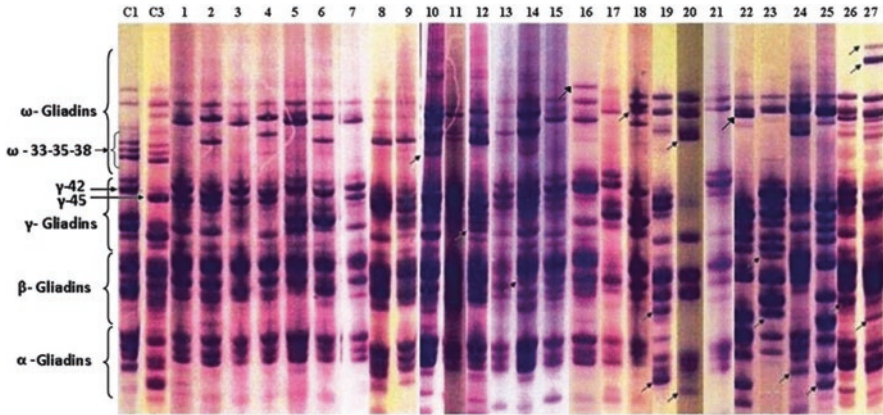


Fig. 4.4 Gliadin band patterns screened by A-PAGE method in Turkish emmer wheat populations. (Source: Özbek et al., 2011, *Plant Systematics and Evolution*, 296(0), 121–135, with permission of Springer Nature)

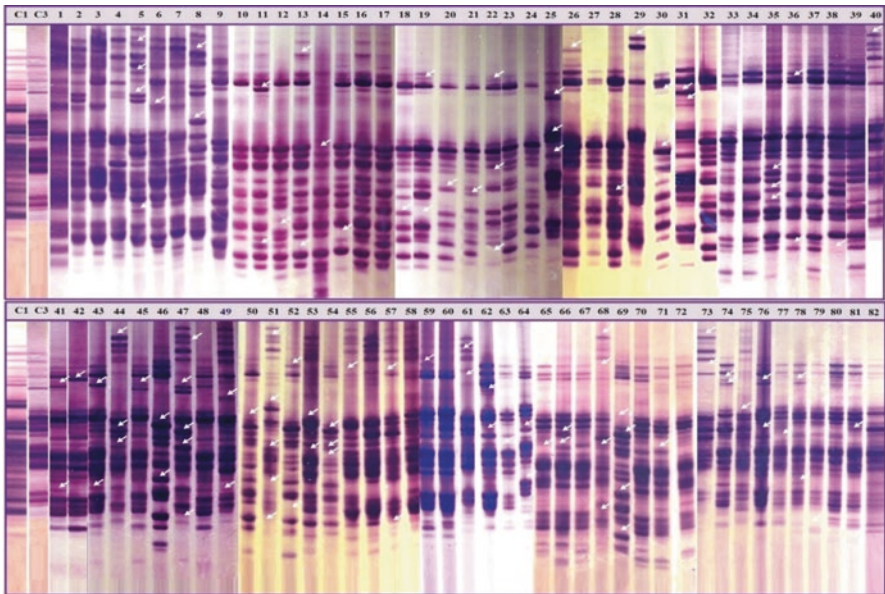


Fig. 4.5 Gliadin band patterns screened by A-PAGE method in Turkish einkorn wheat populations. (Source: Keskin et al., 2015)

Molecular weights of HMW glutenin protein, A-type subunits range between 80 and 120 kDa (Payne & Corfield, 1979). Common wheat cultivars bear 3–5 HMW glutenin subunits (Payne, 1987). Although, HMW and LMW glutenins are alcohol soluble in general, and some LMW glutenin subunits make heteropolymers with HMW glutenin subunits, which are insoluble in alcohol. These heteropolymers are soluble in dilute acid or alkali solutions in native state (Pogna et al., 1994).

LMW glutenin subunits are classified into two basic groups as major group of B subunits (MWs range 42–51 kDa) and minor group of C subunits (MWs range 30–40 kDa) due to breakage of disulfide bonds (Payne & Corfield, 1979; Pogna et al., 1994). Özbek et al. (2012) investigated the genetic diversity of high-molecular-weight (HMW) glutenin subunits in 18 cultivated emmer wheat populations, originating from Turkey, using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). The emmer wheat populations displayed a high level of genetic diversity ($h = 0.31$), genetic differentiation ($D = 0.24$), and gene flow between the different populations ($m = 0.16$). The eco-geographical variables might have significant influence on HMW glutenin diversity that was analyzed by using statistical analysis methods such as multiple regressions, principal component analysis, and *Pearson's* correlation. Examples of HMW glutenin subunit band patterns of Turkish emmer wheat and einkorn wheats (Keskin et al., 2015) are given in Figs. 4.6 and 4.7, respectively.

(iii) Protein Content

The amount of protein content varies between 10% and 15% in dry weight of cereal seed grains (Shewry & Halford, 2002). Protein content is important as much as the different gluten protein combinations related with good bread-making quality in wheat seed grains, because proteins are the components that absorb water and produce dough, which have viscose and elastic structure and contribute to have a high-volume potential (Oleson, 1994). For high-quality bread wheat, the protein content must be 11.5% on a 13.5% moisture basis. If it is less than 11%, it is unsuitable for bread making, when it is used alone (Tipples et al., 1994). However, the types of proteins and their expressions in cereal seed grains controlled by genetic factors, growing within sulfur- and nitrogen-rich environmental conditions, determine the protein contents (Tipples et al., 1994).

Konvalina et al. (2013) analyzed the baking quality of emmer wheat. Although they found high amount of protein (16.05–19.00%) in seed grains, emmer wheat has a lower baking quality compared to bread wheat varieties. Thus, they suggested that emmer wheat had a great potential for production at an industrial level for end use products such as biscuits, pasta, muesli, and various types of mush in addition to traditional use such as making bulgur, homemade macaroni, etc.

Protein content of Turkish durum wheat landraces was investigated in previous studies (Turchetta et al., 1995; Alsaleh et al., 2016). Turchetta et al. (1995) analyzed durum wheat landraces from the gene bank and found the protein concentration in the range of 11–19%, while Alsaleh et al. (2016) analyzed the collection of Kunduru-1149, an Anatolian landrace from six geographical provinces of Turkey, and found protein concentration between 12.10% and 14.90%. These studies indicated that Turkish durum wheat landraces had a wide variation in their protein content. However, Turchetta et al. (1995) reported that protein composition (HMW and LMW glutenin subunits) contributed 78% to the variation in SDS sedimentation, whereas protein content contributed only 6%. The remaining of variation is provided by other factors affecting quality such as other loci encoding storage or non-storage proteins and other chemical ingredients in the grain. The protein concentration found by Alsaleh et al. (2016) was lower than how much Turchetta



Fig. 4.6 HMW glutenin subunits screened by SDS-PAGE method in Turkish emmer wheat population from Sinop province (Source: Özbek, 1998)

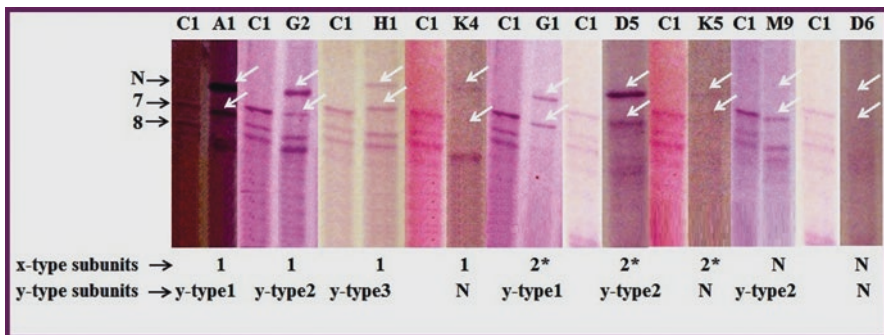


Fig. 4.7 HMW glutenin subunits screened by SDS-PAGE method in Turkish einkorn wheat populations. (Source: Keskin et al., 2015)

et al. (1995) found. However, Turkish durum landrace Kunduru-1149 populations are still quite large protein content; besides, they have shown great variation for other quality traits such as vitreousness (75–100%), TKW (31.80–56.70 g), YP (4.70–8.00 ppm), b*-value (14.30–19.50), and gluten strength (14–60 ml). The variation in these quality traits is also an indication of allelic variation in germplasm of Turkish durum wheat landraces that can be exploitable in breeding programs. Alsaleh et al. (2016) reported that Kunduru-1149 landraces are under the risk of extinction.

4.4 Status of Ancient Wheat's Genetic Resources in Turkey: Maintenance, Protection, Conservation Strategies, and Usage

Turkey has large animal and plant biodiversity due to its geographical location on the world. Turkey is located like a golden bridge between the continents of Europe and Asia. Based on the geographical structure, climate and environmental conditions show a wide range of diversity in Turkey. It has three different types of biogeographic regions, and each one has unique characteristics and natural ecosystems: the Caucasian mountain forests, Central and Eastern Anatolian steppe grasslands, and the cypress forests in the Mediterranean region. Turkey is the center of origin/or diversity center of some crop plants, which includes the wild relatives and wild ancestors of cereals that include those of wheat (*Triticum boeoticum*, *T. dicocoides*), *Aegilops* spp., barley (*Hordeum spontaneum*, *H. bulbosum*, *H. marinum*, and *H. murinum*), oats (*Avena* spp.), and rye (*Secale* spp.), primitive wheat forms (*Triticum dicocum*), and cultivated wheat forms. In Turkey, 3000 out of 9000 taxa identified are endemic plants (Tan, 2010).

Evaluation of genetic diversity in ancient wheats is a preferential action to determine the level of genetic diversity and population status, to determine intra- and inter-species relationships, and to manage the proper uses of gene pools of the species of interest for improvement of new varieties in breeding programs. In the last century, the ancient wheats have lost their popularities because of their low yield compared to modern new wheat varieties, and their cultivations were almost disappeared except in a few countries in the world. However, ancient wheats are rediscovered recently due to interest of healthy foods and organic farming practices. Ancient wheats are observed at all ploidy levels in the genus *Triticum* (Table 4.1).

Of these ancient wheats (primitive wheats or landraces), einkorn, emmer, and spelt wheats are still cultivated in a few countries, and the others are local types in the Transcaucasia regions (D'Antuono, 2008). Due to the importance of germplasms of ancient wheats in breeding programs, Turkish governmental agricultural research institutes and gene banks have started to collect ancient wheat seeds for ex situ conservation in earlier of the twentieth century. Turkey is one those countries that ancient wheats are still in cultivation by mountainous region farmers as fodder

Table 4.1 Ancient wheats within the different ploidy levels in the *Triticum* genus

Ploidy level	Species name	Genome formula and composition	Common name
Diploid	<i>Triticum monococcum</i> ssp. <i>monococcum</i>	2n = 2x = 14, A ^m A ^m	Einkorn wheat
Tetraploid	<i>Triticum turgidum</i> ssp. <i>dicoccum</i> (Schränk ex. Shübler) Thell.	2n = 4x = 28, BBAA	Emmer wheat
	<i>Triticum turgidum</i> ssp. <i>paleocolchicum</i> (Menabde) Á. Löve & D. Löve	2n = 4x = 28, BBAA	Georgian emmer wheat
	<i>Triticum timopheevii</i> (Zhuk) Zhuk. ssp. <i>timopheevii</i>	2n = 4x = 28, A ¹ A ¹ GG	
Hexaploid	<i>Triticum aestivum</i> ssp. <i>spelta</i> Thell.	2n = 6x = 42, AABBDD	Spelt wheat
	<i>Triticum aestivum</i> ssp. <i>macha</i> (Dekapr & Menabde) MacKey	2n = 6x = 42, AABBDD	Macha wheat
	<i>Triticum zhukovskyi</i> Menabde & Ericzjan	2n = 6x = 42, AABBDD	

rather than for human consumption. Although ancient wheats are still in use in Turkey, they have not received much interest as a research material and breeding material in wheat improvement breeding programs.

Crop genetic resources are mainly public goods and their contribution to the world is so valuable. They are the roots from which all crop production grows up in the world. Genetic diversity stored in crop genetic resources is the safeguard of future agriculture and food resources of human. Plant breeders spend much of their efforts for the development of new wheat varieties with desirable traits by exploiting wild and primitive wheat germplasms as genetic resources. However, new wheat varieties are pure monocultures and lack genetic diversity to cope with evolving pests (Rubenstein et al., 2005) and global changes in climate. The big genetic diversity loss in crop genetic resources in the last century will be the main threatening factor for future agricultural practices and food production process, because the development of a new variety takes 8–11 years, but loss of its genetic diversity and featured characteristics is about 5 years (USDA, 1990; Rubenstein et al., 2005). Therefore, international agreements have been signed to prevent, to reserve, and to maintain genetic diversity in crop genetic resources and to increase the share of germplasms worldwide such as International Treaty on Plant Genetic Resources for Food and Agriculture, which organizes the exchange of germplasm for crops like wheat, maize, and cotton. In practice, there are problems about exchanging the germplasms due to a lack of consensus between the parties (Rubenstein et al., 2005). Convention on Biological Diversity (CBD), “Article 2: Any material of plant, animal, microbial, or other origin containing functional units of heredity” is another agreement about reservation of genetic diversity and take covers the genetic diversity issues in the world.

One of the principles of the Convention on Biological Diversity is the sustainable use of biological diversity (Anonymous, 1996). The increasing human populations most probably will be faced with nutritional problems due to global warming and

climate changes. The solutions for nutritional problems will be searched in exploitation of plant genetic resources. Therefore, plant genetic resources should have sustainable genetic diversity to be exploited for the development of new varieties as food sources to feed human beings in the future. Genetic resources can be used directly and as gene progenitor in breeding programs. Many of the genetic resources living in the natural environment are the food resources for the people who live in rural areas. People collect them from nature and consume as food and use them for medical purposes or animal fodder, to produce dye for cloths, and for some other uses. The overcollection of genetic resources, particularly wild forms from the natural environment, leads to the extinction risk of some wild plant species, and also, it affects the genetic diversity in wild species, which are not cultivated in farmlands. Therefore, production of those wild plant species and their protection are important issues we should take significant care. Some primitive forms such as *Triticum monococcum* (einkorn) and *Triticum dicoccum* (emmer) have been cultivated for 10,000 years and consumed by traditional or local farmers and their families. This long cultivation period provided a wide genetic diversity in primitive wheat gene pools, which could be used to extend the genetic diversity of modern wheat genetic base varieties. The studies to increase the yield, the quality, and the stress factors, drought, cold-hot stress, resistance to winter conditions and tilt, microelement deficiency, breeding techniques, and resistance to pests and diseases, limit them in wheat production, which had started after the declaration of the Turkish Republic (Özberk et al., 2016). The maintenance of biodiversity/genetic diversity in Turkey is under the protection of the Turkish constitution, laws, regulations, and international conventions in the field of nature conservation, and further, national environmental strategies, plans, and programs are listed in Tan (2010).

4.5 Turkish Seed Gene Bank (TSGB) of Central Research Institute for Field Crops (CRIF)

The first gene bank was founded to identify genetic resources and to collect and to store those genetic materials by single hand at an international standard level in Turkey in 1963 and was named as Aegean Agricultural Research Institute (AARI). Perhaps, besides this gene bank, there were several collection places varying in capacity and the seeds for were stored ex situ for conservation purposes. After that, the second formal gene bank of Turkey was founded within the Central Research Institute for Field Crops with a project contribution supported by the United Nations in 1987 in Ankara (Balkaya & Yanmaz, 2001).

In 2010, a Turkish seed gene bank (TSGB) with large capacity (250,000 specimens) was constructed. The TSGB has 1043 m³ cold space volumes and contains the following sections: documentation, seed preparation including drying and packaging units, breeding and characterization, seven cold rooms, seed physiology laboratory, molecular biology laboratory, a herbarium that has a capacity of 60,000

specimens, and screening room. It is estimated that approximately 30,000 specimens are stored in cold rooms of the TSGB, while in the herbarium, 6000 specimens are stored.

The TSGB is serving to carry out the following issues:

- Collection of genetic materials
- Providing genetic material needed from external origin
- Storing Turkish plant genetic resources materials to be used in breeding and improvement of modern developed varieties or cultivars
- Registration of genetic resources
- Creation of a database for natural genetic resources
- Training of local people about uses and conservation of genetic resources
- Characterization of genetic material, which is stored in seed gene bank with cooperation of universities, research institutes, and other institutions
- Developing the collection of microbial cultivars, which have a potential to be used in industry, in agriculture and as solution for environmental problems

4.6 Wheat Production Systems

4.6.1 Organic Agriculture

Primitive agricultural practices started with domestication of wheat around 10,000 years ago in the Fertile Crescent. Subsequently, domestication of high-yielding wheat species (durum wheat and bread wheat) and advances in knowledge about agricultural production systems led to an increase in food production, which enabled to feed more people. All these practices were called as traditional agricultural production systems, which needed low input and no chemicals such as synthetic fertilizers and pesticides, and certain veterinary drugs were used for production (Snyder & Spaner, 2010). Today, this kind of agricultural system is called organic agriculture, which aims the maintenance of a sustainable ecosystem, food security, well feeding, animal prosperousness, and social justice (IFOAM; Snyder & Spaner, 2010). The land area under organic management worldwide was 11 million ha and 37.2 million ha in 1999 and 2011, respectively (FAOSTAT).

4.6.2 Status of Organic Agriculture in Turkey

Based on the Food and Agriculture Organization (FAO) statistics, the area used for organic agriculture was 2% of total agricultural area (http://faostat.fao.org/static/syb/syb_223.pdf) in 2014. According to TURKSTAT records based on data of the Ministry of Food, Agriculture, and Livestock of Turkey (MFALT), the number of crops, number of holdings, area used, and amount of production in 2002 were 150,

12,428, 89,827 ha, 310,125 t, respectively, while in 2016, records were 238, 67,878, 523,777 ha (1.7%), and 2,473,600 t (35.2%), respectively. Natural harvesting areas were also included. From the statistical records, it is clearly seemed that organic agriculture is also raising trend of interest in Turkey as observed in the world (TURKSTAT). In the remote mountainous areas, traditional farmers still keep the cultivation of wheat landraces in Turkey. Arable areas for agriculture are usually very small, nearly less than <5 ha due to division of fields by heritage from fathers to their descendants. The distance between villages and crop markets is long and results in expensive transportation costs for poor peasants. Therefore, they usually prefer low input agriculture and use landraces or local wheat varieties. Farmers use traditional agricultural instruments such as sharpening stone, which is used to sharpen the reap hook (Figs. 4.8 and 4.9), and the reap hook is used for harvesting the matured crops (Fig. 4.10). After harvesting, the hays or chaffs are broken down into straws, which is used to feed animals during long winter season by using simple machinery mechanisms (Fig. 4.11).

4.6.3 Conventional Agriculture

The increase in the size of human populations and development of many different types of plant diseases, which cause reduction in the yield capacity and quality of harvested crop seeds due to dynamic evolutionary process of pathogens, global climate changes, and limited areas for agricultural production on the world, increase the demand for high-yielding new wheat varieties with good quality characters and increased agricultural production. These problems compelled to develop modern



Fig. 4.8 An apparatus used to sharpen the reap hook. (Photo taken in 2017 by Özbek with permission)



Fig. 4.9 A reaper is sharpening the reap hook from Gökçeyayla village of Eskişehir province. (Photo taken in 2017 by Ö. Özbek with permission)



Fig. 4.10 A reaper is harvesting the wheat crops in Gökçeyayla village of Eskişehir province. (Photo taken in 2017 by Ö. Özbek with permission)



Fig. 4.11 After harvesting, the villagers are throwing the hays or chaffs into a machine, which is used for copping down to straw. (Photo by Tuncay Ay in 2017, with permission)

conventional agricultural techniques, use chemicals such as synthetic fertilizers to increase the yield, and develop new wheat varieties, which were adaptable to specific environment or adaptable to wide eco-geographical conditions in breeding programs.

The replacement of wheat landraces with high-yielding new wheat varieties reduced the genetic diversity in the last century (Jaradat, 2013). The genetic diversity has been narrowed in high-yielding new wheat varieties due to the applied strong artificial selection methodologies in breeding programs. The new wheat varieties are more susceptible to biotic and abiotic stress factors compared to their wild and primitive wheat relatives. Therefore, it necessitates using pesticides and veterinary drugs to combat with biotic stress factors, while synthetic fertilizers were used to increase the yield. Nevertheless, using pesticides, veterinary drugs, and synthetic fertilizers in agriculture and their accumulation in soil led to soil pollution, which has negative impact on the soil ecosystem, and their precipitates on harvested farm products are supposed to cause severe health problems on the other organisms in the food chains of its ecosystem. The use of fertilizers (kg of nutrients per ha), nitrogen phosphate, and potash were 71.6, 29.8, and 4.7, respectively, in 2014 (FAOSTAT, 2014). The use of high amount of synthetic fertilizers, which dissolve with rainfall or irrigation, contaminates directly underground waters and indirectly the water reservoirs that underground waters reach such as rivers, lakes, and sea ecosystems.

In Turkey, agricultural crop production is based on mainly conventional agricultural breeding, which has high input and used modern agricultural techniques and equipment. Farmers use seeder for dispersion of seeds and harvester for harvesting grains and separating seed grains from their chaffs and then evacuate the seed grains



Fig. 4.12 A harvester, which is used for harvesting matured crop spikes of bread wheat field. (Photo by E.M. Demircan in 2017 with permission)



Fig. 4.13 After harvesting, the harvester separated seed grains from their chaffs, and it is evacuating the seed grains into a trailer. (Photo by E.M. Demircan in 2017 with permission)

into a trailer (Figs. 4.12 and 4.13). All these agricultural practices increase the input of conventional agriculture in Turkey. Therefore, the Ministry of Food, Agriculture, and Livestock of Turkey provides financial support of 13 TL/ha and 4 TL/ha for diesel fuel and fertilizer, respectively, to the farmers, who registered in Farmers Registration System in 2017 (<http://www.tarim.gov.tr>).

4.7 Breeding of Ancient Wheats

A renewed interest for organic agriculture brought forward einkorn and emmer wheat in Turkey simultaneously with the world. Einkorn and emmer wheat landraces are cultivated at remote mountainous regions, where the soil and climatic conditions are not favorable for high-yielding new wheat varieties. Therefore, einkorn and emmer wheats have a great potential for organic agriculture or for production of local foods.

A study of “Wheat Landraces in Farmers’ Fields in Turkey: National Survey, Collection and Conservation” was carried out between 2009 and 2014 under the concern of a project entitled as “International Winter Wheat Improvement Program (IWWIP)” coordinated by the Ministry of Food, Agriculture, and Livestock of Turkey (MFALT), the International Maize and Wheat Improvement Center (CIMMYT), and International Center for Agricultural Research in the Dry Areas (ICARDA) and the participation of the Food and Agriculture Organization (FAO) in the last 2 years of the project (Kan et al., 2014; Kan et al., 2016). In this study, the inventory of wheat landraces of Turkey was built up, and breeding techniques and socio-economic conditions of local wheat producers were surveyed by making questionnaire with 1873 landrace-producing farmers by asking questions face to face in 65 provinces (Kan et al., 2017). In this survey, 162 different wheat landraces were collected, and the names of the 12 wheat landraces came out were “Zerun, Ak, Kırmızı, Sarı, Karakılıçık, Kırık, Siyez, Koca, Topbaş, Şahman, Üveyik, and Göderedi.” Partition of collected material from 65 provinces was as follows: 58.28% used for bread making such as Zerun, Kırmızı Buğday and Kırık; 37.93% used for macaroni and bulgur such as Siyez, Şahman, and Sarı Buğday; and 3.79% used for animal fodder. The pictures of some landraces, which were cultivated in this project in the fields of Bahri Dagdas International Agricultural Research Institute (BDIARI), were given below (Figs. 4.14, 4.15, 4.16, 4.17, and 4.18).



Fig. 4.14 Cam wheat (red, Kırmızı) from Tokat. (Photo by Ö. Özbek in 2017 with permission of BDIARI)



Fig. 4.15 Üveyik wheat from Tokat. (Photo by Ö. Özbek in 2017 with permission of BDIARI)

4.7.1 *Einkorn Wheat Breeding*

The name einkorn is given for both the wild diploid wheat *Triticum boeoticum* and domesticated *Triticum monococcum*. Einkorn wheat (from German einkorn, literally “single grain”) has $2n = 14$ chromosomes and genome formula $A^m A^m$, which was the most popular cereal in 7500 BC; nowadays, it is a relict crop and cultivated in limited countries including France, Morocco, former Yugoslavia, Italy, and Turkey in the world (Cooper, 2015). Its Turkish local names are “siyez,” “kavılca,” and “gernik.” It is an ancient-type primitive wheat due to its tough glume, in which its tough, tight case encloses the seed grain, and it is not threshed from the seed. It has a new place in markets as healthy food such as bulgur (cracked wheat), and it is grown mainly as fodder for animals today (Cooper, 2015).

Archaeological remains displayed that einkorn wheat was used by civilizations existed in Anatolia such as Hittite Empire and Phrygia Kingdom. Hittites gave the first local name “zız” to einkorn wheat. Nevertheless, einkorn wheat is growing only



Fig. 4.16 Kırmızı Evlek wheat from Manisa. (Photo by Ö. Özbek in 2017 with permission of BDIARI)

in few provinces: Çankırı, Çorum, Kayseri, Sinop, and Kastamonu province particularly in its İhsangazi district in Turkey today (Fig. 4.19). The total wheat production is 160,760 t, and 3500 t of it is einkorn wheat in Kastamonu province, while the amount of einkorn wheat yield is 1250 t (15%) of 8250 total wheat production in İhsangazi district, and the area used for einkorn cultivation is around 5000 da (Özberk et al., 2016).

In a Hittite tablet, some important information was given about the agricultural activities of ancient societies such as proprietorship of land, cereal species, their cultivation, plowing, sowing, properties of the field (fertility, hilly, plain, irrigation, crop), yield, reaping, harvesting, processing of food, diseases that affect plants, types of pests, jars, pithos, and the earth dug out holes. In addition, it is reported that Hittites had 180 kinds of bread, pastry, cakes, and noodles. The text also referred that slaves were charged and forced to be participated in agricultural activities such



Fig. 4.17 Ormece wheat from Tokat. (Photo by Ö. Özbek in 2017 with permission of BDIARI)

as plowing, reaping, garnering, and threshing. According to archaeobotanical samples collected by European, Japanese, and American archaeologists, the common attested cereal species were einkorn, emmer, bread wheat, club wheat, and ancient and naked barleys (Ünal, 2016).

4.7.2 *Emmer Wheat Breeding*

A mutation event occurred in the gene that controlled the brittle rachis trait in wild emmer wheat (*Triticum turgidum* L. ssp. *dicoccoides*], which gave rise to emmer wheat [*Triticum turgidum* L. ssp. *dicoccon* (Schrank) Thell.] with the genome



Fig. 4.18 Sorghul wheat from Mardin. (Photo by Ö. Özbek in 2017 with permission of BDIARI)

formula of *BBAA* ($2n = 4x = 28$) and non-free-threshing ancient wheat. Its local names are “gernik,” “çatal siyez,” “kablıca,” “kabalca,” and “kavılca” (Kıpçak, 1951; Şekercioğlu, 2012). It has extremely tough case enclosing the seed grain that makes it hard and adaptable to cold climatic conditions compared to modern wheat varieties. The milling and grinding process is also very hard, which requires extra energy due to its hard and tough case. Therefore, farmers are not willing to cultivate emmer wheat anymore. Only few traditional farmers are growing it for their home consumption as bulgur, couscous, homemade macaroni, and animal fodder. In Kars province, people like to eat goose meat with emmer bulgur, and also, it is used for making lahana dolması, a traditional Turkish food, in which cabbage leaves are stuffed with a mixture of bulgur and minced meat. Emmer wheat presents low but a stable yield to farmers. Cultivation of ancient wheats started to decrease in the



Fig. 4.19 A map displaying the cultivation areas of einkorn wheat (*Triticum monococcum* ssp. *monococcum*) today in Turkey. (Source: Özberk et al., 2016)



Fig. 4.20 A map displaying the cultivation areas of emmer wheat (*Triticum turgidum* ssp. *dicocum*) today in Turkey. (Source: Özberk et al., 2016)

second half of the twentieth century with the development of high-yielding wheat varieties. According to data of a study carried out in 1992, einkorn and emmer wheats are growing in six provinces (Fig. 4.20), and the amount of total production of ancient wheats is 22 million tonnes, which is a tenth of a one whole production of wheat in Turkey (Karagöz, 1995; Özberk et al., 2014).

In 2006, a civil organization called Yer Gök Anadolu (Ground Sky Anatolia) Association started a project about the refreshing and expansion of agriculture of local ancient wheat varieties in Kars province. The project was carried out between 2007 and 2010, and the United Nations organization supplied one-year financial support to the project. At the beginning, 2000 tonnes seed grains of emmer wheat were collected from the different districts of Kars province, and about 100 kg seeds were distributed to local farmers, who were willing to grow local wheat landraces in their farms. Today, more than 450 farmers are growing emmer wheat. The project was awarded and got great attraction and appreciation in the world (Anatolia Foundation, 2017).

4.7.3 Hexaploid Ancient Wheat Breeding

Hybridization between a *T. turgidum* subspecies (Kihara, 1944; McFadden & Sears, 1946) and the diploid goat grass *Ae. tauschii* led to a hexaploid ancient wheat type, which might have been similar to Asian spelta (*T. aestivum* ssp. *spelta* L., $2n = 6x = 42$, *AABBDD*) due to the presence of *Tg1* gene from *Ae. tauschii* around 9000 BC (McFadden & Sears, 1946). The free-threshing hexaploid bread wheat (*T. aestivum* ssp. *aestivum* L., $2n = 6x = 42$, *AABBDD*) evolved by gaining free-threshing character from this subspecies (Faris, 2014). The different hexaploid ancient subspecies of *T. aestivum* are ssp. *macha* Dekapr. et Menabde, ssp. *European spelta* L. (Thell.) and ssp. *vavilovii*. *Triticum spelta* L. wheat was grown mainly in European countries particularly in southwestern Germany and Switzerland, Spain, Italy, former Czechoslovakia, Poland, Ukraine, and the Asian spelt (*T. spelta* L. ssp. *spelta*) in Iran and *T. spelta* L. ssp. *kuckuckianum* Gökg. in Tajikistan, Uzbekistan, and Turkmenistan (Blatter et al., 2004) until the end of the eighteenth century. However, the number of countries cultivating spelta wheat decreased. An increasing trend for healthy food renewed the interest for spelta wheat farming in European countries at the end of the twentieth century. Spelt wheat is used for the production of pasta products, muesli, and flakes rather than for bread making for which it is not suitable (Bojňanská & Frančáková, 2002).

4.8 What Factors Are Threatening the Wheat Biodiversity in Turkey?

Wild and primitive wheat landraces are endangered species in the world today. Their primary importance is their gene pools, which are important genetic resources to improve new wheat varieties with desirable traits. First, the threatening factors of the wheat biodiversity should be determined properly, and conservation strategies should be managed based on those factors to protect the wheat biodiversity at a stable level.

The factors threatening the wheat biodiversity are the following:

- The use of pesticides and veterinary drugs for plant diseases, pathogens, and weeds.
- Over usage of synthetic fertilizers to increase the yield causing the pollution in the soil, underground water, and finally large water resources.
- The replacement of local varieties due to low yield with high-yielding wheat varieties that decreased the genetic diversity. Fallow systems are causing loss of soil and decreasing the productive layer in the soil that also decreases the yield.
- Preferences for high-yielding wheat varieties, which are monocultures that cause a decrease in the genetic diversity. Because a few registered new varieties are developed for specific eco-geographic and climatic conditions and monotype.

Therefore, they cannot bear all the genetic diversity stored in wild and primitive wheat forms.

- Firing stubbles is destroying the ecosystems in the soil. The members of the soil ecosystems are rotating the natural cycles in the soil. Damaging those ecosystems will diminish the fertility of soil.
- Drying wetlands for expansion of agricultural lands, increasing salinity due to dry climate regime, decreasing amount of underground water, and vanishing natural habitat among the farming fields have negative impacts on biodiversity worldwide.
- An increase in soil erosion decreases productive layer of the soil, pollution of natural water resources affects all living organisms, and a decrease in the amount of organic nutrients in soil also decreases the fertility, yield, and genetic diversity.
- Urbanization, industrialization, and damaging agriculturally productive fields for investment of tourism are destroying the natural habitats and leading to a decrease in biodiversity.

4.9 Conclusion

After the wild wheats were subjected to the domestication process, domesticated wheat varieties were cultivated by traditional farmers; nevertheless, they were replaced by the cultivation of naked free-threshing wheat varieties. However, the wild wheats and primitive wheats or landraces have substantial genetic diversity; harboring unique genotypes makes them adaptable to different environmental conditions, particularly adverse conditions in remote mountainous places. The local environmental conditions have enabled landrace populations to have a dynamic genetic structure and to continue their evolutionary history in the farms where they can easily adapt to changing environmental conditions. Genetic diversity is an important issue to describe population structure and to determine the destiny of populations for their future. Crop breeding for the development of high-yielding and resistant new varieties by using the germplasms of wild or primitive (landraces) relatives is a fundamental process to supply food for the increasing human population on the world. Therefore, efficient management of genetic resources and assessment of genetic diversity are important. For evaluation of genetic diversity, morphological markers, biochemical markers, and molecular markers have been used for several decades. The level of genetic diversity within or between individuals of a population or a species is also important for maintenance of ecological balance in an ecosystem or between ecosystems.

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Chapter 5

Conservation Strategies



Hakan Ulukan, Nusret Zencirci, and Mehmet Örgaç

5.1 Introduction

It is interesting that according to ethnobotanical records, at the level worldwide, more than 7000 plant species are cultivated or harvested from the wild (Moudry et al., 2011); and crop domestication and improvement can be described as a process of successive rounds of selection that ultimately results in the isolation of genetic diversity valuable to agriculture from ancestral wild species, some key requirements for the more effective use of crop wild relatives (CWRs) in breeding (Dempewolf et al., 2017). The ancient wheats (einkorn [wild/*Triticum boeoticum* and domesticated/*T. monococcum*], emmer [wild/*Triticum turgidum* subsp. *dicocoides*, domesticated/*Triticum turgidum* subsp. *dicoccum* and *Triticum turgidum* conv. *durum*] and spelt [*T. spelta*]) are the wild wheat species, and they are among the most ancient cereal crops of the Mediterranean region, as known (Fig. 5.1) (Padulosi et al., 1996; Longin et al., 2016). In addition, these crops have non-fragile spikes, and their ancient grains were the earliest species domesticated and may be the oldest, almost 10000 years ago during the Neolithic period. In addition, these crops are currently underutilized or simply neglected in the past and were widely used as shown by the historical records, and they are among the most ancient cereal crops in Europe, where remains have been found in the later Neolithic sites (2500–1700 BC) in eastern Germany, Poland, and Jutland (Caballero et al., 2007). The abovementioned ancient wheats were originated in the Fertile Crescent (Fig. 5.2).

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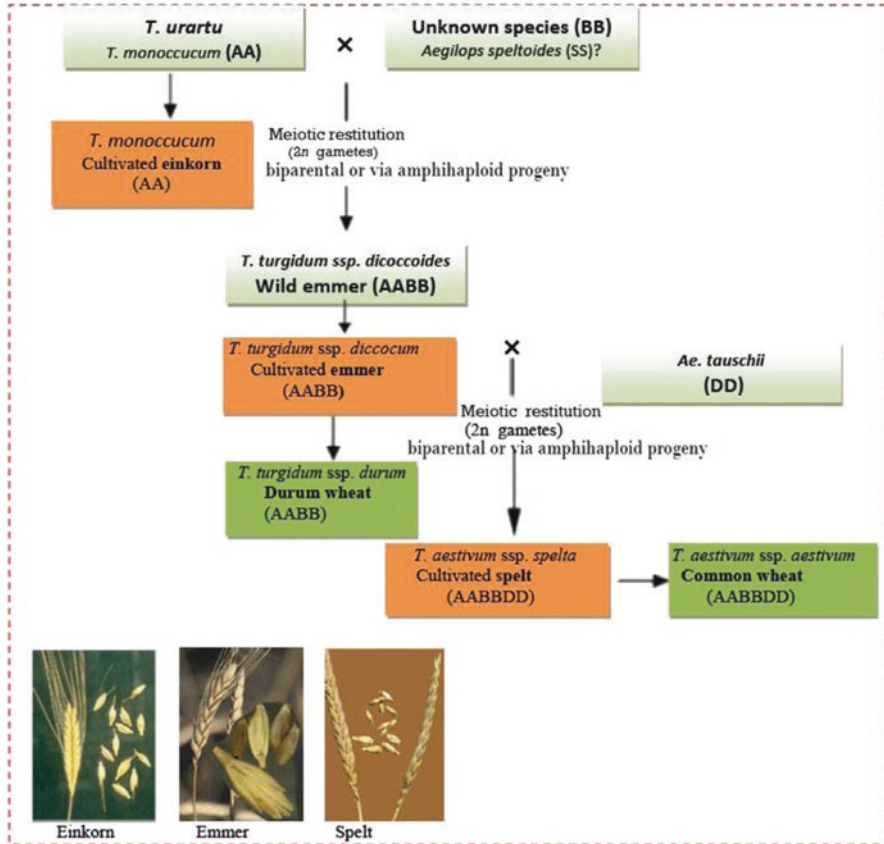


Fig. 5.1 Illustration of taxonomy and evolution of the ancient wheats (Arzani & Muhammad, 2017)

The term “ancient/hulled wheat” is applied to the wild or cultivated species of the genus *Triticum* having the characteristic to present the glumes close to the grain even after the normal threshing (Caballero et al., 2007). The grains of the ancient wheats (as mentioned above) are covered by glumes (hulls) even after harvest. If we grow them in marginal regions, they provide a lower but more stable yield rate (Moudrý et al., 2011). However, the disadvantage(s) of these wheats is/are that their grains are difficult to thresh because the hulls remained attached after harvest and threshing. Top it all off, the toughened glumes enclosed the grain and provide protection against pests and disease during the storage. But, at the same time, this structure(s) has/have some difficulties that, due to the hardness of itself and they easily block for the water and O₂, etc. intake to the grain during the germination, caused the partly dormancy, etc.. Nearly all cultivated wheats were developed from the wild wheats using related breeding techniques (such as hybridization, selection, doubled hybridization, tissue culture, mutation, molecular marker), and all these processes have bestowed the grain yield advantages to the modern wheat over its



Fig. 5.2 The Fertile Crescent area where the ancient wheats originated (Arzani & Muhammad, 2017)

ancestral ancient wheats (Arzani & Muhammad, 2017). But this is a crystal clear fact that all the wild relatives of crop plants constitute invaluable gene resources for crop improvement (Nevo, 1998). The ancient wheats – emmer, einkorn, *spelta*, *tauschii*, and *vavilovii* – are very important not only in terms of taxonomy but also for seed conservation, for good parents in plant breeding programs, for ecosystem services, and ecophysiology but also very valuable for unexplored the PGRs. For example, as will be discussed in further chapters, they are sources of various biotic or abiotic stresses or relevant quality characteristics. Although their yield level is low, therefore, as mentioned above, they are being preserved in situ (mostly) in the farmer’s field and at the seed store facilities and ex situ (minor) in national and/or international seed banks.

PGRs (including wilds) in wheat plants can be classified into 6 broad groups (Anonymous, 2017):

1. Modern cultivars in current use
2. Obsolete cultivars, often the elite cultivars of the past and often found in the pedigrees of modern cultivars
3. Landraces
4. Wild relatives of crop species in the Triticeae tribe
5. Genetic and cytogenetic stocks
6. Breeding lines

Furthermore, the kinds of plant material in the flora can be mentioned as follows (Şehirali & Özgen, 1987):

1. Agricultural centers

- (a) Currently improved cultivars for particular aims
- (b) More or less homogenous cultivars with a narrower genetic base than natural landraces
- (c) No longer used old cultivars
- (d) Primitive or local cultivars

2. Gene centers (which means wild species and transition forms)

3. Plant improvement centers

Usually, the ancient wheats are considered as either wheat landrace or wild wheats due to their genetic base broadness. Wheat landrace collections contain a wider genetic diversity than most breeding programs, and this diversity includes adaptation to different conditions according to the place of origin. But according to records, there are two potential bottlenecks in wheat diversity. The first one is the recent origin of the bread wheat, the presumption that there are relatively few tetraploid and diploid progenitor crosses. Hence, only a portion of the diversity of *T. dicoccoides* and *Aegilops squarosa* exists in bread wheat; and the second one is to founder lines for (local) populations where breeding programs often rely on a relatively limited number of parents in developing germplasm pools.

As known, wild relatives of crop plants constitute invaluable gene resources for crop improvement. As it is clear, it is essential to conserve by collecting, conserving, evaluating, and utilizing the PGRs, in other words, genetic diversity since they disappear very fast. Conservation operation is divided into two major classes: (i) in situ (on site, in habitat/ecosystem) and ex situ (off site, out of place, in seed banks). The fact remains that the genetic diversity and richness for resistance genes against biotic and abiotic stress factors of the ancient wheats should be kept at the genebanks (with ex situ and in situ conservation methods), and as the need arises, they should be used in the hybridizations as a parent.

Gaps in Collections

There was inadequate conservation of landraces, primary wild relatives, and cultivars (in descending order of importance). This contrasts sharply with the responses from collection users whose perception of the deficiencies in the current conservation of wheat genetic resources was lack of mapping populations, mutant and genetic stocks mutants, and the wider range in them. Genetic erosion and genetic pollution, are caused by several factors such as anthropogenic and/or natural changes, the gene flow from conventional and biotechnologically bred crops, and the introduction of exotic and alien species (especially treat to in situ conservation), being one of the major factors contributing to the decrease of the ancient wheat diversity, can be defined as “*the permanent reduction in richness or evenness of common localized alleles or the loss of combination of alleles over time in a defined area*” (Carvalho et al., 2012). Again, according to Carvalho et al. (2012), these two genetic effects will do a domino effect, thus affecting the agricultural sector. There is a wide variation among the different varieties within each species especially for

grain yield, plant height bearing a high risk of lodging, heading time especially (tall) the plant height, and high protein content (Longin et al., 2016). In addition, wheat landrace (including the ancient wheats) collections contain a wider genetic diversity than the most breeding programs, and this diversity includes adaptation to different conditions according to the place of origin. On the other hand, to be able to use the genetic variation in the wheat landraces, current breeding methods are direct crossing and introgression for all the adaptive traits, which are attractive breeding aims, selection(s), and mutation.

Preservation Methods for Genetic Diversity in Ancient Wheats

5.2 Conservation

5.2.1 Ex situ Conservation Situ

This conservation technique implements the conservation target's natural habitat (Heywood & Duloo, 2015); it has some difficulties (mainly money, time, labor force, etc.) and obligated process (such as storing in huge climate factors). For this, large farming areas for cultivation, human power for harvesting and other breeding activities, and financial funds for covering all these considerably expensive expenses are needed. In addition, during these breeding activities, there are some risks such as climate, botanical, practical for selfed plants, which may cross-pollinate or mix the seeds of different genetic materials by mistake, and great attention should be given to prevent reduction in genetic diversity due to strong selection factors and interspecific completion during ex situ regeneration of seeds (Özbek, 2014).

Genebanks conserve seed samples as basic or active collections, or non-seed propagated material conserved at the field genebanks in various research institutions (Şehirali & Özgen, 1987).

This ex situ conservation process is carried out outside the following growth systems (Şehirali & Özgen, 1987):

- (a) Cultivation of fruit trees in artificial plantations that requires larger areas in practice
- (b) Conservation of vegetative propagating plants with tissue culture
- (c) Sexually propagated plant seeds in seed banks
- (d) DNA storage and conservation
- (e) Pollen storage and conservation
- (f) Field genebanks
- (g) Botanical gardens

This method has some advantages (A) and disadvantages (D), too:

- (i) Ex situ is usually much more preferred by national/international seed banks in the world (A/D).

- (ii) When natural habitat become insecure or insufficient, the species are withdrawn from the environment and flora. These are picked up for *ex situ* conservation (A/D).
- (iii) It is preferred mostly by academicians, science men, seed banks, etc. (A).
- (iv) *Ex situ* conservation is used for the seeds of more dangerous and strategic PGRs (A).
- (v) The maintenance and conservation of the seeds of the CWRs are readily available gene sources against pests and diseases (Hoyt, 1988; Davies, 1991) (A).
- (vi) Foremost, among other reasons, capturing, particularly preserving and utilizing genetic variation in the CWRs (poorly known genomes, low seed setting and production, viability, seed recalcitrance, etc.) are the difficulties, which carry often higher costs (Berjak & Pammenter, 1997) (D).
- (vii) *Ex situ* conservation requires lots of research activity (D).

Due to the *in situ* conservation is more often used by plant breeders and geneticists (A/D). It has eight steps: (1) regeneration (a must for maintenance of an optimal number of accessions), (2) CWRs (proved as sources of biotic and abiotic stress factors), (3) collecting (to fill gaps in genebanks and preserve exclusive diversity before they are lost), (4) crop descriptors (international information in genebanks of accessions as passport info, characterization, and related agronomic and evaluation data), (5) information systems (computerized documentary and information system for the accessions), (6) user priorities (genetic diversity, agricultural necessities, technology[ies], security concerns, genetic erosion, etc.), (7) modern know-how and/or inquiries (*in vitro* storage, cryopreservation, tissue culture, etc.), and (8) challenges to building a robust strategy for rational conservation (planning, capacity, global crop strategies, echoing concern, etc.) (Khoury et al., 2010).

Some of the cons of *ex situ* conservations are as follows:

- (a) Endangered species in the wild or in nature would not recover anymore.
- (b) Although it works well for isolated species, more specialized organisms fail to keep up, are isolated in captivity and do not evolve with their environment.
- (c) Gene pool is too small, and selfing/inbreeding activity is a serious problem.
- (d) The natural habitat might disappear in the wild even if restoring a population is possible.
- (e) Originally, the role of botanical gardens is cost-effective, curative, and appealing.
- (f) Therefore, botanical gardens can only be preferred for these purposes.
- (g) The range of species collected *ex situ* is limited.
- (h) This type of conservation may cause germplasm resources (seeds, pollen, organisms, etc.) to move away from their original habitat or environment.
- (i) Botanical gardens, zoological parks, and aquariums have a crucial role as many plant and animal species are increasingly threatened and have future uncertainty in the wild.
- (j) Genebanks and seed banks, zoological and botanical gardens, etc., are *ex situ* conservation organizations.

- (k) Unfortunately, the evolution there stops since the interaction between the species and the environment fails.
- (l) Hence, both in situ and ex situ preservation types complement each other.

Genebank Operations

They were first established over 50 years ago to conserve threatened crop diversity in local landraces that were being displaced by new improved varieties and destruction of natural habitats. At the level of worldwide, the number of genebanks and the stored seed in them has been rapidly increased due to benefits to the humanity such as their genetic treasures, etc. First of all, getting more storage of PGRs and CWRs require huge storage facilities, handling, computerized characterization, documentation, periodically refreshing, qualified experts, persons... employment necessity is vital. In addition, relevant research and development activities, followed by “seed bank” policy, publication, facilities, material exchange possibilities, food security consequence, etc., are very important. On the other hand, for instance, we estimate that under baseline assumptions, the present value of conserving the existing accessions in perpetuity at CIMMYT is \$7.95 million–\$4.42 million for storing the 17,000 maize accessions and \$3.53 million for the 123,000 wheat samples (Wilkes, 1993; Pardey et al., 1999; Anonymous 2007).

- (i) Seeds can be maintained for long years under controlled conditions (<5% humidity and -20°C).
- (ii) Not all species are right for this handling.
- (iii) Seeds need to be repeatedly germinated to refurbish stock, or they ultimately lose their viability.
- (iv) There are risks such as sudden power cuts, earthquakes, natural disasters, fire, floods, etc. in seed banks.
- (v) Doubling stocks are highly recommended for buffering the conservation.
- (vi) The seeds stored in the seed bank are not affected, to a minimum extent, by changes in environmental conditions.

An extreme ex situ conservation genebank is Svalbard Global Vault:

For the ex situ conservation, an erratic and extreme example is Svalbard Global Vault. This is the world’s the most secure and the biggest genebank on the Norwegian island of Svalbard, about 1300 km from the North Pole. It is maintained by the Global Crop Diversity Trust, the Norwegian government, and the Nordic Genetic Resource Center. The seeds in Svalbard Global Vault are safety duplicates. They are stored free of charge and placed in Svalbard Global Vault on black box terms – only the depositor of the seeds has the ability to withdraw them. Depositors retain ownership rights over the seeds sent to the facility. Officially opened on February 26, 2008, the vault currently holds more than 830,000 samples (accessions including the PGRs) of crop diversity from more than 60 institutions and all over the world, and it has the capacity to conserve 4.5 million seed samples.

Importance of Ex Situ Conservation

The CWRs' ex situ conservation particularly makes them possible to deploy and to use. Without ex situ conservation, the CWRs are extremely difficult or impossible to benefit from using them as exogenous genetic material. Collecting and storing the CWRs in genebanks make them more accessible to breeders and enable their use in agricultural research and breeding while allowing samples to be backed up as safety duplication in multiple locations (Anonymous, 2010). About the 700,000 CWRs accessions are currently held ex situ in genebanks around the world, about 10% of the total holdings. They have held ex situ in national crop diversity collections and international genebanks: CGIAR genebanks, the Millennium Seed Bank, and the Svalbard Global Vault.

5.2.2 In Situ Conservation

Some sources called this type of conservation as “natural reserves/conservation, national parks, wildlife sanctuaries, etc.,” where wild species, forests, and some crusted and crustless fruits are conserved. It allows extremely important natural evolution to continue for the evolution of the genes against biotic and/or abiotic stress (Porceddu et al., 2013; Vernon, 2015). The in situ conservation means “on site/in place/in habitats” conservation and comes with a set of conservation techniques such as designation, management, and monitoring of biodiversity in a given area.

Current in situ conservation methods are the following (Şehirali & Özgen, 1987; Tan, 1998; Özgen et al., 2000; Tan, 2009; Anonymous, 2017):

1. National parks
2. Nature parks
3. Nature conservation
4. Nature monuments
5. Wildlife improvement areas
6. Wildlife breeding stations
7. Protection forest
8. Gene conservation forest
9. Seed stands
10. Special environment protection regions
11. Ramsar areas
12. Natural archeological protection areas
13. Natural assets and gene conservation areas
14. Management areas

5.3 What Are the Advantages (A) and Disadvantages (D) of the In Situ Conservation?

Conservation of species in their site of origination (in situ) has some advantages (A) and disadvantages (D):

- (a) In situ conservation involves the natural system(s) to take care of ecosystem(s) (A).
- (b) It is a very cheap and convenient way of conserving biological diversity (A).
- (c) It ensures the survival of the species besides the protection of the entire natural habitat/the ecosystem (A).
- (d) It has many possibilities to protect a large number of organisms simultaneously known or unknown (A/D).
- (e) It helps the emergence of biotic and abiotic factors in the environment indirectly and induces their interactions with species (A/D).
- (f) Ex situ conservation conserves the sample after it is taken from the target area and helps the target species in the scene to be selected for in situ conservation (A).
- (g) Conservation in spot needs more space than all the other types of conservations and assures a good number of samples (A).
- (h) In situ conservation requires larger areas (D).
- (i) The species in situ that continue to evolve in their natural environments are buffered from environmental changes, and larger populations can be kept (A).
- (j) Wild populations of the CWRs contain a much more genetic diversity than is generally captured in an accession in a genebank, and maintaining viable populations in the wild also allows their evolution, with the potential appearance of new adaptive traits (A; Nevo et al., 2012).
- (k) Similarly, CWRs are the primary target of the in situ conservation and help to conserve several wild relatives of wheat and other cereals (A; Hunter & Heywood, 2011).
- (l) It allows the species not only to sustain their life but also to reproduce and evolve at the same time (A).
- (m) Conservation project aims not only a few selected species but a whole wildlife area (A).
- (n) We are not sure how many species that are endangered by natural habitat or environmental changes would react (D).
- (o) The natural habitat is sufficient for species (D).
- (p) Neither the in situ nor ex situ CWRs conservation is opposite to each other. In situ, ex situ, and on-farm conservation differs from each other based on different criteria (Table 5.1; A/D).
- (q) Ex situ conservation is necessary for the use of CWRs in breeding, but it tends and requires maintaining a greater amount of (genetic) diversity and ensures continued growth and breeding of the species (A/D).

Table 5.1 Regional distribution of genebanks with their cereals and cereal landrace collections (Carvalho et al., 2012)

Geographical region	Number of seed banks maintaining Ancient wheats including landrace (LR) collections	
Africa	23	11
Asia	62	37
Europe	88	44
Latin America and Caribbean	32	9
Oceania	4	2
North America	11	7
Total	220	110

- (r) In the last decade and in most parts of the world, in situ conservation is now more favorable for the CWRs (A).
- (s) During the in situ conservation, it might be necessary to renew the area and control the weeds (A).
- (t) Ex situ conservation stops or interrupts ongoing evolution process (Şehirali & Özgen, 1987) (D) (Table 5.2).

5.4 Which Conservation Type Is Better?

This is a fair but, at the same time, relative question. The answer depends on the type of conservation, germplasm, availability, possibilities, and the environmental conditions. In situ is preferred because it requires less effort and finance. On the other hand, ex situ conservation is useless if species cannot adapt themselves to new environmental conditions. Therefore, an appropriate decision is to be made to consider all the circumstances for the type of conservation desired. To be able to better understand the conservation types for the wild germplasm, we can consult to Table 5.3.

5.4.1 Agrobiodiversity Conservation

Agrobiodiversity is a biological diversity of food and agriculture. Its basic elements make up agricultural ecosystems, all the changes in the organisms, their genetic structure, species and ecosystem levels, and their sustainability. On the other hand, it is an interaction of million-year-old genetic resources and both natural and artificial (human) selection. Agrobiodiversity conservation is clearly essential for mankind survival. A widened genetic variation decreases the risk of climate, harmful diseases – while increases the flexibility ability. Agrobiodiversity is very important for farmers to meet tolerant/resistant/durable cultivar requirements against stress

Table 5.2 Ex situ, in situ, and on-farm conservation of plant and animal genetic resources by different criteria (Hammer et al., 2003)

Criteria	Ex situ	In situ	On farm
Taxa	Species cultivated with the infraspecific taxa; potential wild species	Wild species and their infraspecific taxa; hemerophobes (unadapted to cultivation).	Cultivated species, their taxa and hemerophilous relatives (adapted to cultivation).
Objective	Conservation and investigation	Maintenance.	Management.
Methods	Keeping individuals or minimum populations outside their natural habitat in an artificial environment, mainly without parasites, predators (special case: in vitro)	Caring vital populations and their native, the environment under natural conditions, and natural selection burden (with increasing human influence)	Keeping tame animal and plant populations, shielding whole ecosystems under farming, counting grassland circumstances, and selection by farmers and clients
Limitations	Personal interests; missing collecting strategies and sampling concepts; finances; space for storage and reference collections; scientific capacity; low level of knowledge regarding optimum and bad living conditions, biology, ecology, behavior, etc.; limited access and missing data on traditional preparation, application, or use; genetic shift and drift	Expanding human population with irreversible effect on the native environment including sheltered areas	Reduced quantity of experienced farmers; the reduced biodiversity of plant varieties and animal races, compulsory by modern marketing systems and industry; fast fluctuations in land use and administration
Institutions and people involved	Genebanks, botanical and zoological gardens, special collections, research stations, breeder's collections, amateurs and professionals, a decreasing number of specialists	Protected areas, national parks, biosphere reserves, specialists in research, amateurs and professionals	Farms, gardens, grassland, forests, biosphere reserves including ruderal areas, experts in research and breeding, farmers, hunters, consumers
Experience	500 years	10,000 years	10,000 years unintentionally +
Networking	Global and regional exchange of material and material-related data, internationalization of breeding work; specialization of collections (new)	Target-dependent networks exist or networks are under construction with the aim of harmonizing action of activities	Regional networking, cooperation with ex situ collections should be established; land use within protected areas depends from applied techniques (e.g., in biosphere reserves, see the UNESCO program MAB)
Security	To understand the most secure method for characters and knowledge for the future		

Table 5.3 Maintenance methods and their relative worth for different categories of diversity (Hammer, 2003)

Methods of maintenance			
Category of diversity	Ex situ (genebanks)	On farm (agro-ecosystems)	In situ (other ecosystems)
Intraspecific diversity	C	C** ^b	Co ^c
	W	Wo ^c	W
	WP* ^d	WP** ^b	WP* ^d
Species diversity	C** ^b	C*** ^a	C ^o
	W* ^d	Wo ^c	W
	WP* ^d	WP** ^b	WP* ^d
Ecosystem diversity	C ^o	C** ^b	Co ^c
	W0 ^e	Wo ^c	W
	WP ^{e0}	WP** ^b	WP ^{e0}

* Little meaning α Very important; **^b Important; ^o unimportant; ^ψ; *^d Little meaning
 C= cultivated plants, W= wild related to cultivated plants, WP = weedy plants

induced by climate change, drought, and temperature extremes, etc. Therefore, conservation of the PGRs including the CWRs is of great importance (Ulukan, 2011; Sebastian et al., 2012).

For example, *Aegilops* spp. are very important plants for the cultivated wheats (hulled and naked) and can be given as a leading example for this issue (Table 5.2). At the same time, *Aegilops* spp. have been used as a bridge between cultivated and wild wheats particularly for the disease resistance. From them, leafstripe, and stem rust (*Puccinia* spp.) resistance were successfully transferred to the cultivated wheats with serial hybridization, artificial selections, mutation practices, etc. (Vanderplank, 1963; Zadoks & Bowman, 1985; Stubbs et al., 1986; Roelfs et al., 1992; Ulukan, 2020). There are 137 *Aegilops* spp. populations in Israel, in Russia, and in Turkey, 4000 entries of *Aegilops* sect. *Sitopsis* species, and 5000 entries of ancient *Triticum turgidum* subsp. *dicoccoides* in Israel (Maggioni et al., 2003). The genetic base of the CWRs is narrow, but it possesses valuable genes for resistance to pest and diseases, high protein percentage, etc. (Table 5.4).

5.5 Conservation and Utilization of Wheat

There are three different gene pools, namely, primary, secondary, and tertiary (Ulukan 2020) on earth, and most plants including naked and ancient wheats with their ancestors (the CWRs) originated from them (Figs. 5.3, 5.4 and 5.5). The in situ CWRs conservation community accomplished much more to enlighten politicians and the community. Some of the advanced endorsements might be rapidly and quite cheaply completed. With modest revisions in a few major databases, the CWRs around the world could be substantially located by simply using available

Table 5.4 Classification of domesticated wheats and closely related wilds: the *Aegilops* and *Triticum* spp. (Feldman & Millet, 2001)

Species	Genomes	Wild hulled	Domesticated	
			Hulled	Free threshing
Diploid ($2n = 14$)				
<i>Ae. speltoides</i>	S (= G)	All	–	–
<i>Ae. bicornis</i>	S ^b	All	–	–
<i>Ae. sharonensis</i>	S ¹	All	–	–
<i>Ae. longissima</i>	S ¹	All	–	–
<i>Ae. searsii</i>	S ^s	All	–	–
<i>Ae. tauschii</i>	D	All	–	–
<i>T. urartu</i>	A	All	–	–
<i>T. monococcum</i>	A ^m	spp. <i>aegilopoides</i> (wild einkorn)	spp. <i>monococcum</i> (domesticated einkorn)	
Tetraploid ($2n = 28$)				
<i>T. timopheevii</i>	GA	spp. <i>armeniicum</i>	spp. <i>timopheevii</i>	
<i>T. turgidum</i>	BA	spp. <i>dicoccoides</i> (wild emmer)	spp. <i>dicoccon</i> (domesticated emmer)	spp. <i>parvicoccum</i> * spp. <i>durum</i> spp. <i>turgidum</i> spp. <i>polonicum</i> spp. <i>carthlicum</i>
Hexaploid ($2n = 42$)				
<i>T. zhukovskyi</i>	GAA ^m	–	spp. <i>zhukovskyi</i>	–
<i>T. aestivum</i>	ABD	–	spp. <i>spelta</i>	spp. <i>aestivum</i> (common or bread wheat)
			spp. <i>macha</i>	spp. <i>compactum</i> (club wheat)
			spp. <i>vavilovii</i>	spp. <i>sphareococcum</i>

information effectively. Increasing the official acceptance of selected sites would be harder and requires better planning, training, collaboration, and public relation.

The international agricultural and conservation organizations must lead this worldwide effort. Agro-morphological and molecular characterization and evaluation for resistance/tolerance to biotic and abiotic stresses are coordinated by international programs with the cooperation with national plant breeding programs. The characterization of field crops, forages, fruit trees, medicinal and aromatic plants, vegetables, and even ornamental plants is already carried out. Fortunately, the participatory breeding programs to improve the landraces and the informal seed production systems in some remote and marginal areas are already being given priority in recent years. Material at the national genebanks is freely available for breeding programs and other scientific studies. The exchange of PGRs depends on the seed amount and number of accessions for distribution. With an existing *material*

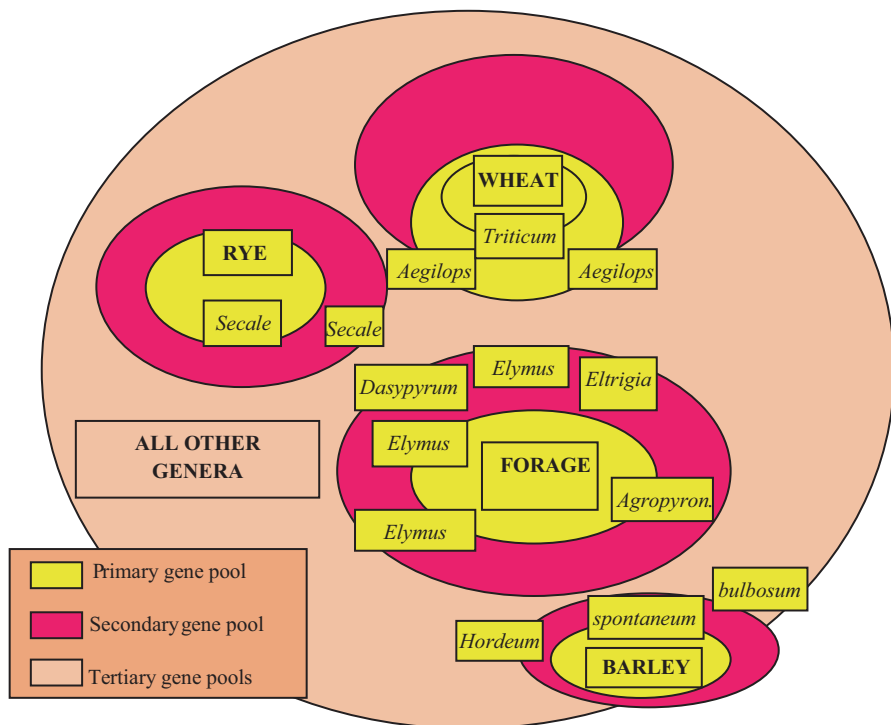


Fig. 5.3 Gene pools in Hordeae. (Formerly Triticeae; modified from Hammer, 2003; Ulukan, 2020)

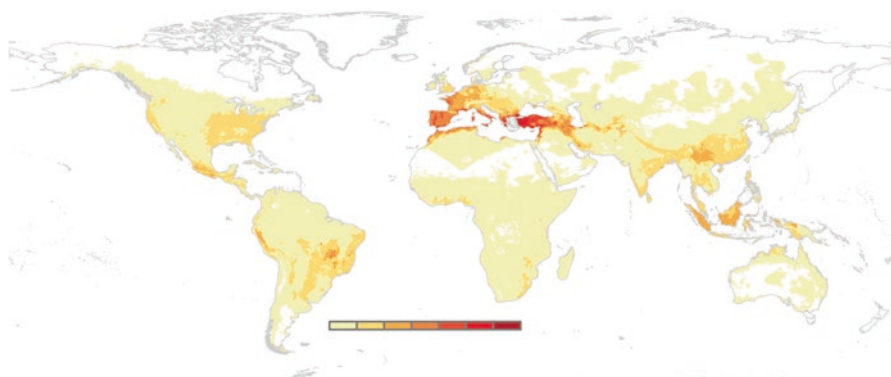


Fig. 5.4 The CWRs richness map. (Source: Castañeda-Álvarez et al., 2016; dark red color indicates denser CWRs distribution)

transfer agreement (MTA) among the institutions, feedback is requested for the accessions, either dispatched from a national genebank or collected jointly. Various improved cultivars either directly released or benefited as genitors (Tan, 1998; 2009).

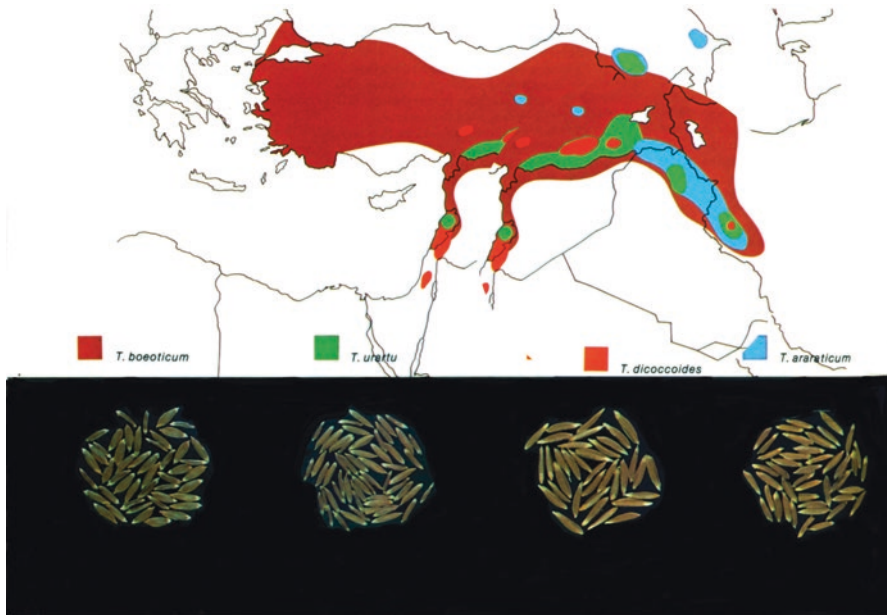


Fig. 5.5 Wild wheats origin and distribution center and their seeds (Johnson & Waines, 1997)

5.6 Crop Wild Relatives (CWRs)

They are crop-related wild plant species (typically in the same genus as the crop species), constitute a valuable genetic resource for crop breeding and climate change adaptation, benefited the improvement cultivars of rice (*Oryza* spp.), sugarcane (*Saccharum* spp.), wheat (*Triticum* spp.), barley (*Hordeum* spp.), tomato (*Lycopersicon* spp.), sunflower (*Helianthus* spp.), and other crops. The CWRs have alone provided traits of \$115 billion in one year, 1997, to global agriculture worth (Anonymous, 1997). This is like wild crop plants, of which approximately 22% are threatened with extinction and face a number of threats according to the 2010 Sampled Red List Index. These threats include changes in land use, increase in many invasive species, the intensification of agriculture, and climate change (Ureta et al., 2011).

Thirty percent of the CWRs taxa were not present in genebanks at all, with a further 24% represented by fewer than ten samples (Castañeda-Álvarez et al., 2016) in the recent study analysis (f. the CWRs can be conserved both ex situ in genebanks, field collections, and botanical gardens and in situ in their natural habitats). Ex situ conservation is essential both to prevent loss of genetic diversity and to facilitate their diversity use in crop breeding. In situ conservation, as a complement, involves the maintenance and recovery of populations in their natural surroundings and ensures continued evolution, including the natural gene exchange with each other and with their cultivated cousins. The CWRs are wild plant species. They

closely relate to cultivated crops, including their wild ancestors – the wild “cousins” of our cultivated plants. The CRWs are an enormously diverse untapped source of raw material for crop improvement. They provide useful genes for pest and disease resistance or tolerance to heat, drought, and other stresses. The common ancestry with crops facilitates the use of their genes in traditional and modern breeding (Ulukan 2011). The CWRs have made significant contributions to modern agricultural production through the contribution of the characteristics to plant cultivars. The CRWs have increased the worldwide productivity of economically important plants, that is, wheat, maize, rice, potatoes, barley, and oats. They have also increased nutritional values. The natural populations of many crop wild relatives are increasingly at risk, mainly due to habitat loss, degradation, and fragmentation. Moreover, the increasing industrialization of agriculture is reducing populations of crop wild relatives in and around farms. Crop wild relatives are often missed by conservation programs, falling between the efforts of agricultural and environmental conservation actions.

5.7 Future Value of Wheat Landraces

The landraces are currently one of the most threatened components of plant diversity in many world countries (Carvalho et al., 2012). Cereal landraces (including the ancient wheats) have emerged in different regions of the world as a result of centuries of crop evolution in traditional agrosystems, especially since the 1950s, they have been increasingly replaced by modern varieties, mainly due to low yield capacity, which offer superior yields but are less adapted to changing environmental conditions, pest, and diseases, etc., and therefore require higher inputs. They are dynamic and essential agrobiodiversity germplasms for the productivity of new crop varieties; however, agricultural biodiversity can make even a far greater contribution (Jaradat, 2012). They are rarely cultivated in developed countries and almost disappeared in developing ones because of their lower yield and higher disease susceptibility potential than modern cultivars under high external input farming systems. However, landraces and old cultivars possess better quality attributes and out-yield/high-yielding cultivars under organic and low-input farming systems. Farmers’ selection for desirable agronomic and quality traits shapes wheat landrace dynamics a lot. Therefore, sustainable utilization of them through in situ or on-farm conservation could ensure their continued evolution and contribution to local food systems.

The future of wheat landraces may depend on: (i) how strongly our food traditions could be linked to the conservation of them, (ii) how increased knowledge of food traditions and improved culinary arts help to create or expand market demand for their products, (iii) how environmental factors could impact quality and culinary attributes of their products, and finally (iv) how the accrued knowledge, along with on-farm conservation, improves livelihoods of individual farmers and rural communities. Additionally, some wheat landraces grow well in various newer environments

in culinary qualities unless the farmers' options are limited. Therefore, non-breeding on-farm conservation would ensure the future wheat landraces.

5.8 On-Farm Dynamic Conservation of the Ancient Wheat and Sustainable Utilization of Wheat Landraces

The scientific concept for on-farm conservation is relatively new and has been offered for the conservation of cereal fields together with their wild relatives allowing them genetic interchange in gene centers (Kuckuck, 1974). The management of the PGRs on farm became the main issue, and in situ and on-farm features of conservation of biological range for food and agriculture have lately been much tenser in the events of International Plant Genetic Resources Institute (IPGRI) later (IPGRs), Rome, Italy. Researchers, governmental decision-makers, and others in developed countries deliberated a lot mainly about rising countries (Engels, 1995).

5.9 Public and Community Seed Saving and Exchange Systems

Unlike their bigger seed and genebank equals, public seed banks are relying less on long-term safeguarding and more on the allocation of seed season to season. They are, therefore, called "seed libraries" as well. No matter what they are called, the core of all community seed banks is the same: they are a principal place where seeds (often locally grown) are stored and shared with local planters. Most deal their seed for free since the philosophy behind public seed banks is that seed is not a product but is a shared public reserve.

Resident seed banks serve as the center of operations for saving the range of local food schemes. These banks have numerous functions:

- Guard and acme the value of inheritances and nearby adjusted seed
- Tutor societies on gardening, seed saving, and breeding for local needs
- Magnify the diversity of people involved in seed stewardship

What are the welfares of starting a seed bank in public?

- Avoiding the loss of genetic diversity and the vanishing of local seed varieties
- Generating adjusted seed to bloom in the soils and climate in a certain region
- Growing access to seed for all, but particularly for the first-time, low-income, and public gardeners
- Educating the public on how to save seeds and linking them to the shared history of seed as a public resource
- Firming the local food structure and growing food security, beginning with seeds
- Hovering consciousness and activating movements around seed issues such as patenting and genetic manufacturing

5.10 Farmer's Evaluation and Conservation of Crop Genetic Resources

Farmer's evaluation and conservation of crop/plant genetic resources are directly related with/depend on growing knowledge, agrobiodiversity, ecology/ecosystem, and conservation method. Agricultural biodiversity or agrobiodiversity is the basis of this topic. Therefore, many researchers, institutes (national/international), etc., are occupied with these activities, which resulted in many lists for plant genetic resources. Among the PGRs, the CWRs have high priority in any wheat breeding programs for agrobiodiversity and biodiversity. The lists prepared are very useful for providing/generating massive data sets (such as the IUCN Red List of Threatened Plants) of persons who collected, conserved, and evaluated (Maxted & Hawkes, 1997; Hawtin & Hodgkin, 1997). Directed research is, however, needed to discover the many unsolved problems to identify/describe and manage the CWRs best on this topic. In situ, the CWRs conservation training programs, particularly for the staff of the protected areas, are likewise needed, and luckily, the theoretical and methodological base and the protected sites exist today. Easy and pleasant in situ CWRs organization rules for the staff of the protected areas and other workers to consult are needed. The creation of the protected areas and their management for the CWRs require the presence and the value of CWRs within the existing and proposed protected areas. The importance of in situ CWRs conservation should be publicized and involve ordinary people inappropriate activities. Educational, agrobiodiversity, forestry, and biodiversity preservation organizations must more effectively coordinate the common objectives of the CWRs conservation. The in situ CWRs conservation efforts reported here have been increasingly collaborative, but trying to broaden and deepen cooperation must still remain a priority. Country-level agricultural and environmental protection administrations must try to overcome pillow wars, while they deal with the advancement of the CWRs conservation. International organizations have strategic, operational, and backing policies for mounting and enabling the in situ for the CWRs safeguarding.

5.11 Seed Genebanks in Turkey

As known, Turkey is in the subtropic zone between 36°–42° N and 26°–45° L. It has a total area of 77,945,000 ha, which is divided by the Dardanelles, the Sea of Marmara, and the Bosphorus in the west and is surrounded by the Black Sea in the north, The Aegean Sea in the west, and the Mediterranean Sea in the south. Turkey is the gene center of many cultivated and wild (the CWRs and the PGRs) crops (particularly wild and cultivated naked and ancient wheats, rye, chickpea, oat, and barley) and the domestication as well (Harlan, 1950, 1995; Vavilov, 1951; Sears, 1954; Harlan et al., 1973; Hawkes, 1983; Davis, 1985; Zohary & Hopf, 1988; Anonymous, 1993; Kaya et al., 1997; Kaya, 1998; Bennett et al., 1998; Tan, 1998; Tan & Tan, 1998; Zohary, 2004; Gepts, 2006; MacDonald et al., 2015; Khoury et al., 2016).

In Turkey, genetic resources are conserved mainly with the ex situ conservation method in genebanks. But Aegean Agricultural Research Institute (AARI) has ex situ and in situ conservation methods with related facilities for the PGRs and the CRWs. There are more than 22,000 Turkish wheat landraces in ex situ collections worldwide (Morgounov et al., 2016). But we still have no information for the ancient wheats that were stored in the AARI at the level of genera and species bases.

The AARI or the Aegean Agricultural Research Institute maintains a collection of 3216 accessions of cultivated *Triticum* species. In 1948 to 1949, Prof. Dr. J. Harlan collected 2128 wheat accessions from each province in Turkey (Harlan, 1950). In 1990, Prof. Dr. C. Qualset and his colleagues initiated research on wheat genetic diversity at the farm, village, and regional level in Turkey to assess the extent and the impact of changing social and agricultural conditions on wheat diversity. The research was in three provinces of the western transitional zone, which is located among the Taurus Mountains, located between the Aegean and Central Anatolian regions, in order to study the changes in genetic erosion. The transitional zone appeared to retain old farming methods higher than the intensively cultivated (irrigated) coastal zone or the Anatolian plateau (Aresvik, 1975).

In Turkey, seed genetic conservation is practiced by national and well-equipped genebanks in İzmir (the AARI) and in Ankara (CRIFC). It was established in 1972 to protect the PGRs. Historically, the first one was initiated by the Republic of Ministry of Food, Agriculture, and Livestock (MFAL) in 1930, and the second one is established and renamed after Prof. Dr. Osman TOSUN at the Department of Field Crops, Faculty of Agriculture, University of Ankara in 1933. At the first one, a total of 50,000 cereal and leguminous seed samples collected by Yeşilköy Agricultural Research Institute up to 1960 years were conserved. The AARI's seed collection facilities meet the requirements of long- and medium-term storage operations, while it could work on these seeds as well. The AARI seed facilities also meet the international standards (Tan, 1998; 2009).

Ex situ conservation has been started in 1964 at the AARI for **basic/base/core collection** (long time) and short and middle collections. Seeds collected are local or released varieties, breeding lines with good agronomic traits, wild relatives of the cultivated plants, wild species, and the transition forms. The first stage in the studies was to determine the phytogeographical and agroecological distribution of plant species and to collect the most distinct plant samples. Collection is being applied following the prepared annual programs. These programs are carried out in groups: cereals, fodder plants, pulses, fruits and vegetables, industrial plants, ornamental plants, aromatic and medicinal plants, and vine plants. Endemic species are also collected.

Basic collections are stored as two sets in the AARI at $-18/-20$ °C for a long time, and active collections at the 0 °C. Today, there are more than 55,000 samples and more than 2700 species in the AARI. Of those materials, about 20,000 belong to 2221 wild species. Conservation facilities at the AARI are also available to keep the safety duplicates of national collections up to 8405 accessions in field crop species. Total accessions are, in other words, about 63,000 at both genebanks. There are also up to 7000 vegetative propagated the PGRs, mainly fruits.

National field genebanks are mainly at different institutes including the AARI in Turkey. The field genebank collections of vegetative propagated species consist of over 100 species (Tables 5.5 and 5.6). The main users of the conserved materials are the plant breeders, researchers, scientists, institute persons, and academicians at the level of national and international. The security backups of some proportion of base collection of the AARI have recently been started to be backed up in the national seed genebank in Ankara at the Field Crops Central Research Institute (CRIFC) in Turkey, which was opened on the date of February 3, 2010. Its capacity is 250,000 seeds, which comes after the one in China (300,000 seeds) and the one in the United States (280,000 seeds in 2016).

There were 63,756 species from the 3650 plant and 120,995 cultivar seed and samples conserved there. To mention, the samples gathered from all over Turkey during 1960–1972, under the “*new culture plant project*” are under protection. The following are ex situ conserved for short and long term: 3700 wheat (*Triticum* spp.), 3000 barley (*Hordeum* spp.), 1600 chickpeas (*Cicer* spp.), 500 lentils (*Lens* spp.), and 1600 other plant species. This arrangement continues to work on the classification, documentation, and preservation of cereals, legumes, forage plants, vegetables, and oranges and ornamental, medical, and aromatic, and scented plants.

Tissue culture is the process of producing new tissue, plant, or plant products totally under the controlled aseptic conditions from plant parts (explants) such as cells, tissues, or organs in an artificial nutrient environment. The tissue culture studies were first started as micro-studies in universities and agricultural research institutes. Of these laboratory techniques, there are various preservation methods. Cryopreservation, for example, means the conservation of vegetative plant cells and tissues at ultra-low temperatures such as $-196\text{ }^{\circ}\text{C}$ by liquid nitrogenous. DNA banks, as another method, consist of DNA isolation from the material as the first step toward this protection. DNA is, then, stored as total genomic DNA and/or chloroplast DNA. The isolated DNA is protected at low temperatures ($-20\text{ }^{\circ}\text{C}$ or $-80\text{ }^{\circ}\text{C}$) depending on the use.

Table 5.5 Number of species and number of accessions at national genebank at the AARI in İzmir (Tan 1998; 2009)

Plant groups	Number of species	Number of accessions
Cereals	169	15,806
Industrial crops	58	5703
Vegetables	92	7753
Ornamental plants	127	2301
Forages and fodder plants	398	8683
Food legumes	34	7443
Medicinal and aromatic plants	198	2616
Endemic species	874	4297
Other species (incl. wilds utilized as vegetables)	735	924
Total	2685	54,523

Table 5.6 Plant species supplied from the national genebank at the AARI, in İzmir in 1998–2008 (Tan 1998; 2009)

Plant species	Total no. of accessions	To foreign users	To domestic users
<i>Agropyron</i> spp.	51		51
<i>Allium</i> spp.	56		56
<i>Astragalus</i> spp.	6		6
<i>Avena</i> spp.	147	4	143
<i>Beta</i> spp.	139	6	133
<i>Brassica</i> spp.	31	8	23
<i>Bromus inermis</i>	17		17
<i>Cannabis sativa</i>	38		38
<i>Capsicum</i> spp.	472	6	466
<i>Carthamus tinctorius</i>	52		52
<i>Cicer</i> spp.	606	5	601
<i>Citrullus vulgaris</i>	96		96
<i>Cucumis flexuosus</i>	26		26
<i>Cucumis melo</i>	332	8	324
<i>Cucurbita</i> spp.	27		27
<i>Cuminum cyminum</i>	14		14
<i>Dactylis glomerata</i>	74		74
<i>Daucus</i> spp.	80	9	71
<i>Dianthus</i> spp.	63		63
<i>Festuca</i> spp.	13		13
<i>Glycine max</i>	15		15
<i>Gypsophila</i> spp.	17		17
<i>Helianthus annuus</i>	290	4	286
<i>Hibiscus esculentus</i>	53		53
<i>Hordeum</i> spp.	492	11	481
<i>Isatis</i> spp.	9		9
<i>Lathyrus sativus</i>	13		13
<i>Lens</i> spp.	48		48
<i>Linum usitatissimum</i>	33		33
<i>Lolium</i> spp.	45		45
<i>Lotus</i> spp.	14		14
<i>Lycopersicon esculentum</i>	472	9	463
<i>Medicago</i> spp.	102		102
<i>Nicotiana tabacum</i>	123	2	123
<i>Onobrychis</i> spp.	9		9
<i>Origanum</i> spp.	123		121
<i>Petroselinum hortense</i>	18		18
<i>Phaseolus vulgaris</i>	718	7	711
<i>Pisum sativum</i>	92		92
<i>Salvia</i> spp.	123		123
<i>Sesamum indicum</i>	201	4	197
<i>Solanum melongena</i>	341	8	333

(continued)

Table 5.6 (continued)

Plant species	Total no. of accessions	To foreign users	To domestic users
<i>Spinacia oleracea</i>	37		37
<i>Thymus</i> spp.	9		9
<i>Trifolium</i> spp.	8		8
<i>Triticum</i> + <i>Aegilops</i>	3436	10	3426
<i>Vicia</i> spp.	690	12	678
<i>Vicia faba</i>	249	6	243
<i>Zea mays</i>	1108		1108
Total	11,228	119	11,109

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Chapter 6

Chemical Composition of Einkorn (*Triticum monococcum* ssp. *monococcum*), Emmer (*Triticum dicoccum*), and Spelt (*Triticum spelta*)



Yunus Şahin and Fatma Pehlivan Karakas

6.1 Introduction

Wheat (*Triticum* spp.), which is cultivated for centuries in the Middle East, Central Asia, Europe, North Africa, and, then, in America, is a leading staple crop around the world. Its marginally grown ancestor, einkorn (*Triticum monococcum* ssp. *monococcum*), possesses desired agronomic and quality characters and, moreover, bioactive compounds, which reduce and prevent chronic diseases such as diabetes, cancer, Alzheimer, and cardiovascular diseases. As more attention has been given to wheat cultivars with strong gluten, protein content, starch composition, and resistance to biotic and abiotic stresses in bread wheat and yellow-colored pasta products in durum wheat health compounds such as fibers, phytochemicals, and bioactive compounds have, however, been undervalued so far. Thus, demand for functional foods with desirable protective substances steadily is increasing in order to combat malnutrition; thus, in this case we discuss the chemical composition of ancient wheat species in detail in the present chapter.

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6.2 Wheat Grain Morphology

All cereal species taxonomically belong to the *Gramineae* (*Poaceae*), one of the largest Angiosperm families. In this family, the seeds cannot easily separate from the fruit tissue because the pericarp and the seed coat are tightly united. Hence, this peculiar type of infructescence is called caryopsis or, colloquially, kernel. Wheat grains range mainly between 5 mm and 9 mm in length. Moreover, their shapes vary from spherical to long, narrow, or flattened.

Wheat grain is composed of three main parts: germ, endosperm, and bran. The wheat germ is the embryo that would eventually develop into the wheat plant. Endosperm part is the storage region mainly for starch, contributing in feeding in the germination phase of the grain. Bran part encloses the grain and is composed of several layers. Phytochemicals, proteins, carbohydrates, cellulose, and other molecular compounds are unevenly distributed among the layers and subsections of grain (Fig. 6.1).

The bran is divided into compact cell layers and a pigment strand: aluerone layer, hyaline layer, testa, inner and outer layer, and pericarp. These layers are rich in soluble and insoluble dietary fiber (xylans, glucans, raffinose, stachyose, fructans), proteins (sulfur amino acids and glutathione), phenolic acids, carotenoids, lignans, anthocyanins, isoflavonoids, vitamin E, B vitamins (thiamine, riboflavin, and

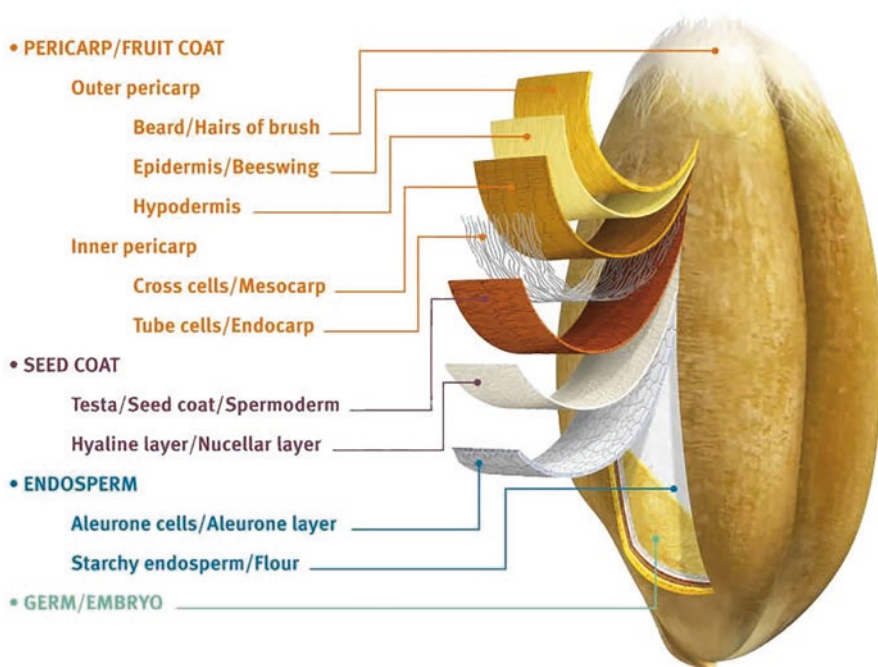


Fig. 6.1 Morphological structure of wheat grain

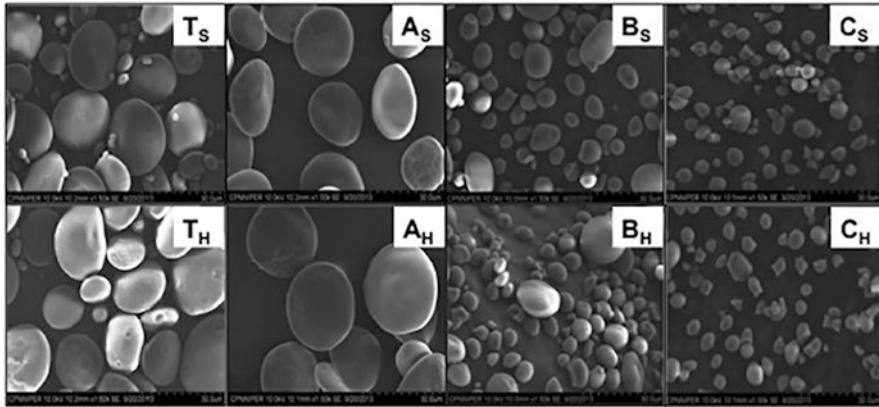


Fig 6.2 Starch granules

pyridoxine) (Batifoulier et al., 2006; Karakas et al., 2021), minerals (K, P, S, Mg, Ca, Zn, Fe, Mn, Cu, Na, Al, Ba, Sr, B, Rb, and Mo) (Ozkan et al., 2007; Suchowilska et al., 2012), and phytic acid, betaine, choline, and enzymes.

The endosperm is composed of large, thin-walled cells, filled mainly with starch and proteins. In the sub-aleurone area, especially on the dorsal side of the grain, the dominant cells are elongated in the direction of the endosperm center containing 70–75% of proteins. Starch grains are enclosed between thin layers of adherent proteins and within a protein matrix which fills the individual cells of the endosperm at varying degrees (Fig 6.2).

The germ lies at one end of the grain. It is rich in proteins (25%) and lipids (8–13%). The mineral level is also higher (4.5%) than that of bran and endosperm part. Wheat germ is available as a separate entity. It is an important source of vitamin E, which has only one half the glutamine and proline of flour, but the levels of alanine, arginine, asparagine, glycine, lysine, and threonine are twice as much as the levels in flour.

6.3 Carbohydrates

Carbohydrates are macromolecules that are found in certain foods. Sugars, starches, and fiber are carbohydrates. Starch makes up the most abundant fractions up to about 65–75% of the wheat grain. Starch presenting as granules is the main constituent of wheat grain and flour and made up of amylose and amylopectin (Fig. 6.3).

The linear amylose, composed of α -(1-4) bound glucose molecules and the highly branched amylopectin, with a backbone linear structure of glucose molecules is linked with α (1-4) glycoside bonds and branching side-chains every 24 to 30 glucose units linked to the backbone by α (1-6) bonds. During the digestion, the starch is mainly degraded by two enzymes (alpha- and beta-amylase): the amylose

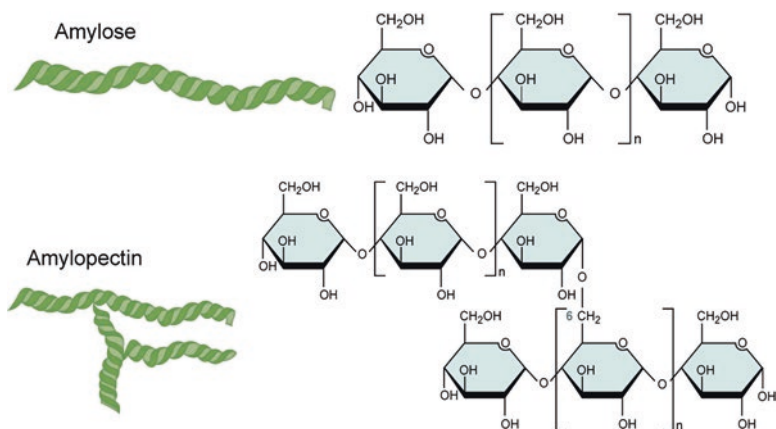


Fig. 6.3 Molecular structures of amylose and amylopectin

is completely hydrolyzed to maltose, while the amylopectin gives maltose (about 60%) and dextrin (about 40%). Amylose accounts for only around one-quarter of the starch granule with the frequency of its molecules greater than that of amylopectin ones. It is the reason for the much smaller size of amylose compared to amylopectin. Nonetheless, its higher resistance to hydrolytic enzymes is due to being more tightly packed, thus causing the low postprandial levels of glycemic and insulinemic responses which, in turn, rise to reduce post meal blood insulin and glucose, yielding longer satiety (Giacintucci et al., 2014).

Starch content of ancient wheat species is lower than modern ones. It is mainly attributed to the selection of high starch concentrated modern cultivars by breeding programs and/or by needs and demands by farmers. Starch molecules are arranged in semi-crystalline granules of different dimensions. *Triticum* ssp. granules have a typical bimodal size distribution as larger granules (A-type, with a 12–24 μm diameter) and smaller granules (B-type, with a 5 μm diameter) (Fig. 6.2). The frequency of A-granules is less than that of B-granules. On the other hand, einkorn does not show the same bimodal shape; even those of einkorn (A-type granules) are smaller than those of common wheat. Rapid hydrolysis of both starch granules plays an important role in determination of lower postprandial glycemic and insulinemic levels. The fraction which resists digestion and absorption in the human small intestine is defined as “resistant starch.” The aspect of resistant starch content in einkorn is poor (Asp, 1994; Brandolini et al., 2011; Lindeboom et al., 2004; Stoddard, 1999). In emmer, two typical distinct populations of starch granule sizes were observed (Giacintucci et al., 2014). The larger granules were lenticular shaped, and the dimensions of these larger granules manifested as wide dispersion, while the smaller granules had a spherical appearance and were more uniform in size. Emmer wheat kernels and flour appeared to have more similarities with the common wheat. Spelt contains approximately eight to ten fold the amount of resistant starch as compared to common wheat (Abdel-Aal and Rabalski, 2008a, b).

6.4 Proteins

Proteins are synthesized from common 20 amino acids. Amino acids that covalently bound each other produce polypeptides. Since each of amino acids has distinctive chemical properties, they contribute to enormous variability of each protein in their functions. Cells can produce proteins with different combinations by joining 20 amino acids. Those combinations lead to widely diverse products as enzymes, hormones, antibodies, transporters, and other substances having distinct biological activities. Wheat stores a bunch of proteins in its seed. They are used in growth and development as source of amino acid when seed starts to germinate. Wheat seed proteins are the topics of this chapter.

6.4.1 Seed Storage Proteins

In wheat grain, storage proteins can be classified as gluten and non-gluten proteins. Gluten protein is subdivided into two fractions: glutenin and gliadin. The glutenins are comprised of aggregated proteins, therefore known as glutenin macropolymers. Their complex polymeric structure increases the strength and the elasticity of dough, whereas the gliadins are monomeric proteins responsible for dough viscosity in wheat meal flour valuable for bread and pasta making. Protein quality depends on amino acid contents, especially the proportion of indispensable amino acids. In addition, the quality also depends on digestibility and rate of physiological utilization after absorption. Wheat storage proteins are low in some essential amino acids, scarce in lysine, but higher in proline and glutamic acid. The variation of amino acid composition was demonstrated in several *T. monococcum* ssp. *monococcum* accessions. Acquistucci et al. (1995) found that variation in its amino acid composition was relatively low, except for tyrosine, arginine, and proline.

The unique properties of hexaploid common wheat are primarily related to its gluten-forming storage proteins. Since the gluten network is mainly responsible for dough extensibility and elasticity, understanding the role of storage protein fractions on bread texture is crucial for technological applications. It is quite documented that the strength and elastic properties of dough are primarily imparted by glutenin proteins, whereas gliadin proteins have been indicated to play a role in determining dough extensibility. Polymeric glutenin proteins, with molecular masses ranging from approximately 300 kDa to one million kDa, can be further classified into two subunit groups: low molecular weight (LMW-GS) glutenin subunits and high molecular weight glutenin subunits (HMW-GS). Low molecular weight glutenin subunits are similar in size and structure to γ -gliadin (30–40 kDa). LMW-GS subunits are biochemically classified into B, C, and D types (Fig. 6.4).

B-type subunits are named according to their first amino acid residue: m=methionine, s=serine, and i=isoleucine (LMW-m, LMW-s, and LMW-i types). On the basis of LMW-GS's ability to form different numbers of intermolecular

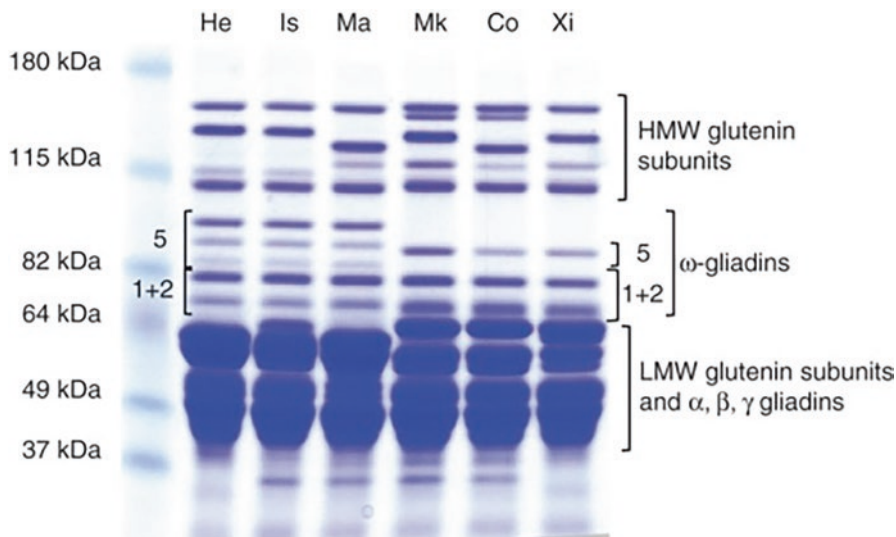


Fig. 6.4 SDS page of wheat glutens. Upper part and lower part indicate glutenin subunits

disulfide bonds, LMW-GS can be classified as chain extenders or chain terminators. *i*-type subunits are linked by interchain disulfide bonds, which may subsequently extend to produce glutenin polymers. Chain terminators are characterized by gliadin-like LMW-GS (i.e., α -, γ -, ω -gliadin) and block subunits from becoming extended polymer chains due to the lack of additional free cysteine molecules necessary for interchain linkages. High molecular weight glutenin subunits (HMW-GS) range in molecular mass from ~65 to 90 kDa. Each wheat variety contains three to five HMW-GS that can be further grouped into two different types: *x*- and *y*-type. *x*-type HMW-GS subunits are characterized by molecular weights that range from 83000 to 88000, while the molecular weights of *y*-type HMW-GS subunits range from 67000 to 74000 Da. While all hexaploid wheats contain 1Bx, 1Dx, and 1Dy subunits (1Bx, 1Dx, and 1Dy indicate the locations of *x*- or *y*-type subunits in A, B, and D chromosomes of wheat), only some cultivars contain 1By and 1Ax, additionally. The composition of HMW-GS alone may account for up to 60% of variation observed in the quality of bread flour. HMW-Dx5, HMW-Bx17, HMW-Dy3, HMW-Dy10, HMW-By15, LMW-m, LMW-s, and LMW-i are commonly found subunits in wheat starch. The most influential subunits of glutenin on wheat quality are Dy10, Dy3, and Dx5, while the most effective gliadin subfraction is α -/ β -gliadin (Wieser & Kieffer, 2001).

Wheat storage proteins include many active forms such as bioactive peptides. For example, DPP-IV is an enzyme that cleaves peptides at position 2 of the N terminus of a peptide sequence containing either alanine or proline residue. There are two substrates for DPP-IV in humans: GLP-1 and glucose-dependent insulinotropic peptide (GIP). These two important incretin hormones enhance meal-stimulated insulin secretion from pancreatic β -cells, but are inactivated by DPP-4; GLP-1 and

GIP are natural therapeutic agents in managing glucose levels in blood, thus effective in treating type-2 diabetes. Due to its rapid breakdown in the body by DPP-IV, GLP-1 cannot survive long enough (Thornberry & Gallwitz, 2009). LMW glutenin is high in DPP-IV and celiac toxic peptides. Moreover, LMW glutenin includes a small number of opioids which acts as analgesics or as antioxidant peptides. Angiotensin-converting enzyme is a central component of the renin-angiotensin system (RAS), which controls blood pressure by regulating the volume of fluids in the body. ACE inhibition is a serious process since the ACE lead to enhance angiotension II which increase blood pressure. In wheat storage protein, some ACE inhibitor peptides have been identified. HMW glutenin is especially high in ACE inhibitor peptides, DPP-IV inhibitor, and antioxidant peptides. HMW glutenin also carries some anticancer peptide sequences. Alpha-gliadin contains high amounts of ACE inhibitors, as well as DPP-IV and PEP inhibitors. Both gamma- and omega-gliadin have similar occurrence frequencies of ACE inhibitor, DPP-IV inhibitor, and antioxidant peptides, though omega-gliadin is higher in celiac toxic peptides, while gamma-gliadin contains some hypotensive rennin inhibitor (Cavazos & Gonzalez de Mejia, 2013).

Considering the protein content of grain fraction in einkorn, germ has 2–3 times more than that of endosperm and bran. Although protein content of einkorn is superior to bread wheat, their amino acid compositions are almost similar with that of bread wheat. In addition, protein content of bread wheat is lower than that of durum wheat and emmer, and there is no diversity between that of bread and spelt wheat.

6.5 Lipids

6.5.1 Dietary Fiber and β -Glucans

Dietary fibers are indigestible compounds, derived from plants. They are classified as soluble fiber which dissolves in water and insoluble fiber which solidify in water; the dietary fiber includes all nondigestible carbohydrates, that is, non-starch polysaccharides, resistant starch, resistant oligosaccharides, and other nondigestible: lignin, cellulose, and fractions of hemicellulose. These components vary in their distribution among the different grain portions. Outer pericarp is rich in lignin, arabinoxylan, and cellulose. The aleuron cells are also rich in fiber, which comprises cellulose, arabinoxylan, β -glucan, and glucomannan. By contrast, the starchy endosperm cells contain only the major components: β -glucan, cellulose, glucomannan, and arabinoxylan. In addition to cell wall polysaccharides, fructans are other carbohydrates which contribute to its dietary fiber contents of wheat (Andersson et al., 2013; Barron et al., 2007).

Total dietary fiber content, lignin, and β -glucan have been quantified as a part of HEALTGRAIN Project, including five lines of each einkorn, emmer, and spelt (Gebruers et al., 2008). Diversity in dietary fiber content is lower due to limited

number of species subjected to the research, especially in einkorn and emmer. That of einkorn is lower (less than 100 g/kg) than others; in addition similar results were obtained for other ancient wheats (emmer, 98 g/kg dry matter; spelt, 120 g/kg dry matter) (Andersson et al., 2013).

These differences in diversity and distributions of dietary fiber components are attributed to smaller grains which may have higher concentrations of dietary fiber components (and other components concentrated in the bran) than larger grains, due to having a higher ratio of bran to flour. Since grain size is determined mainly by starch, it may lead to “yield dilution” of other components.

6.6 Microelements

6.6.1 Ancient Wheat for Combating Microelement Malnutrition

Mineral nutrients play a major role in numerous processes of biological systems, included in many biochemical and physiological functions and metabolisms. These essential elements are divided into two groups as those obtained from water and carbon dioxide and those obtained from the soil. Hydrogen, oxygen, and carbon are obtained from water and carbon dioxide, though the rest of the essential elements are brought from the soil. Essential elements obtained from soil are subdivided into two groups as micro- and macroelements. Nitrogen, potassium, calcium, magnesium, phosphorus, sulfur, and silicon are macroelements, though chlorine, iron, boron, manganese, sodium, zinc, copper, nickel, and molybdenum are microelements. The concentration of micro- and macroelements studied in several ancient wheat species and bread wheat were presented in Tables 6.1 and 6.2.

Higher plants can obtain these minerals from soil for their metabolic processes; however, animals depend mostly on higher plants to stream their biological system with mineral nutrients. Micro- and macroelements are classified according to their usage density in the metabolism. Deficiencies of these elements in biological systems might cause a set of problems, including death. Micronutrient malnutrition affects more than three billion people (Welch & Graham, 2004). The most prevalent deficiencies of micronutrients are Fe, Zn, and I. It has been estimated that each Fe

Table 6.1 K, P, S, Mg, and Ca is expressed as g/kg, and the rest of the values are in mg/kg

	K	P	S	Mg	Ca	Zn	Fe	Mn	Cu	References
<i>T. boeoticum</i>	–	–	–	–	–	89	79	–	–	Cakmak et al. (2000)
<i>T. monococcum</i>	4.3	5.2	1.9	1.6	0.4	55	47	54	6.4	Ozkan et al. (2007) and Suchowilska et al. (2012)
<i>T. dicoccum</i>	4.4	5.1	1.9	1.7	0.4	54	49	24	4.1	Suchowilska et al. (2012)
<i>T. spelta</i>	4.2	4.7	1.8	1.5	0.4	47	50	27	5.0	Suchowilska et al. (2012)
<i>T. aestivum</i>	5.0	4.2	1.4	1.4	0.4	35	37.5	26	3.9	Suchowilska et al. (2012)

Table 6.2 Other microelements

	Na	Al	Ba	Sr	B	Rb	Mo	References
<i>T. monococcum</i>	7	2.5	2.6	5.4	0.8	0.8	1.2	Ozkan et al. (2007)
<i>T. dicoccum</i>	12	3.8	2	2.6	0.6	0.8	1.0	Suchowilska et al. (2012)
<i>T. spelta</i>	10	4.4	3.5	3.6	0.7	1.1	0.7	Suchowilska et al. (2012)

and Zn deficiency affects about one-third of the world's population (International Zinc Nutrition Consultative Group (IZiNCG) et al., 2004). Selenium deficiency is another common one. Globally, between 0.5 and 1 billion people may have inadequate intakes of Se and, therefore, higher deficiency incidence of Se in the developed countries.

Einkorn, wild emmer (*T. dicoccoides*), emmer, and spelt were examined for their micronutrient content to date and suggested a wide variation in their micronutrient concentration (Suchowilska et al., 2012). A clear micronutrient variation was evidently reported by Ozkan et al. (2007) in the seed of 54 einkorn accessions. Zinc in the seeds varied from 0.21 to 2.16 µg/seed, with an average of 1.21 µg/seed, and Fe content varied from 0.54 to 3.09 µg/seed, with an average of 1.27 µg/seed. A similar variation in Mn and a lower variation in Cu were determined. In addition, grain mineral nutrient concentrations in *T. dicoccoides* under five environments (Gomez-Becerra et al., 2010) varied among the genotypes. Given the macronutrients, Ca concentration had the largest variation in the grains, whereas for genotype-dependent P, K, S and Mg, variations were lower. Among the micronutrients, the largest variation was in Mn concentration. Grain Fe and Zn concentrations also showed significant variation. Cultivated spelt accessions at two different environmental sites showed great genotypic variation for micronutrients. Iron (19–72 mg/kg) and Zn (29–102 mg/kg) had the largest variation (Suchowilska et al., 2012). Moreover, the genotypes having high concentrations of mineral nutrients had also larger seed size or seed weight resulting from the greater amount of minerals/seed. The grain of all ancient wheats contained significantly more Zn (from 34% to 54%), Fe (from 31% to 33%), and Cu (from 3% to 28%) than other wheat species and cultivars (Suchowilska et al., 2012).

When all these findings and evidence are considered, ancient wheat species (einkorn, emmer, and spelt) present higher genetic variability. These can be used to improve the microelement content of modern wheat species or cultivars and offer opportunity to develop more functional foods.

6.7 Phytochemicals

Phytochemicals are secondary metabolites which are produced by plants. Even though the exact mechanisms they involved are not clearly elucidated, some roles have been investigated in the processes of metabolic mechanism such as color change, flavoring, UV protection, antimicrobial defense, antioxidant, and natural

pesticide properties in the metabolic processes of plants. Phytochemicals are not considered as nutrient, especially because they are not essential for human/animal growth and development. However, they are called as phytonutrients in the literature since many of these substances exert protective effects or physiological functions in mammals. Nevertheless, they may preserve health throughout life, including prevention of chronic diseases such as some cancers, diabetes, and cardiovascular diseases (Dembinska-Kiec et al., 2008; Seal & Brownlee, 2015; Zhang et al., 2015).

Phytochemicals are classified according to the arrangement and the number of carbon atoms and characterized by one aromatic ring and at least one hydroxyl groups. They are originated from carbohydrates that shikimate or phenyl-propanoid pathways are involved in biosynthetic process. They are divided into three main divisions as phenols, terpenes, and alkaloids. Depending on the type of plant family, species, and/or diverse tissues (also organs), they show uneven distribution and production rates. Shikimate pathway is responsible for the biosynthesis of three amino acids: phenylalanine, tyrosine, and tryptophan. Biosynthesis of numerous phytochemicals originates from tyrosine and tryptophan end products of Shikimate pathway. On the other hand, phenylalanine and tyrosine products are used for biosynthesis of phenylpropanoids. Phenylpropanoids are common throughout the plant kingdom, and they serve as essential components of a number of structural polymers (Fraser & Chapple, 2011).

The function of these molecules is still debated, scientist continuously investigate and thus discover their functions and effects in the plant metabolism.

6.8 Phenols

Phenols comprise a large group of molecules among secondary plant metabolites. They offer essential roles in the reproduction and the growth of plants (i.e., acting as defense mechanisms against pathogens, parasites, and predators, as well as contributing to color formation). In addition to their roles in plants, phenolic compounds play a role in human diet. They are classified as aromatic alcohols since the benzene ring is attached with a hydroxyl group. In the structure of simple phenols, hydrogen is replaced by a hydroxyl group on the aromatic ring. They are categorized as phenolic acids, flavonoids, stilbenes, coumarins, and tannins. Figure 6.5 represents some basic structure for the major phenols.

Composition of phenolic compounds synthesized by plants can vary depending on the species. In cereal grains, even though the yields may vary, specific phenol compounds such as ferulic, *p*-coumaric, and caffeic acids are mainly found.

Phenols are the major class of bioactive compounds present in wheat grain. A number of benefits of secondary metabolites with the widest variety derivatives make the key component of cereals present in grains. Until now, quality parameters, colors, yield, and starch and protein content of cereal grains have been taken into consideration by plant breeders, but little attention has been given to the secondary

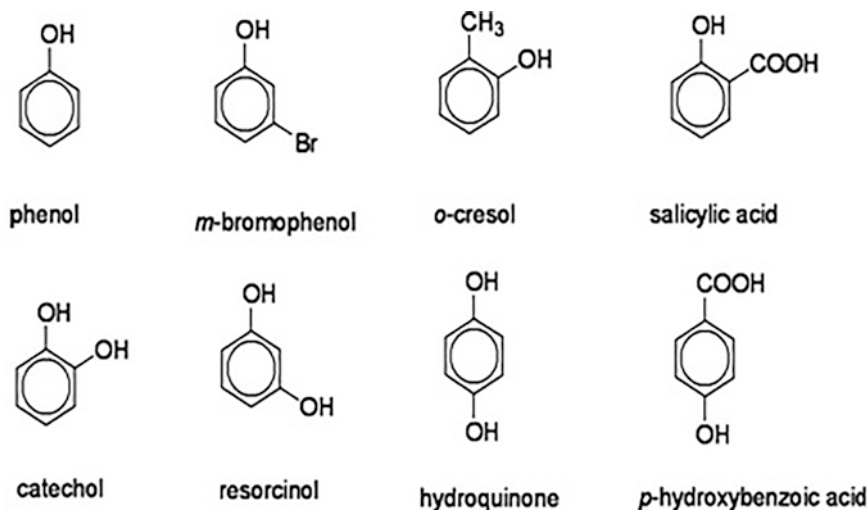


Fig. 6.5 Molecular structures of the major phenolic compounds

metabolites, especially bioactive compounds. Thanks to today's advanced techniques (i.e., MS, HPLC, GS, etc.), more accurate data can be collected in terms of phenolic content quantification and quantitation. For example, the values of total phenolic content quantified by HPLC have become more reliable than quantified by Folin-Ciocalteu method.

Phenols can be detected in all tissues of wheat grain, including embryo. In their study, Adom and his colleagues (2003) suggested that phenols present in wheat grains are in three forms as soluble (free), conjugated soluble, and insoluble.

They exposed the grounded grains of selected bread and durum wheat variety samples to alkaline environment, thus breaking down the ester bounds between cell components and phenolic acids, and, hence, released the insoluble phenolic acids. Their study showed that the past studies had been carried out without considering the actual phenol contents of wheat grains, and it came out that most of phenols – especially phenolic acids – was esterified to cell wall of wheat grain (Adom et al., 2003). Today, it is obvious that both conjugated and free soluble phenol acids are located mostly in pericarp, though insoluble phenolic acids are located mainly in the cell walls, associated with lignins.

Phenolic acid derivatives fall into two classes as hydroxybenzoic and hydroxycinnamic acids. Hydroxybenzoic acid comprises gallic, *p*-hydroxybenzoic, vanillic, syringic, and protocatechuic. Hydroxycinnamic acids include coumaric, caffeic, ferulic, and synaptic acid. A few studies have shown that phenolic acids are major wheat grain secondary metabolites and the amount of total phenolic content is greater than the other classes of secondary metabolites and other derivatives of phenols present in wheat grain. Ferulic acid, *p*-coumaric acid, syringic acid, vanillic acid, and *p*-hydroxybenzoic acid are common phenolic acids in wheat grain, and a

few studies have shown that phenolic acids are mostly located in the wheat grain bran (Adom et al., 2003; Verma et al., 2009). Table 6.3 summarizes the quantities of major phenolic acids among ancient wheat species which includes *T. monococcum* ssp. *monococcum*, *T. boeoticum*, *T. urartu*, *T. dicoccoides*, *T. dicoccum* Schrank., *T. macha*, and *T. spelta*.

In ancient wheat species, ferulic acid is the predominant phenolic acid type and mostly located in the bran part. It was shown that the amount of ferulic acid in the grains of ancient wheat could range between 816.00 µg/g and 184.73 µg/g and the weighted mean of ferulic acid was 613.56 µg/g. *p*-Coumaric acid was the higher phenolic acid (especially in the bran part) than ferulic acid. The concentration in

Table 6.3 Major phenolic acid contents of ancient wheat species or cultivars have been published in the distinct studies over the last decades

	<i>Ferulic</i>	<i>p-Coumaric</i>	<i>Syringic</i>	<i>Vanillic</i>	<i>p-Hydroxybenzoic</i>
<i>T. monococcum</i>					
Range:	184.73–746.04	23.10–310.3	2.90–4.90	3.10–5.23	0.80–4.46
WM ^a :	574.77	117.6	3.68	3.89	1.80
N ^b :	46	56	28	29	35
<i>T. dicoccum</i>					
Range:	236.30–816.00	21.80–294.00	1.70–4.10	2.40–7.80	1.10–7.29
WM:	617.31	102.89	2.98	4.25	2.44
N:	73	62	33	36	48
<i>T. spelta</i>					
Range:	236.20–562.00	13.80–35.70	3.10–6.00	1.89–3.70	1.21–2.40
WM:	396.94	22.16	4.53	2.92	1.81
N:	24	21	19	21	21
<i>T. dicoccoides</i>					
Range:	219.36–559.80	ND ^c	4.60–5.70	9.00–10.20	ND
WM:	408.72	26.05	5.15	9.60	3.60
N:	17	14	14	14	14
<i>T. boeoticum</i>					
Range:	ND	ND	ND	ND	ND
WM:	226.44	ND	ND	ND	ND
N:	2	ND	ND	ND	ND
<i>T. macha</i>					
Range:	787.00–804.00	44.00–77.00	ND	ND	ND
WM:	795.50	60.50	ND	ND	ND
N:	2	14	ND	ND	ND
<i>T. urartu</i>					
Range:	ND	ND	ND	ND	ND
WM:	ND	60.20	7.20	ND	ND
N:	ND	1	1	ND	ND

^aWeighted mean

^bN: the number of ancient wheat sample has been involved in studies until now

^cNot detected

157 ancient wheat samples ranged between 130.30 $\mu\text{g/g}$ and 13.80 $\mu\text{g/g}$ (weighted mean of 97.90 $\mu\text{g/g}$). Vanillic acid was in the range from 10.20 $\mu\text{g/g}$ to 1.89 $\mu\text{g/g}$, and the mean value was 4.01 $\mu\text{g/g}$ in the 101 ancient wheat species and cultivars. Syringic acid was in the range of 1.20 $\mu\text{g/g}$ and 7.20 $\mu\text{g/g}$. Its mean value was 3.57 $\mu\text{g/g}$ in the 57 ancient wheat species and cultivars. *p*-Hydroxybenzoic acid concentration was in the range of 7.29 $\mu\text{g/g}$ and 0.80 $\mu\text{g/g}$. The mean value was 2.22 $\mu\text{g/g}$ in 199 ancient wheat species and cultivars.

In addition, another predominant compound is lipophilic alkylresorcinol (AR). AR is also included in polyphenol group, which has potential antioxidant activity and protective ability to counteract oxidative damage and so on.

In wheat kernel, the major site of AR location is the outer cuticle of testa/inner cuticle of pericarp. Minor amounts of AR are also present in the outer cuticle of pericarp (Landberg et al., 2008). Within the scope of HEALTHGRAIN diversity project, Andersson et al. (2008) investigated durum wheat, bread wheat, einkorn, emmer, and spelt for AR. The AR contents in spelt, einkorn, and emmer wheat varieties were 490–741 (mean = 605), 545–654 (mean = 595), and 531–714 (mean = 581) $\mu\text{g/g}$ of dm, respectively. It was evident that the AR content of durum wheat was in the same range as common wheat, whereas the contents of spelt, einkorn, and emmer wheats were significantly higher.

Wheat grain phenols are thought to play a protective role against the chronic diseases. Those substances abovementioned have been recently subjected to many epidemiological studies. Epidemiological studies showed that polyphenols are potent inhibitors of LDL oxidation of which one of the key mechanisms is to develop chronic inflammatory diseases such as atherosclerosis (Aviram et al., 2000). In addition, polyphenols can improve endothelial dysfunction associated with different risk factors for atherosclerosis before the formation of plaque. Plaque is a prognostic tool for coronary heart diseases (Schachinger et al., 2000). Polyphenols may also exert antithrombotic effects by means of inhibiting platelet aggregation via preferential inhibition of cyclooxygenase 1 (COX 1) activity (Pirola & Fröjdö, 2008).

Polyphenols influence the metabolism of pro-carcinogens by modulating the expression of cytochrome P450 enzymes involved in their activation to carcinogens. Higher level of insulin like growth factor-1 (IGF-1) is associated with a higher risk of development of prostate cancer. IGF-1 binding to its receptor is a part of signal transduction pathway which causes cell proliferation. The association of polyphenol and IGF-1 is phenol compound found to block IGF-1-induced progression of cells into S phase of cell cycle. Numerous studies report the antidiabetic effects of polyphenols. Polyphenols may affect glycemia through different mechanisms, including the inhibition of glucose absorption in the gut or of its uptake by peripheral tissues (Kamaraj et al., 2007; Scalbert et al., 2005; Sharma & Rao, 2009).

6.9 Tocols (Vitamin E)

Vitamin E is seriously taken into consideration by medical and nutrition science due to understanding of the relationship between oxidative stress and impact onset of many chronic diseases. Since its availability, strong marketing potential, overall health beneficial, and its positive role in oxidative stress, consuming food rich in these compounds becomes crucial for human health and prevented diseases caused by the absence of these plant-derived molecules. Vitamin E molecules are synthesized only by plants and cyanobacteria; human metabolism is not capable of producing these molecules (Yang et al., 2011).

Vitamin E refers to a group of eight fat-soluble vitamins which can be divided in four tocopherols and four tocotrienols. Tocopherols have a phytyl chain, while tocotrienols have an unsaturated side chain with double bonds at positions 3', 7', and 11'. These two groups of vitamin E have four isomers each, designated as alpha and beta, which differ in number and position of methyl groups in the chroman ring. The shikimic acid pathway present in plants, algae, and bacteria but not in animals as a key pathway yielding homogentisic acid which is precursor for the synthesis of tocopherol isomers (Fig. 6.6) (Yang et al., 2011).

As an antioxidant, vitamin E has many functions in human metabolism and is involved in many processes for protecting against several diseases. Vitamin E protects cell membranes and lipoproteins from oxidative damage by scavenging lipid hydroperoxyl radicals and reactive oxygen species. Beside these actions in the metabolism, vitamin E compounds are subjected to preclinical treatments. *RRR*-tocopherol is one of the active forms of vitamin E. It inhibits proliferation of vascular smooth muscle cells by modulating activity of PKC in cell cultures, low-density lipoprotein-stimulated smooth muscle cell proliferation, and PKC activity *in vitro*. Vitamin E prevented formation of cholesterol-induced atherosclerotic lesions and induction of PKC activity, suggesting an involvement of signal transduction events in the protective effect of vitamin E against atherosclerosis (Ozer et al., 1998; Sirikci et al., 1996). *CD36* gene has a role in the atherosclerotic lesions. The studies on the activity of vitamin E on *CD36* mRNA expression revealed that vitamin E has reduced atherosclerotic lesions. Recently, it has been shown that vitamin E supplementation downregulates vasculature mRNA expression of *CD36* and upregulates mRNA expression of *PPAR γ* , *LXR α* , and *ABCA1* (Tang et al., 2014). The gene-regulatory activity of α -tocopherol on *CD36* expression has also been observed in the liver (Barella et al., 2004). Dietary vitamin E crosses the blood-brain barrier (Mardones & Rigotti, 2004), thus being available to reach different areas of the brain. Vitamin E may influence the complex interplay among different mechanism of neuronal cell loss, such as autophagy, apoptosis, and necrosis, in postischemic brain and may confer neuroprotection or early/postischemic secondary prevention, reducing, therefore, the long-term consequences of the insult. In addition, the considerations may sustain the general assumption that dietary vitamin E is "more essential" for normal development of the brain and for neuroprotection during the period of growth and even more during the uterine life due to the rapid development

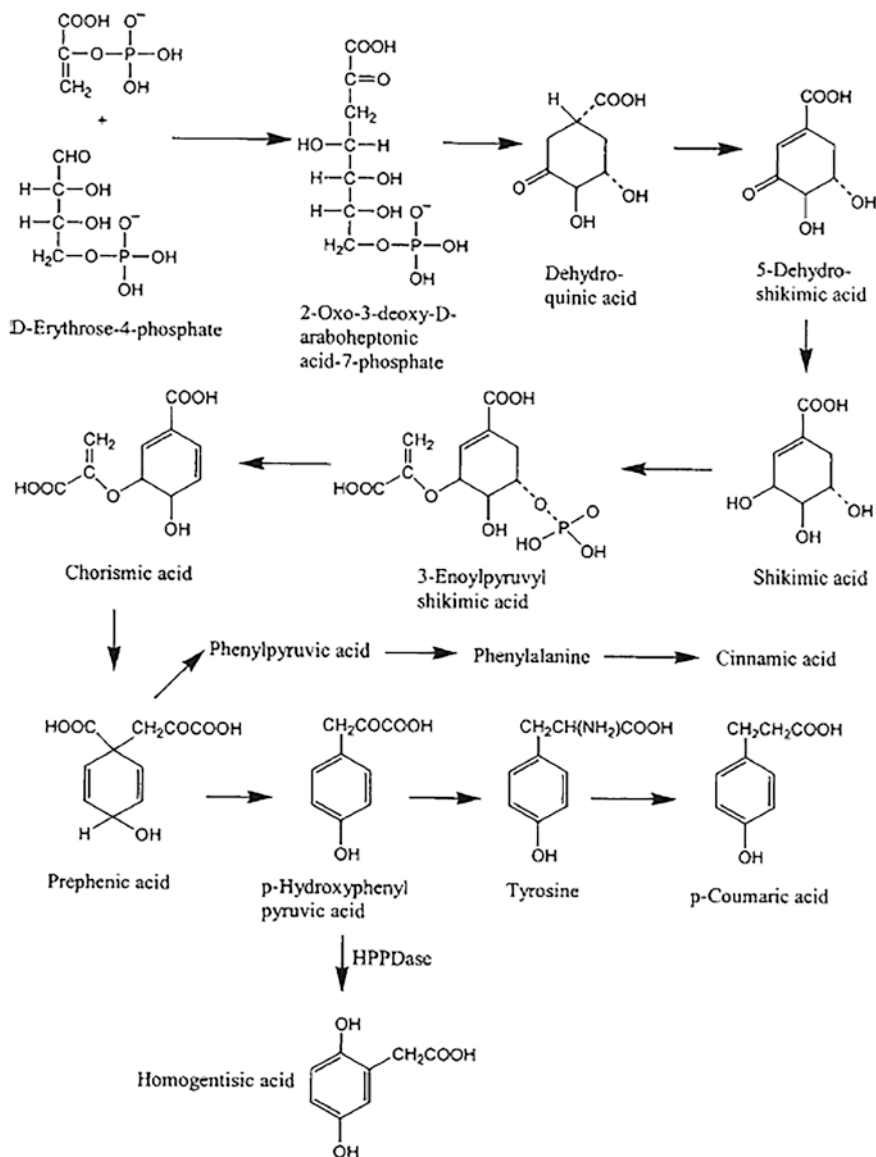


Fig. 6.6 Biosynthesis of homogentisic acid. Homogentisic acid forms the backbone of tocopherol compounds. This reaction provides the substrate for the formation of tocotrienols by 2-methyl-6-phytylbenzoquinol methyltransferase activity

of embryonic and fetal tissues. Besides the roles in developing the brain, neuroprotection effects of vitamin E have been proposed to extend also to other phases of brain life with effects on key detoxification and anti-inflammatory genes (Calabrese et al., 2003). The intake and circulating levels of different forms of vitamin E have

been observed to influence the risk of mild cognitive impairment and Alzheimer's disease (Mangialasche et al., 2012, 2013). Different aspects in the antioxidant-dependent and antioxidant-independent functions of vitamin E may have a role in hepato-protection and improved lipid metabolism of NASH liver. A shortage of these functions could be anticipated in the pathobiology of this disease state. In fact, the lipid abnormalities of fatty liver may interfere with the metabolism and vitamin status of vitamin E and possibly of other fat-soluble vitamins.

Several studies have been reported so far in terms of vitamin E content of ancient wheat. However, there is limited knowledge with respect to the tocochromonal compounds present in ancient wheat. *T. urartu*, *T. monococcum*, *T. dicoccum*, *T. spelta*, and *T. macha* were evaluated for the content of tocochromonols. β -Tocotrienol is the predominant tocochromonol type. β -Tocotrienol is mostly located in the bran part. The amount of β -tocotrienol in the grains of ancient wheat may range between 7.81 $\mu\text{g/g}$ and 46.96 $\mu\text{g/g}$, and weighted mean of β -tocotrienol was 26.87 $\mu\text{g/g}$ in 94 ancient wheat samples. The second highest levels were measured for α -tocotrienol. α -Tocotrienol concentrations ranged from 1.80 $\mu\text{g/g}$ to 17.60 $\mu\text{g/g}$ with a weighted mean of 5.91 $\mu\text{g/g}$. In addition, the majority of the tocopherols (>90% of tocols) was detected in the germ. The range α -tocopherol concentration was between 5.69 $\mu\text{g/g}$ and 16.05 $\mu\text{g/g}$, and the mean value was 10.07 $\mu\text{g/g}$ for a total of 94 ancient wheat species and cultivars. β -Tocopherol was in the range of 2.00 $\mu\text{g/g}$ and 10.86 $\mu\text{g/g}$. Its mean value was 5.72 $\mu\text{g/g}$ (Abdel-Aal ve Rabalski, 2008a; Brandolini et al., 2015; Digesu et al., 2009; Giambanelli et al., 2013; Hejtmánková et al., 2010; Hidalgo et al., 2006, 2008; Lachman et al., 2013; Pehlivan Karakas et al., 2021; Piironen et al., 2008).

Karmowski et al. (2015) evaluated the antioxidant activities of vitamin E compounds by using photochemiluminescence, indicating that the antioxidant activity of α -tocopherol was superior to that of β -tocopherol. Just like this, that of β -tocotrienol was greater than that of α -tocotrienol. The reason why β -tocotrienol has greater antioxidant activity is due to its low redox potential (Table 6.4).

There are a few factors which affect the tocol content of wheat grains; genotype and environmental conditions are considered as a main source of variation for tocol content. It is reported that there is a positive correlation for protein/tocol and lipid/protein content of einkorn wheat. Another observation on tocol content reported by Digesu and colleagues is that cultivars released before 1970 had lower YPC than recently released cultivars (Digesu et al., 2009). In this case, climate change might affect the amount of tocochromonol contents in wheat grains in positive way. However, it is reported by Giambanelli et al. (2013) that genotype and crop year affected the content of majority of the component of wheat grain, while lipids and tocol were not affected by these factors (Giambanelli et al., 2013). They investigated the association of wheat grain tocol and sowing date when grain samples were sowed in either fall or spring. Einkorn accessions had the highest average values of tocol in fall sowing, whereas macha and emmer wheat had higher tocol content in spring sowing. In addition, they could not detect the significant effect of sowing date for tocol amounts in ancient wheat grains. However, genotype and tocochromonal content in ancient wheat is positively correlated.

Table 6.4 Major phenolic acid contents of ancient wheat species or cultivars have been published in the distinct studies over the last decades

	α -Tocopherol	β -Tocopherol	α -Tocotrienol	β -Tocotrienol
<i>T. monococcum</i>				
Range:	7.20–9.10	1.16–5.60	2.94–17.60	9.30–45.45
WM ^a :	8.99	3.05	9.34	28.67
N ^b :	30	30	30	30
<i>T. dicoccum</i> Schrank.				
Range:	6.11–14.50	3.10–10.86	1.58–7.24	7.81–46.96
WM:	10.55	6.38	4.69	26.44
N:	47	47	47	47
<i>T. spelta</i>				
Range:	7.20–16.05	3.00–10.16	2.60–10.05	12.40–37.52
WM:	10.71	6.07	4.30	21.52
N:	14	14	14	14
<i>T. macha</i>				
Range:	7.05–8.34	4.08–6.40	3.89–5.07	33.62–46.91
WM:	7.70	5.25	4.48	40.26
N:	2	2	2	2
<i>T. Urartu</i>				
Range:	–	–	–	–
WM:	9.90	2.9	9.4	37.9
N:	1	1	1	1

^aWeighted mean^bN: the number of ancient wheat sample has been involved in the studies until now

In conclusion, the aspect of YPC in wheat species is taken into consideration by breeders due to the demand in the markets. In this way, the amount of tocochromonals content of durum wheat is higher than the others in current literature. α -Tocopherol is used as a biomarker for YPC by breeders; nonetheless, the amount of β -tocotrienol is considerably greater than α -tocopherol concentration; thus, β -tocotrienol would be a better alternative for the evaluation of YPC.

6.10 Carotenoids

Carotenoids are secondary metabolites, mainly C40 isoprenoids with more than 700 members (Hirschberg, 2001). A variety of organisms such as plants, algae, fungi, and bacteria have required pathways for biosynthesis of carotenoids. Animals, on the other hand, are nutritionally required to receive carotenoids and many of their derivatives. They are involved in a number of processes, for example, photosynthetic system, photoprotection, and synthesis of phytohormones (i.e., ABA and strigolactones). In addition, they provide an aroma to flowers to attract insects for seed dispersion and pollination, protecting seed from environmental factors as well as inducing a response mechanism to environmental stresses like photooxidative

ones. Since they are used as feed additives to enhance pigmentation of fish and eggs and as colorizing agents for human foods, cosmetics, and pharmaceutical product, carotenoids have a higher market value.

The core carotenoid pathway is conserved in most plant species although some plants accumulate special and rare carotenoids via unique biosynthetic routes (Fig. 6.7).

Lutein and zeaxanthin are the most abundant pigments in human macula, supporting the protective role of lutein in age-related macular degeneration; thus, intake of those carotenoids has been taken into account over the years (Landrum & Bone, 2001). Besides, a few studies have shown a positive correlation between the consumption of carotenoid and a decreased risk of several cancer types such as lung, prostate, esophagus, larynx, and pharynx. Lung cancer and dietary carotenoids support the observation of decreased morbidity upon β -carotene supplementation in nonsmoking adults. Several carotenoids, as well as carotenoid-rich food, might be involved in the risk reduction of the prostate cancer. Lycopene is regarded the most potent agent against the prostate cancer. The human trials show that β -carotene has the potential for preventing oral, pharynx, and larynx cancers. Furthermore, many evidence indicates a negative correlation between the intake of carotenoid-rich fruits and vegetables and the risk for colon cancer development. Carotenoids are lipophilic compounds, and they can efficiently diffuse through the cell membranes due to their hydrophobic nature. This phenomena helps cells for protecting the membrane integrity against ROS-mediated disruption due to their antioxidant activity which in turn protect body against cardiovascular diseases (Chan et al., 2005; Giovannucci, 2002; Le Marchand et al., 1993; Marshall, 2009; Mayne et al., 1994; McGarr et al., 2005; Mills et al., 1989).

Animals do not have metabolism for carotenoid derivatives synthesis; thus, they rely on their diet to obtain these compounds ready. In wheat, these compounds are partially present (Table 6.5). Lutein is a predominant carotenoid compound, mainly concentrated in the germ fraction of grain. Beside lutein, zeaxanthin is present in trace amount (Hidalgo et al., 2008).

Carotenoid concentration varies substantially among ancient wheat species. Table 6.6 depicted average carotenoid content of several ancient wheat species. Lutein, zeaxanthin, α -carotene, β -carotene, β -crypto-xanthin are unevenly present in ancient wheat grain (Abdel-Aal & Rabalski, 2008a; Digesu et al., 2009; Giambanelli et al., 2013; Hidalgo et al., 2006; Lachman et al., 2013; Serpen et al., 2008; Ziegler et al., 2015, 2016).

Zeaxanthin, β -crypto-xanthin, α -carotene, and β -carotene concentration is very low. The amount of lutein in the grains of ancient wheat ranged between 0.78 $\mu\text{g/g}$ and 8.10 $\mu\text{g/g}$, and weighted mean of lutein was 2.88 $\mu\text{g/g}$ in total 250 ancient wheat

→
Fig. 6.7 (continued) subsequently can be converted to lycopene molecule. Lycopene is the branching point in the pathway. While one way goes to α -carotene, the other way goes to β -carotene by the action of cyclase enzymes which eventually results in production of lutein and zeaxanthin, respectively. *PDS* phytoene desaturase, *ZDS* ζ -carotene desaturase, *β LCY* β -cyclase, *ϵ LCY* ϵ -cyclase, *β OH* β -hydroxylase, *ϵ OH* ϵ -hydroxylase, *ZE* zeaxanthin epoxidase, *NXS* neoxanthin synthase, *VDE* violaxanthin deepoxidase

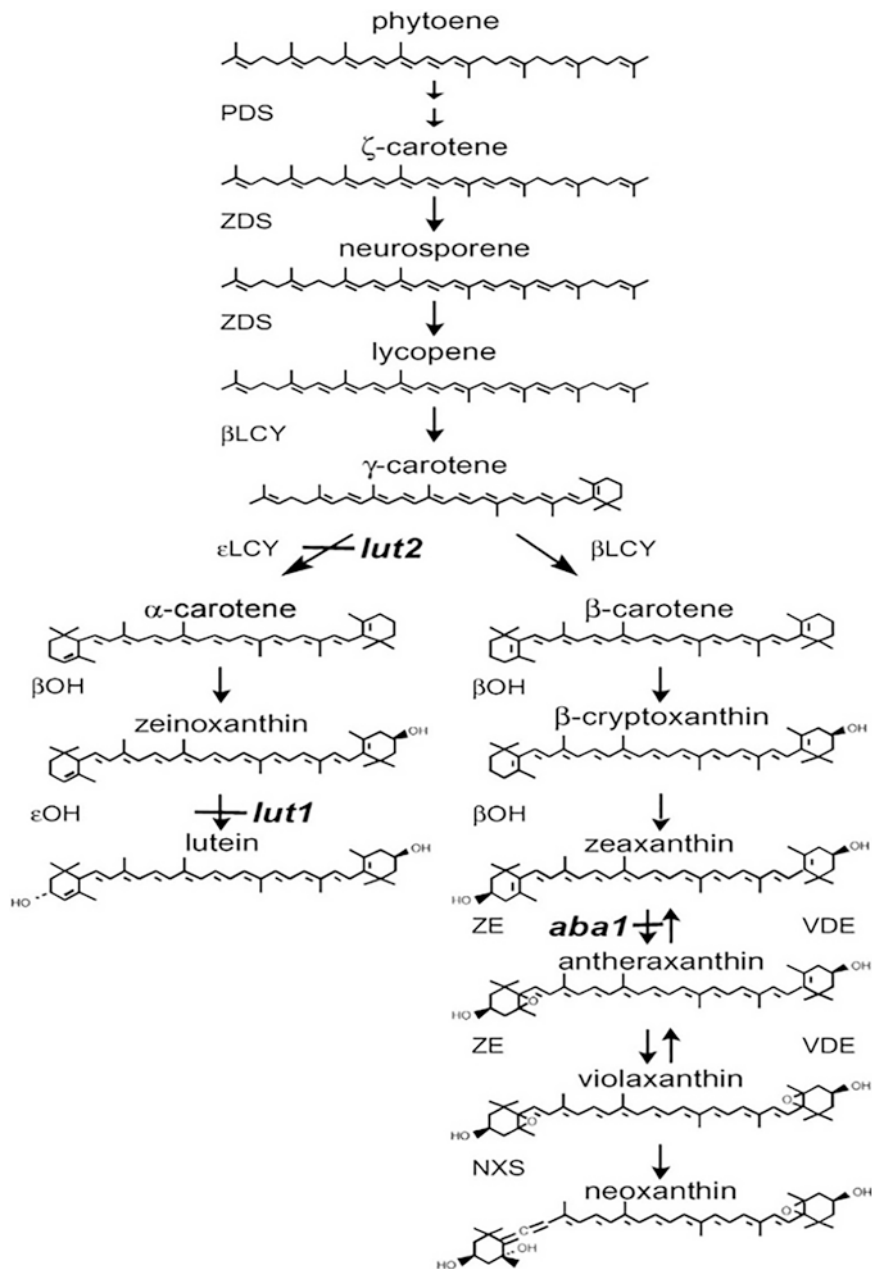


Fig. 6.7 Biosynthesis of carotenoids. Carotenoids are derived from MEP pathway which is taken place in plastids of the organisms. Pyruvate, synthesized from the glucose in cytoplasm, and glyceraldehyde-3-phosphate, synthesized in Calvin cycle in stroma, act as primary substrates to synthesize GGPP which is a common precursor for carotenoids biosynthesis and several other terpenoid compounds. The condensation of GGPP molecule results in phytoene which

Table 6.5 Lutein, zeaxanthin, β -crypto-xanthin, and $\alpha+\beta$ -carotene in ancient wheats

	<i>Lutein</i>	<i>Zeaxanthin</i>	<i>β-crypto-xanthin</i>	<i>$\alpha+\beta$-carotene</i>
<i>T. monococcum</i>				
Range:	3.26–8.10	0.24–1.03	0.18–0.33	
WM ^a :	5.81	0.57	0.28	
N ^b :	130	18	8	
<i>T. dicoccum</i>				
Range:	0.78–3.97	0.16–0.67	0.09–0.17	
WM:	1.45	0.46	0.12	
N:	74	30	25	
<i>T. dicocoides</i>				
Range:	ND	ND	ND	
WM:	1.07	0.15	ND	
N:	8	8	ND	
<i>T. spelta</i>				
Range:	0.87–2.10	ND	ND	
WM:	1.48	ND	ND	
N:	36	ND	ND	
<i>T. macha</i>				
Range:	ND	ND	ND	
WM:	1.26	0.39	0.22	
N:	2	2	2	

^aWeighted mean^bN: the number of ancient wheat sample has been involved in the studies until now**Table 6.6** Carotenoid contents of ancient wheat species or cultivars have been published in the distinct studies over the last decades

Study	<i>T. monococcum</i>	<i>T. dicoccum</i>	<i>T. dicocoides</i>	<i>T. spelta</i>	<i>T. macha</i>
Ziegler et al. (2016)	✓	✓	ND ^a	✓	ND
Ziegler et al. (2015)	✓	✓	ND	✓	ND
Lachman et al. (2013)	✓	ND	ND	ND	ND
Giambanelli et al. (2013)	✓	✓	ND	✓	✓
Digesu et al. (2009)		✓	✓	ND	ND
Abdel-Aal and Rabalski (2008b)	✓	✓	ND	✓	ND
Serpen et al. (2008)	✓	✓	ND	ND	ND
Hidalgo et al. (2006)	✓	ND	ND	ND	ND

^aND: Not available data

samples. The second greater one was zeaxanthin; 58 ancient wheat samples ranged from 0.15 $\mu\text{g/g}$ to 1.03 $\mu\text{g/g}$ in the weighted mean of 0.48 $\mu\text{g/g}$. Einkorn is rich in carotenoid among ancient wheat species. In addition to these compounds, α -carotene and β -carotene has been detected in trace amounts (Digesu et al., 2009; Hidalgo et al., 2006). As in the other chemical components of grains, variability of carotenoid concentration in ancient wheat grain is attributed to their genetic background and environmental factors.

6.11 Other Components

The B vitamin complex comprises eight water-soluble components which often occur together in the same foods and is initially considered to be a single component. Wheat, and in particular wholegrain, is an important source of B vitamins: thiamine (B1), riboflavin (B2), niacin (B3), pyridoxine (B6), and folates (B9) (Piironen et al., 2008).

Folate is currently one of the most studied vitamins. The reason why is its role in the prevention of neural tube defects in the fetus. Foliates have impacts beyond preventing the classical folate deficiency and megaloblastic anemia (Pitkin, 2007). Foliates are the only one that has been screened in ancient wheat species, as part of the HEALTHGRAIN project. Durum wheat (0.74 mg/g dry weight) and emmer (0.69 mg/g dry weight) showed higher concentration compared to the other species (0.56–0.58 mg/g dry weight), and good and poor folate sources were evident in all wheat types, including einkorn, emmer, and spelt. Thus, most of the genotypes, in particular einkorn, emmer, and spelt, were in the range of 400–450 ng/g dry weight. In wheat kernel, the most abundant folates vitamer is 5-HCO-H₄-folate, and 5-HCO-H₄-folate, which is one of the more stable vitamers, was reported to be the main vitamer in wheat grains, but it is found to be a little bit lower in spelt, einkorn, and emmer than in modern species (Piironen et al., 2008).

Phytosterols are well-known for their ability to lower serum total and low-density lipoprotein (LDL) cholesterol levels by inhibiting the absorption of cholesterol in the small intestine. Most abundant phytosterols are sitosterol, sitostanol, campesterol, and campestanol subset of steryl ferulates which is esterified with ferulic acids. There is a considerable variation, particularly in spelt and einkorn, among ancient wheat species. The ranges of the total phytosterol contents reported by Nurmi et al. (2008) of durum, spelt, einkorn, and emmer wheat genotypes were 871–1106, 893–963, 976–1187, and 796–937 $\mu\text{g/g}$, respectively. Smaller kernel sizes and higher bran yields result in higher phytosterol contents in ancient wheats (Nurmi et al., 2008).

Besides nutritious content of ancient wheat, harmful or antinutrient compounds that wheat does not have in its structure are also vital. The presence of antinutritional factors limits the bioavailability of the essential nutrients present in wheat grain, in particular minerals. The antinutrient content of these ancient wheat species is not clearly known; on the other hand lowering antinutrient, if it is considerably

present, will contribute to its functional food attribute. Trypsin/ α -amylase inhibitor family, belonging to the cereal trypsin/R-amylase inhibitor family, are active against insect and mammalian R-amylases, but not against the cereal enzymes. Fontanini et al. (2007) identified the emmer R-amylase inhibitory fraction that is composed of two newly identified proteins (emmer dimeric inhibitor 1 (EDI-1) and emmer dimeric inhibitor 2 (EDI-2)) sharing very high identity levels with related proteins from *T. aestivum*.

6.12 Antinutrients

6.12.1 *Is Einkorn Promising for Celiac Disease Patients?*

Celiac disease is an autoimmune disease, which is caused by a reaction to **gluten**. Upon exposure to gluten, an abnormal **immune** response may lead to the production of several different **autoantibodies** that can affect a number of different **organs**. In the small bowel, this varying autoantibodies may produce shortening of the villi lining the small intestine caused by an **inflammatory reaction**. These cases that have taken place in the small bowel limit the absorption of nutrients, mostly leading to a decrease in the total amount of red blood cells or hemoglobin. The incidence of celiac disease is steadily rising, and 1% of the population around the world suffers from it (Lebwohl et al., 2015). In addition, there has not existed an effective treatment for the improvement or remedial factor of celiac disease so far; thus, the patients who have celiac disease should be under gluten-free diet throughout their lifespan. For the patients, availability of foods that do not lead to toxic effects in the consumption of cereals and cereal byproduct is vital. Wheat species which has high gluten protein content might be toxic for celiac patients, especially to those consuming bread wheat. This toxic effect is mainly due to its gluten protein content. It is known that these peptides are encoded by several loci.

Gliadin subunits of gluten protein are documented as the main cause of the toxicity. Gliadin is a prolamin which is an alcohol-soluble protein and is a group of plant storage proteins having a high proline content and is found in the seeds of cereal grains. Comparative investigations of prolamins of different cereals have shown a close relationship between the amino acid composition and celiac toxicity, since tertiary structure of gliadin is not implemented with its toxicity. On the other hand, it is likely that the toxicity is somehow related with the composition of certain amino acids. For instance, the toxic prolamins of wheat are characterized by high contents of glutamine (about 36%) and proline (17–23%). The causal agent resides mainly in the gliadin fraction of gluten: all three main structural types of gliadins, α -, β -, γ -, and ψ -gliadins, are active, and other components also exacerbate celiac disease. These prolamins have been assigned into three groups: sulfur-poor (S-poor), S-rich, and high molecular weight (HMW) prolamins. The S-poor prolamins consist of ψ -gliadins and contain little or no cysteine residues. They are predominantly

monomeric (Mr 30–80 kDa) and comprising a single domain made up almost entirely of a single repeat motif. A group of S-poor prolamins may associate with disulfide bonded glutenin polymers, behaving as low molecular weight glutenin subunits (LMW-GS). They are known as D-type LMW glutenins and are considered mutant ψ -gliadins in which the presence of single cysteine residues allows cross-linking. The S-rich prolamins account for about 70–80% of the prolamins fraction (Mr 30–55 kDa) and include both monomeric α -, β -, γ -, and ψ -gliadins and polymeric LMW glutenins. They consist of a repetitive N-terminal domain, representing up to half of the molecule, and a non-repetitive cysteine-rich C-terminal domain. In addition, α -, β -, and γ -gliadins contain two and one polyglutamine regions and six and eight conserved cysteine residues, respectively. Cysteins (Cys) form either three or four intra-chain disulfide bonds; additional Cys can be present allowing the incorporation of α -, β -, and γ -gliadins into gluten polymers as *bound* gliadins.

α -gliadin subunit contains a stable 33mer fragment. This 33mer fragment is naturally formed by digestion, binding well to DQ2 – most likely confers disease susceptibility by their ability to bind the deamidated gluten peptides and form complexes that are recognized by interferon producing CD4⁺T cells within the celiac lesions – after deamidation by tissue transglutaminase, and it is recognized much more effectively by intestinal T-cell lines than shorter peptides (Molberg et al., 2005; Shan, 2002; Sollid, 2002). Molberg et al. (2005) demonstrated that 33mer fragment is encoded by α -gliadin genes on the wheat chromosome 6D and thus absent from gluten of einkorn and even certain cultivars of the tetraploid pasta wheat.

It is noteworthy that alcohol-soluble peptides able to prevent agglutination of K562(S) cells by toxic prolamins also occur in monococcum wheat, suggesting that inactivity of prolamins from *T. monococcum* ssp. *monococcum* and *T. dicoccum* Schrank. against intestinal cells could be partly due to the presence of protective sequences in these species. In addition, some studies suggest a reduced or absent toxicity of *T. monococcum* ssp. *monococcum*, which lacks a highly immunoreactive α - β -gliadin peptide. This peptide is encoded by genes located on chromosome 6D, which are absent in the diploid A genome einkorn.

On the other hand, einkorn (like its *Triticum* relatives) still may not be safe for consumption for people suffering from celiac disease. Some preclinical treatments and the examination of genes involved in toxicity show that einkorn has the full potential to induce the celiac disease syndrome.

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Chapter 7

Nutritional and Technological Aspects of Ancient Wheat



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7.1 Introduction

Wheat, after maize, is the second most important grain, with worldwide production of 760.9 million tons in 2020, followed by rice. The top five wheat-producing countries are China, India, Russia, the USA, and Canada (FAOSTAT, 2021). The most important species of wheat are common and durum wheats because about 95% and 5% of the wheat grown in the world are common and durum wheats, respectively (de Sousa et al., 2021). However, there is a renewed interest in ancient wheats which have limited production and utilization compared to common and durum wheats (Geisslitz & Scherf, 2020). Ancient wheat species are among the most valuable

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crops of the Mediterranean and neighbouring Near East regions (Heller & Padulosi, 1996).

The ancient wheat term is reserved for particularly three ancient wheat species: *T. monococcum*, a diploid minor wheat species; *T. dicoccum* Schrank, a primitive tetraploid wheat which is also identified as emmer; and *T. spelta*, a hexaploid wheat identified as spelt which is still grown in Europe and Asia and now a specialty wheat in Canada and the USA (Morrison & Wrigley, 2004). In limited amounts, macha, vavilovii, and Khorasan wheat (known Kamut®) are also produced in some countries.

Einkorn, emmer, and spelt wheats are the main three cultivated species of ancient wheat, which are the bridge species between wild and cultivated (common and durum) wheats (Kulathunga et al., 2020). Ancient wheat species are known by the names “farro” in Italy (Cubadda & Marconi, 1996) and “kaplıca” in Turkey. Kaplıca means “covered” or “hulled” in Turkish. More definitely, the diploid species (einkorn) is called “siyez” or “IZA”, and the tetraploid one (emmer) is called “gernik” or “kavılca” (Karagöz, 1996).

Common wheat is particularly used for producing breads, cookies, cakes, noodles, etc. Durum wheat is mostly used for producing pasta and bulgur as well as bread and couscous. Ancient wheats are frequently utilized as whole-grains in salads and soups, although they are processed differently, with emmer and einkorn used primarily for pasta and spelt used generally for bakery products (Benincasa et al., 2015; Boukid et al., 2018). The einkorn and emmer wheats are still being used in rustic areas to cook traditional foods such as bulgur in Turkey, breads or pastry in Germany and Italy. Each product has its own features, processing conditions, and specific requirements (Peña, 2002; de Sousa et al., 2021).

The nutritional quality of wheat is closely related to its chemical composition; however, the technological quality is a result of the balance among various compounds in wheat and flour, such as starch, proteins, lipids, etc., as well as the relationships within and between these compounds (Johansson et al., 2020). The main component of the grain is starch, which accounts for 54–72% of the wheat grain; however, proteins, which consist of 8–20% of the wheat grain, are regarded the most important components for the wheat technological quality (Pomeranz, 1988; Johansson et al., 2020). The technological quality of wheat is influenced by the protein content and composition of endosperm, especially the contents and proportions of two gluten fractions, gliadins and glutenins, as well as their high molecular weight (HMW) and low molecular weight (LMW) subunits (Mefleh et al., 2019). Qualitative variations in composition and properties of gliadins and glutenins account for much of the differences in quality among wheat genotypes (Schofield, 1994).

Flour is a product that exists between wheat and finished products (bread, pasta, biscuits, etc.) and has a direct impact on end-product quality. Determining the rheological properties of flour is important for estimating the quality of the end-product. Each end-product has its own technological quality requirements (Johansson et al., 2020). In general, hard wheat flour with strong gluten and high protein content ($\geq 12\%$) is suitable for high-volume bread-making (Tipples et al., 1994; Khan & Shewry, 2009). Soft wheat flours with low protein content (7–10%), low

damaged-starch content, low water absorption, and fine granulation are more appropriate for making biscuits and crackers (Faridi et al., 1994). Wheats with high vitreousness, protein content, gluten strength, and semolina yield are preferred for producing pasta as well as bulgur (Matsuo, 1994).

Wheat is utilized for a wide range of end-products with differing quality requirements. Recent research projects to assess the potential of ancient wheats in terms of both nutritional and technological quality for human consumption are underway in various countries including Turkey, Germany, the USA, and France. The aim of this chapter is to review and compare the results accessible in the literature on the nutritional, technological, and end-product quality of ancient wheat.

7.2 Quality Characteristics of Ancient Wheat

7.2.1 Physical Properties

Grain size is one of the primary physical quality characteristics subjected heavily to the selection. The grain size has a strong effect on numerous compositional and qualitative characters because big and heavy kernels contain a higher amount of starchy endosperm, lesser proportions of aleurone layers, and the external pericarp (Brandolini et al., 2011). Kernels of einkorn have wide variation with 16.7–33.4 g, but lower kernel weight than those of emmer, spelt, common, and durum wheats (45.6–55.5, 36.7–41.7, 28.9–41.2, and 46.0–58.4 g, respectively) (Brandolini et al., 2008). In another study to compare kernel weights of cereal species, the highest kernel weights were obtained from spelt and common wheat genotypes with an average of 44.8 ± 0.87 g and 44.6 ± 5.03 g, respectively. Hexaploid common wheat was followed by durum (41.1 ± 7.43 g), emmer (39.5 ± 5.08 g), and einkorn (30.1 ± 5.95 g) wheats (Hejcman et al., 2016). Kernel weights belonging to 30 einkorn genotypes ranged between 28.2 ± 0.23 and 39.5 ± 0.18 g (Emeksizoğlu, 2016).

Environmental factors and agronomic applications, in addition to species and cultivars, may influence kernel weights. Hidalgo and Brandolini (2017) found that the thousand kernel weights of einkorn cultivars varied between 23.4 ± 0.45 g and 30.5 ± 0.21 g. Kernel weight also changed slightly between years because the protracted rain favoured the development of heavy kernels. Furthermore, the fertilization caused small irregular weight changes. On the contrary, fertilization increased grain weight in other wheat species (Makowska et al., 2008).

Grain hardness is one of the most important quality characteristics of wheat. The grain hardness is regulated by *Ha* hardness genes (Pin a and Pin b), which are located on the short arm of chromosome 5D, but it is also influenced by other small-effect loci (Pasha et al., 2010). In hexaploid wheat, Pin a and Pin b have different alleles (Morris & Bhawe, 2008). The diversity in Pin function has a significant influence on the milling and end-product quality of wheat (Pasha et al., 2010). The grain hardness is a trait that is significantly associated with wheat species. Durum wheat

lacks the D-genome and has a harder grain texture (Morris & Bhave, 2008). Durum means “hard” in Latin, and it is the hardest of all wheat species (Hare, 2017). Wheat is divided into soft, medium-soft, medium-hard, hard, and extra-hard based on the hardness of the kernels. This categorization provides a fundamental basis for discriminating the world wheat grain trade (Pasha et al., 2010).

Einkorn kernels exhibited extra-soft grain texture (99–306 g). It was followed by spelt (205–214 g), common (383–458 g), emmer (596–685 g), and durum (756–885 g) wheats (Brandolini et al., 2008). The confirmed extra-soft texture of einkorn (21.6 ± 17.0) with a lower hardness index than the samples of spelt (34.4 ± 4.0), emmer (87.1 ± 1.6), and common (45.7 ± 5.8) wheats was observed by Løje et al. (2003). Endosperm texture is so important that it often determines the type of wheat that will be utilized for particular products. Medium to hard hexaploid (*T. aestivum*) wheats with 11–13% protein content are required for bread-making, while soft to medium wheats with 8–12% are generally preferred for cakes, cookies, and noodles. Semolina from durum wheat is preferred for pasta-making (Schofield, 1994; Peña, 2002).

Grain hardness is as well important for milling and baking quality, being directly associated with the force required and the energy spent during grinding. It determines the level of starch damage during the period of milling. The adhesion between the protein matrix and the starch is higher in hard-grained wheats than in soft-grained types. Hard wheats have a higher flour extraction rate, starch damage, and energy consumption during milling (Bedo et al., 2010). Water absorption is generally higher for hard wheat flours than for soft wheat flours at the same protein level since damaged starch granules absorb more water than intact or undamaged granules. The water absorption is a significant quality trait for bakers since it directly affects the quantity of bread they can produce from a given weight of flour. Water absorption also has a significant impact on crumb softness and bread storage properties (Tipples et al., 1994).

7.2.2 Protein Content and Gluten Quality

Protein content and gluten quality are crucial in determining the functional characteristics of wheat for making bread or pasta (Cubadda & Marconi, 1996).

By comparing five wheat species, it has been found that the protein contents of einkorn, emmer, spelt, common, and durum wheats changed between 15.5 and 22.8, 14.0 and 16.0, 17.1 and 18.7, 13.7 and 15.7, and 14.0 and 18.3 g/100 g of dry matter, respectively (Brandolini et al., 2008). Grain protein contents of the ancient wheats were generally higher than those of modern counterparts, which were cultivated under the same growing conditions (Abdel-Aal et al., 1997; Løje et al., 2003; Stolickova & Konvalina, 2014). However, this doesn't justify the classification of ancient wheats in common as a protein-rich crop because their high protein content may be due to low grain yields (Čurná & Lacko-Bartošová, 2017). A negative

association existed between protein content and grain yield as shown by Shewry et al. (2013).

Grain quality characteristics, such as protein content, differ within and across species. But they are also highly affected by the environmental conditions (Arzani & Ashraf, 2016). The variation in grain protein content is strongly dependent on the wheat cultivar, growing conditions, soil fertility, and fertilizer, particularly nitrogen (Carson & Edwards, 2009). The genotype effect was found the strongest on grain protein content by Hidalgo and Brandolini (2017). However, according to Shewry et al. (2013), nitrogen fertilization had a greater effect on grain protein content than the genotype. Working with *Triticum* species, einkorn, emmer, spelt, common, and durum wheats, genotype–environment interactions were found to exist for grain protein content across all five wheat species except for emmer. This caused a large range of heritability for grain protein content, with estimates of heritability ranging from 0.29 to 0.91 for durum and common wheats, respectively (Longin et al., 2016). In terms of the limited nitrogen supply, einkorn, emmer, and spelt might have higher protein content than common and durum wheats (Longin et al., 2016).

Sedimentation tests (SDS and Zeleny) depend on the swelling and flocculation characteristics of gluten protein in dilute lactic acid, and the findings are linked to gluten strength and baking quality. SDS sedimentation is an extensively utilized predictive test in breeding programmes, especially when only a small amount of sample is available. The simple and rapid sedimentation test is a useful technique for forecasting the alveograph energy (W) value, confirmed by a significant relationship between sedimentation volume and alveograph energy (W) value (Vázquez et al., 2012).

The sedimentation volume in SDS varies from poor to good among genotypes. In a comprehensive study carried out by Quaranta et al. (2014), including 219 emmer and 203 spelt wheats, the range of SDS sedimentation values was found to be similar for both species (emmer, 14–80 mL; spelt, 15–90 mL). SDS sedimentation variation among genotypes of einkorn was also found out to change between 11 and 95 mL (Castagna et al., 1996) and 7 and 97 mL (Corbellini et al., 1999). SDS sedimentation values changed between 11 and 37 mL, while Zeleny sedimentation values belonging to the same eight emmer genotypes ranged from 6 to 19 mL (Stehno et al., 2011). Emmer with a low sedimentation value (20–27 cm³) could be challenging to use for baking (Desheva et al., 2016). Zeleny sedimentation values were 10–14 mL for einkorn, 12–18 mL for emmer, 31–42 mL for spelt, and 50 mL for common wheats (Stolickova & Konvalina, 2014). In another study including five wheat species, SDS sedimentation values were 14–66 mL, 20–27 mL, 53–63 mL, 45–75 mL, and 31–38 mL for einkorn, emmer, spelt, common, and durum wheats, respectively (Brandolini et al., 2008).

The genotype effect on SDS sedimentation value was strongest according to Hidalgo and Brandolini (2017). Similarly, in a comparative study of ancient and modern wheats, beside genotype effect, genotype–environment interactions were observed for SDS sedimentation value, and heritability changed from 0.85 (einkorn) to 0.96 (emmer and common) by Longin et al. (2016).

Wheat gluten was isolated in 1728 by Beccari who used solvent-fractionation procedures to discriminate four types of proteins: albumins, globulins, prolamins (gliadin), and glutelins (glutenin) (Wrigley, 2010). In wheat flour, gluten consisting of gliadins and glutenins represents about 80% of the total proteins (Hoseney et al., 1969; Shewry et al., 2009). The glutenins are polymeric proteins and contribute to the strength and elasticity of the dough, while the gliadins are monomeric proteins and responsible for dough viscosity and extensibility. Therefore, gluten plays a significant role in the baking quality of wheat flour by providing water absorption capacity, viscosity and elasticity, and cohesivity for dough (Wieser, 2007).

Thirty-nine glutenin and 44 gliadin bands were detected as a result of electrophoretic analysis of 668 einkorn accessions by Brandolini et al. (2003). The bread-making quality of einkorn, evaluated by the SDS sedimentation test, was improved by eight glutenin and eight gliadin bands. Especially relevant was the impact of three linked glutenin fragments that combined with diminish or absent ω -gliadins (Hidalgo & Brandolini, 2019). HMW glutenins have a greater impact on the elasticity and strength of dough (Gupta et al., 1991; Gupta & MacRitchie, 1994), which affects the bread-making quality of wheat (Šramková et al., 2009). Degaonkar et al. (2005) determined the HMW subunits in emmer storage proteins linked with good bread-making quality. However, emmer was found to be unsuitable for classical bakery processing but suitable for non-yeast products like pasta and biscuits (Konvalina et al., 2013).

Aside from the individual impacts of gliadins and glutenins, the ratio of gliadins to glutenins (Gli/Glu) is important for the bread-making quality, such as the high volume of bread (Wrigley et al., 2006). The dough mixing time and dough stability are decreased when the content of gliadins is higher than that of glutenins, but a higher quantity of glutenins, namely, HMW-GS and LMW-GS, is positively related to bread-making quality (Rodríguez-Quijano et al., 2019). A recent research, comparing the protein compositions of ancient and modern wheats grown at four locations, revealed that ancient wheats exhibited higher protein and gluten contents than common wheat regardless of location. With regard to protein composition, Gli/Glu ratio of common wheat was typically 1.5–3.1, and those of ancient wheats were much higher (spelt, 2.8–4.0; emmer, 3.6–6.7; einkorn, 4.2–12.0) (Geisslitz et al., 2019). Similarly, einkorn was characterized by a high gliadin/glutenin ratio and low amounts of HMW glutenins compared to common wheat (Wieser et al., 2009). These results are most likely the main explanation for the poor baking qualities of ancient wheats since high glutenin content is linked with good baking quality (Geisslitz et al., 2019).

Wet gluten is extracted from wheat meal or flour using an automated gluten washer. It is well known that the amount of wet gluten positively correlates with the amount of dry gluten (Desheva et al., 2014). In a study, including 38 accessions of emmer wheat, the wet gluten changed from 22.6% to 45.8%, while dry gluten varied from 7.95% to 15.83% (Desheva et al., 2016). Stolickova and Konvalina (2014) reported that the average wet gluten contents of einkorn, emmer, spelt, and common genotypes were 37.0%, 38.4%, 42.6%, and 29.0%, respectively. The wet and dry

contents of 30 einkorn genotypes were changed from 19.3% to 46.3% and from 6.8% to 18.3%, respectively (Emeksizoğlu, 2016).

The gluten index value indicates both gluten quantity and quality, with higher values representing stronger gluten. There is a wide range of gluten index within wheat species. The gluten index was found to be between 12.8% and 17.2% for einkorn (Konvalina et al., 2011), between 17% and 56% for emmer (de Vita et al., 2006), between 10% and 50% for spelt (Marconi et al., 1999), between 93% and 95% for common, and between 62% and 89% for durum (Rao et al., 2010) wheats. Emeksizoğlu (2016) reported that gluten index value of 30 einkorn genotypes, grown in Kastamonu, changed between 2.0% and 51.3%.

7.2.3 *Starch and Lipid Contents*

There is a high range of composition across the cereal grains, but all have moderate levels of protein, high starch content, and a rather minor quantity of fat (lipid).

Starch is a main functional component in wheat, and its content and properties are known to have a significant influence on the quality of wheat and its end-products. The total starch content is the main nutritional component of wheat with values ranging from 60.6% to 71.4%, from 70.2% to 70.7%, from 62.1% to 65.5%, from 65.3% to 72.9%, and from 65.2% to 68.7% for einkorn, emmer, spelt, common, and durum wheats, respectively (Brandolini et al., 2008). Variations in total starch contents of species were explained by differences in genotype and growing period conditions (Massaux et al., 2008).

Starch is comprised of 25% amylose (a combination of linear and weakly branched) and 75% amylopectin (monodisperse and highly branched) (Maningat et al., 2009). The ratio of amylose to amylopectin within the starch granules varies, depending on the species and the cultivar (McKevith, 2004).

Health nutrition trends favouring enhanced fibre intake with low glycemic foods have encouraged the growth of high amylose starch as a source of resistant starch that functions similarly to dietary fibre (Bertolini, 2010). Spelt wheat had around eight to ten times the amount of resistant starch when compared to common wheat (Abdel-Aal & Rabalski, 2008).

Lipids are a tiny component of wheat. Nevertheless, they play an important role in nutrition, storage, and processing such as dough mixing and baking. The lipids create complexes with the gluten proteins, which contribute to the stabilization of the gas-cell structure, therefore having a significant effect on the bread volume and final texture (Uthayakumaran & Wrigley, 2010).

The majority of the lipid in wheat is localized in the germ (28.5%) and aleurone (8.0%), with relatively minor quantities in the endosperm (1.5%) (Delcour & Hoseney, 2010). The lipid content varies similarly and narrowly within the wheat species: 2.03–2.85% for einkorn, 1.80–2.85% for emmer, 1.88–1.93% for common, and 1.96–2.82% for durum wheats (Giambanelli et al., 2013). Similarly, a low variability among ancient (emmer and spelt) and naked (common and durum) species

existed which changed from $2.5 \pm 0.1\%$ to $2.7 \pm 0.2\%$ in naked wheat and spelt, respectively (Pelillo et al., 2010). Lipid content of wheat was higher for spring (1.92–2.85%) than fall (1.80–2.65%) sowing, which was affected by sowing date (Giambanelli et al., 2013).

The lipidomic profiles of ancient wheats are different from each other (Righetti et al., 2016). Einkorn contains more monounsaturated fatty acids, less polyunsaturated fatty acids, and less saturated fatty acids compared to durum wheat, which is beneficial for health (Hidalgo & Brandolini, 2014; de Sousa et al., 2021).

7.2.4 Vitamins and Minerals

Wheat is a good source of vitamin B, mainly thiamine (B1), riboflavin (B2), niacin (B3), pyridoxine (B6), and folate (B9) (Shewry & Hey, 2015). Consuming whole-grain products provides 40% of the recommended daily allowance for thiamine, 10% for riboflavin, 22% niacin, 33% pyridoxine, and 13% folate (Uthayakumaran & Wrigley, 2010).

The thiamine content of wheat species varied narrowly from 0.50 mg/100 g to 0.60 mg/100 g. The riboflavin contents of einkorn (0.45 mg/100 g) and common (0.55 mg/100 g) wheats were relatively high, but those of spelt (0.14–0.17 mg/100 g) was relatively low. Nevertheless, niacin (2.0–5.7 mg/100 g) content of spelt was higher as compared to those of einkorn (3.1 mg/100 g) and common wheat (2.3 mg/100 g). The amount of pyridoxine changed among species, ranging from 0.35 mg/100 g to 0.49 mg/100 g (Abdel-Aal et al., 1995). Stehno et al. (2011) found that the levels of riboflavin (0.108–0.135 mg/100 g), thiamine (0.29–0.44 mg/100 g), pyridoxine (0.27–0.45 mg/100 g), and niacin (8.4–10.6 g/100 g) varied significantly across eight emmer genotypes as compared to common wheat with 0.071 mg/100 g riboflavin, 0.36 mg/100 g thiamine, 0.37 mg/100 g pyridoxine, and 6.8 mg/100 g niacin.

Cereal and cereal products are good sources of folate, a water-soluble form of vitamin B9 which is also known as folacin or folic acid. Folate is needed to avoid anaemia, cardiovascular disease, and neural tube defects (Scott et al., 2000; de Wals et al., 2007). In the HEALTHGRAIN project, the folate content of ancient and modern wheat species was screened, and the folate concentrations of emmer (0.69 $\mu\text{g/g}$ dmb) and durum (0.74 $\mu\text{g/g}$ dmb) wheats were slightly higher compared to those of common, einkorn, and spelt (0.56, 0.58, and 0.58 $\mu\text{g/g}$ dmb, respectively) wheats (Piironen et al., 2008).

In Gx E research using 26 lines, heritability values of these vitamins were quite low, with thiamine having the highest value (31%), followed by folate (24%), riboflavin (16%), pyridoxine (12%), and niacin (7%) (Shewry et al., 2013).

Wheat is rich in vitamin E, several B vitamins, and minerals. These are unequally distributed throughout the seed, with the majority found in the germ and the bran (Uthayakumaran & Wrigley, 2010).

Humans need more than 22 mineral elements to meet their metabolic demands. Some, like Na, K, Ca, and Mg, are required in large quantities, while others, including Fe, Zn, Mn, Cu, I, and Se, are required in trace levels (Welch & Graham, 2004).

According to research comparing the macro and micro element contents of einkorn, emmer, spelt, and common wheats, all ancient wheats contained significantly higher Zn (34–54%), Fe (31–33%), and Cu (3–28%) than common wheat (Suchowilska et al., 2012). *T. monococcum* contained larger amounts of minerals (Zn, Mg, Mn, K, and P) except for Fe than *T. dicoccum* (Zengin, 2015). Some mineral element contents of 30 einkorn genotypes changed from 397 to 558 mg/100 g for K, from 109 to 528 mg/100 g for P, and from 103 to 157 mg/100 g for Mg (Emeksizoglu, 2016).

Among the micronutrient deficiencies, zinc and iron insufficiencies are mainly important for affecting the health of humans. The levels of Zn and Fe in the 54 einkorn wheat accessions varied significantly, ranging from 0.21 to 2.16 $\mu\text{g}/\text{seed}$ for Zn and from 0.54 to 3.09 $\mu\text{g}/\text{seed}$ for Fe. This variation is expected to be utilized in the breeding programmes of wheat (Ozkan et al., 2007). Wild emmer is particularly a valuable genetic resource for increasing the zinc and iron levels in modern wheat (Cakmak et al., 2004).

Although there was a significant genotype–environmental interaction effect on Fe and Zn contents, there was a considerable genetic effect on both Zn and Fe accumulation in wheat as well. Further studies revealed that there was no negative relationship between grain yield and Fe and Zn contents in the grain. Hence, it should be able to increase the Fe and Zn contents of wheat grain via breeding (Welch & Graham, 2004).

7.2.5 Phytochemicals and Antioxidant

Ancient wheat has been identified as a possible source of several health-promoting components. Wheat grain is a good source of various health-related substances, notably phytochemicals, antioxidants, vitamins, and macro-micronutrients, along with the primary components of protein, carbohydrate, and lipid (Arzani, 2019).

Ferulic acid is the most abundant phenolic component in both the soluble-conjugated and insoluble-bound fractions of wheat species (Yilmaz et al., 2015). The average ferulic acid contents in spelt, durum, and common wheats were comparable (about 400 $\mu\text{g}/\text{g}$ of dm), higher in emmer (476 $\mu\text{g}/\text{g}$ of dm), and lower in einkorn (298 $\mu\text{g}/\text{g}$ of dm) wheats, according to Li et al. (2008). Similarly, Serpen et al. (2008) found that emmer wheat samples had about 2.1-fold more ferulic acid than einkorn wheat samples.

Alkylresorcinols are one of the major classes of phenolic compounds, and they are mostly present in high levels in the grain's outer layers (Landberg et al., 2008). The concentrations of alkylresorcinol varied greatly among the genotypes within each species, and the overall mean contents of the species were 737 ± 90.9 , 697 ± 93.6 , 743 ± 56.7 , 761 ± 92.3 , and 654 ± 47.9 $\mu\text{g}/\text{g}$ dm in einkorn, emmer,

spelt, common, and durum wheat samples, respectively (Ziegler et al., 2016). Alkylresorcinol mean values in the dry matter for emmer, einkorn, common, and durum wheats were 377 ± 17 , 344 ± 8 , 321 ± 18 , and 286 ± 11 $\mu\text{g/g}$, respectively (Ciccoritti et al., 2013). Similarly, the findings of the HEALTHGRAIN research revealed that considerable variation existed in total alkylresorcinol content in wheat species, with ancient wheats (einkorn, emmer, and spelt) having higher alkylresorcinol contents than modern wheats (durum and common) (Andersson et al., 2008).

Very high heritability values were calculated in all five wheat species for tocopherols ($h^2 = 0.88\text{--}0.97$), for steryl ferulates ($h^2 = 0.88\text{--}0.94$), and for alkylresorcinols ($h^2 = 0.69\text{--}0.97$). This demonstrated that the amount of lipophilic antioxidants in einkorn, emmer, spelt, common, and durum wheats was highly genetically controlled (Ziegler et al., 2016).

Tocols are a group of lipid-soluble liquids synthesized only by photosynthetic plants and classified into tocopherols and tocotrienols. A polar chromanol ring and a hydrophobic 16-carbon side chain are present in both. The side chain of tocopherols is a saturated isoprenoid group, while the side chain of tocotrienols comprises three double bonds. Tocopherols and tocotrienols are both made up of four derivatives, α -, β -, γ -, and δ -, and are referred to as tocopherols together (Hidalgo et al., 2006; Lampi et al., 2008; Okarter et al., 2010; Lachman et al., 2013; Ziegler et al., 2016). However, the most present derivatives are α - and β -tocols (Lampi et al., 2008). In wheat, there are more tocotrienols than tocopherols, with β -tocotrienol being the most abundant, followed by α -tocotrienol, α -tocopherol, and β -tocopherol (Hidalgo et al., 2006; Hidalgo & Brandolini, 2017).

While all tocopherols and tocotrienols are antioxidants, only α -tocopherol has vitamin E activity (Schneider, 2005). Besides their antioxidant characteristics, cereals' tocol content may provide health benefits such as decreasing cancer, cardiovascular diseases, and LDL cholesterol risks (Tiwari & Cummins, 2009). Tocotrienols may potentially be used as a neuroprotective dietary factor (Frank et al., 2012).

The mean values of total tocols were higher in einkorn than for the other ancient wheat species, with a wide range of contents by several researchers. In a study comparing the amount of total tocol in some wheat species, the tocol contents of 54 einkorn accessions varied from 61.45 to 115.85 $\mu\text{g/g}$ dm with an average of 77.96 $\mu\text{g/g}$ dm. In the same study, durum wheat samples had the lowest tocol content with 38.87–57.27 $\mu\text{g/g}$ dm. The tocol contents for emmer, spelt, and common wheats were similar and changed between 62.70 and 67.92, 67.48 and 69.18, and 53.16 and 74.94 $\mu\text{g/g}$ dm, respectively (Hidalgo et al., 2006). In a screening study of 175 genotypes belonging to einkorn, emmer, spelt, common, and durum wheat types, the average total tocol concentration ranged from 27.6 to 79.7 $\mu\text{g/g}$ dm with a mean value of 49.4 $\mu\text{g/g}$ dm, displaying a 2.9-fold higher variation among wheat genotypes (Lampi et al., 2008). As reported by Ziegler et al. (2016), the mean values of total tocols were 48.3 ± 4.8 $\mu\text{g/g}$ dm, 41.7 ± 3.0 $\mu\text{g/g}$ dm, 50.6 ± 5.7 $\mu\text{g/g}$ dm, 31.1 ± 3.4 $\mu\text{g/g}$ dm, and 36.9 ± 6.3 $\mu\text{g/g}$ dm in common, spelt, durum, einkorn, and emmer wheat samples, respectively.

Phytosterols, when consumed as part of a healthy diet, may help to prevent colon cancer and reduce total serum cholesterol (Piironen et al., 2008). The most plentiful

phytosterol in whole kernel fractions is β -sitosterol (34.2–42.7% of phytosterols) followed by campesterol, sitostanol, and campestanol (Giambanelli et al., 2016). Einkorn and emmer had comparable total sterol contents (554.3–828.5 and 500.8–816.4 mg/kg dm, respectively), which was higher than common wheat (440.8–661.8 mg/kg dm) but lower than durum wheat (614.8–929.0 mg/kg dm) (Giambanelli et al., 2013).

Carotenoids, which are lipid-soluble antioxidants, are produced by a large number of photosynthetic organisms. They are responsible for the orange, red, and yellow colours seen in a variety of flowers, fruits, and bird feathers. The carotenoid contents of 54 einkorn accessions ranged from 5.33 to 13.64 $\mu\text{g/g}$ dm, two- to four-fold higher than non-einkorn wheats, with an average of 8.41 $\mu\text{g/g}$ dm (Hidalgo et al., 2006). Lutein is the most abundant component in wheat, whereas other carotenoids like β -carotene are only found in minor levels (Hidalgo et al., 2006; Abdel-Aal et al., 2007). A study carried out by Ziegler et al. (2016) using whole-grain flours of 15 genotypes each of einkorn, emmer, spelt, common, and durum wheat species grown at four locations revealed that the concentrations of lutein varied greatly among the species and the overall mean contents of the species were 5.76 ± 0.96 , 1.43 ± 0.33 , 1.59 ± 0.28 , 1.30 ± 0.29 , and 3.13 ± 0.62 $\mu\text{g/g}$ dm in the aforementioned wheat samples, respectively. Einkorn wheat samples, on average, had nearly twofold higher lutein content than emmer wheats (Serpen et al., 2008). Similarly, einkorn had the highest lutein content which is around 6.37–8.46 $\mu\text{g/kg}$ dm in whole einkorn flour with an average value 7.41 $\mu\text{g/kg}$ dm, higher than durum (5.41 $\mu\text{g/kg}$ dm), emmer (3.97 $\mu\text{g/kg}$ dm), common (2.11 $\mu\text{g/kg}$ dm), and spelt (1.47 $\mu\text{g/kg}$ dm) wheats (Abdel-Aal et al., 2007). It is well known that the best way to get the most nutritional benefits from wheat is to consume it as whole-grain products. For instance, whole-grain einkorn bread can help to elevate the daily uptake of lutein since einkorn wheat has nearly twofold higher lutein content than the other species (Grausgruber et al., 2008; Ziegler et al., 2016).

Carotenoid concentration and lutein content were found to be genetically controlled (Lachman et al., 2013; Ziegler et al., 2016) or influenced by environmental conditions (Abdel-Aal et al., 2007). The total yellow pigment content is a widely utilized test in durum wheat breeding programmes to evaluate the quality of semolina and pasta products. There were significant positive relations between total yellow pigment content and lutein content ($r = 0.94$) and total carotenoids ($r = 0.99$). These results demonstrated that the colorimetric method or total yellow pigment content would be a good predictor for determining the lutein or total carotenoid content of wheat (Abdel-Aal et al., 2007).

7.2.6 *Dietary Fibre and β -Glucan*

The development of cereal with health benefits is a new challenge for breeders. This request, which comes from both consumers and health authorities, focuses on particularly dietary fibre, antioxidants, and phytochemicals (Bedo et al., 2010). Dietary fibre is described as the edible components of plants or analogous carbohydrates that are resistant to digestion and absorption in the human small intestine while fermenting completely or partially in the large intestine (Gebruers et al., 2008).

In the HEALTHGRAIN cereal diversity screening programme, common wheat genotypes with 11.5–18.3 g/100 g had the highest level of dietary fibre compared to durum (10.7–15.5 g/100 g) and spelt (10.7–13.9 g/100 g) wheats, while einkorn (9.3–12.8 g/100 g) and emmer (7.2–12.0 g/100 g) contained the lowest dietary fibres (Gebruers et al., 2008). Similarly, common wheat had the highest dietary fibre content of 12.3%, followed by spelt (10.3%), einkorn (8.7%), and emmer (7.9%) wheats (Løje et al., 2003). Fibre is a critical nutritional component. Most dietary fibre in wheat is closely related to the outer layers of grain. After the milling, the refined wheat flour contained 2.0–2.5% dietary fibre (Khan & Shewry, 2009; de Sousa et al., 2021). Increasing the consumption of flours with high extraction rates or whole-wheat is a nutritional aim in many countries because a high fibre diet is positively related to health benefits.

The most important dietary fibre components are arabinoxylans, which are the most prevalent dietary fibre in terms of quantity, mixed-linkage β -glucans, and cellulose, and nonpolysaccharide compound lignin, which are all cell wall components (Gebruers et al., 2008; Bedo et al., 2010). Similar to total dietary fibre, common wheat genotypes had the largest variation in total arabinoxylan content, ranging from 1.35% to 2.75% of dm. Total arabinoxylan contents of durum, spelt, einkorn, and emmer wheat flours ranged from 1.70% to 2.35% of dm, from 1.60% to 2.15% of dm, from 1.45% to 2.35% of dm, and from 1.40% to 1.95% of dm, respectively (Gebruers et al., 2008).

Soluble arabinoxylan is the most easily fermentable dietary fibre in the colon. The amount of water-extractable arabinoxylan in wheat species differed. The largest variation in water-extractable arabinoxylan content in flour was observed for common wheat (from 0.30% to 1.40% of dm), while a narrow variation (from 0.50% to 0.65% of dm) was determined in einkorn. Durum and spelt wheats exhibited comparable ranges of 0.25–0.55% of dm and 0.30–0.45% of dm, respectively. In the same study, emmer wheat had the lowest value (0.15–0.55% of dm) (Gebruers et al., 2008).

High β -glucan is desirable for increasing health benefits by lowering blood cholesterol levels (Lia et al., 1997). β -Glucans are found as cell wall components of grains (Laroche & Michaud, 2007). It was reported that the genotypic effect was the strongest for β -glucan content by Biel et al. (2016). However, β -glucan content in wheat grain varied dramatically among species, varieties, and environmental conditions, ranging from 0.25% to 1.40% of dm (Marconi et al., 1999; Løje et al., 2003; Gebruers et al., 2008; Biel et al., 2016). Gebruers et al. (2008) found significant

variations in β -glucan content of some wheat grains. The values were 0.25–0.35%, 0.30–0.40%, 0.55–0.70%, 0.50–0.95%, and 0.25–0.45% of dry weight for einkorn, emmer, spelt, common, and durum whole-meals, respectively. On average, einkorn, emmer, and durum wheats contained half the level of β -glucan found in other wheat species.

7.2.7 *Rheological and Pasting Properties of Flour*

The rheology of dough plays a key role in determining the quality of wheat-based products (Kundu et al., 2017). Physical dough analyses like farinograph, mixograph, and alveograph are utilized in various parts of the world to evaluate dough mixing properties (Marchylo & Dexter, 2001). Farinograph is widely utilized to determine especially flour water absorption. Water absorption affects each step of the process, the dough yield, and the quality of the end-product. Water absorption is the amount of water required to make a dough of proper consistency. The contents of protein, damaged starch, and non-starch polysaccharide especially pentosans influence the water absorption of flour. When the dough is mixed, protein may absorb nearly double its weight in water. Even some polysaccharides have ability to absorb more water. Depending on grain hardness, milling conditions, and the desired flour specifications, water absorption may range from 50% to 70% (Miskelly et al., 2010). Comparing some wheat species, the lowest water absorption value was determined as 56.3% from emmer, followed by durum (57.9–61.1%) and common (62.7–64.3%) wheats (Rao et al., 2010). In a study with 24 einkorn lines, grown at 2 different years and locations, farinograph water absorption, stability, development time, and degree of softening values were determined as 52.6–62.8%, 24–450 s, 36–240 s, and 50–300 FU after 12 min, respectively (Corbellini et al., 1999). As compared with the common wheat, einkorn had lower water absorption, shorter dough development time and stability, but higher softening degree, indicating lower tolerance to the development of dough (Piasecka-Józwiak et al., 2015). Zengin (2015) reported that einkorn had higher stability, dough development time, and softening degree, while emmer showed higher water absorption. Farinograph water absorption was related positively with protein content and negatively to SDS sedimentation, as estimated with protein quality (Corbellini et al., 1999). Some einkorn genotypes had high SDS sedimentation values and acceptable farinograph stability (360–720 s) and breakdown (20–50 BU) values similar to those of common wheat (Brandolini et al., 2009). Similarly, Emeksizoğlu (2016) reported that protein content, Zeleny sedimentation, water absorption, softening degree, falling number values, and total yellow pigment content of 30 einkorn genotypes changed 11.2–17.7%, 3.4–18.9 ml, 48.4–63.0, 130–240 BU, 321–400s, and 6.07–11.25 $\mu\text{g/g}$ (LE) of dry matter basis, respectively.

The alveograph is one of the rheological analyses. Alveograph parameters are W (work, related to the area of the curve), L (the length, extensibility), P (pressure,

related to the height of the curve), and P/L (tenacity/extensibility ratio). The alveograph W, in particular, evaluates the gluten strength of dough by measuring the power required to inflate the dough bubble until it ruptures. In a study carried out by Cubadda and Marconi (1994) over 2 years on emmer, spelt, and common wheats, alveograph W values changed between 78 and 118, 116 and 181, and 173 and 190 10^{-4} joule, respectively. In different studies, alveograph energy values varied between 7 and 159 10^{-4} joule for emmer (de Vita et al., 2006) and ranged from 76 to 154 10^{-4} joule for spelt genotypes (Marconi et al., 1999). Castagna et al. (1996) reported that common wheat had a higher energy value (235 10^{-4} joule) than 25 einkorn lines ranging from 9 to 127 10^{-4} joule grown in two different locations. In the same study, einkorn lines had 19–57 mm P, 10–205 mm L, and 0.21–3.30 P/L values. Chopin alveograph gluten strength, farinograph stability, and degree of softening of einkorn are generally poor (Corbellini et al., 1999; Hidalgo & Brandolini, 2019).

Until recently, einkorn has been considered inappropriate for the production of bakery products due to its sticky dough and poor rheological characteristics. Einkorn was mostly utilized to feed animals in recent years, while the threshold durum and common wheats were preferred for human consumption. As a result, during the early history, no selection favouring bread-making attitude was exerted, and a similar tendency was likely maintained afterward. However, breads with bread volumes and features comparable to those of common wheat are occasionally obtained (D'Egidio & Vallega, 1994).

The pasting or gelatinization properties of the starch have an impact on dough stickiness, leavening, end-product, and staling kinetics (Hidalgo & Brandolini, 2019). The amylographic viscosity values of einkorn were higher than those of spelt and common wheats and, in most cases, emmer (Løje et al., 2003). Comparing common and emmer wheats, emmer starch is stated to gelatinise a little later and at a slightly higher temperature than wheat starch. However, the peak viscosity of wheat flour was higher than that of emmer flour (Zaparenko et al., 2020). Brandolini et al. (2008) studied the pasting properties of 65 einkorn samples by rapid viscosity analyzer (RVA). Einkorn samples exhibited higher peak viscosity and final viscosity values compared to modern wheat. The differences were most likely due to the smaller granule size, different grading of einkorn starch granules (Hidalgo & Brandolini, 2019), and lower amylose ratio of einkorn flour (Hidalgo & Brandolini, 2011). In terms of bread colour, einkorn has a lighter colour than common and durum wheats, indicating that einkorn has lower heat damage during baking than modern wheats due to lower α - and β -amylases which restrict the starch degradation (Hidalgo & Brandolini, 2011). The Maillard reactions were minimized during food processing due to the decreased production of reducing sugars in the dough. Besides, low lipoxygenase activity in einkorn dough restricts the degradation of carotenoids (Hidalgo & Brandolini, 2014).

Some factors affect the pasting properties of flour. For instance, water content affects the pasting properties of flour. The importance of starch may differ in a cake system where there is considerably more water present compared to a bread product where water is limiting (D'apollonia & Rayas-Duarte, 1994). Besides water

content, parboiling causes the gelatinization of starch granules and the denaturation of proteins, resulting in significant changes in the flour's technical qualities. There were decreases in the values of SDS sedimentation and viscosity after low-moisture parboilization of einkorn wheat; the changes were more occurred under more severe steaming circumstances (Hidalgo et al., 2008). The technical characteristics of flours are also affected by storage conditions: SDS sedimentation values and viscosity of einkorn flour alter during storage, especially at high temperatures (30 °C and 38 °C) (Brandolini et al., 2010).

7.3 Ancient Wheat-Based Foodstuffs

Einkorn, emmer, and spelt, old-world ancient wheats, were used as the raw material of bread, gruel, porridge, soup, cracked wheat, and beer with regard to the ethnographic and archaeological evidence (Nesbitt & Samuel, 1996). In the last centuries, ancient wheat was replaced by modern wheat cultivars with higher yields, lower prices, and easier milling (Giuliani et al., 2009; Longin et al., 2016). However, in recent years, einkorn, emmer, and spelt have renewed attracting interest for breeders, farmers, bakers, and consumers. Particularly, people want to consume high-quality foodstuffs with increased health and taste properties coming from local farming with lower production intensity. This is the main reason for increasing interest in ancient wheat consumption (Longin et al., 2016).

Due to cultural reasons, different societies have focused on some uses rather than others (Nesbitt & Samuel, 1996). Einkorn and emmer consumptions are generally different in the regions of the countries. However, both are to a great amount utilized for human consumption in addition to animal feed.

Nowadays, ancient wheat species can be processed into a variety of foods including bread, pasta, cookies, crackers, breakfast cereals, snacks, and beverages depending on their appropriateness to produce each of these products (Kulathunga et al., 2020).

7.3.1 Bread

Bread is the most widespread end-product. Each bread type has its own properties, processing conditions, and specific requirements for end-product (Peña, 2002; de Sousa et al., 2021). Bread wheat flours with high protein content have a high water absorption and bread volume potential, and they are suitable for producing breads with good storage quality. And, wheats of less than 11% protein content are usually not suitable for bread-making (Tipples et al., 1994). Einkorn flour is usually considered unsuitable for bread-making owing to its sticky dough and poor rheological characteristics (Abdel-Aal et al., 1997; Boukid et al., 2018); however, einkorn bread is gaining popularity as a speciality bread (Kulathunga et al., 2020). The presence

of einkorn genotypes with high bread-making quality has been reported by several studies (Hidalgo & Brandolini, 2011; Brandolini et al., 2011). Some einkorn breads had a bright yellow colour, and their volumes were similar or better than those of common wheat (Brandolini et al., 2009). A wide collection of einkorn with 1099 accessions was screened, and about 16% of them were found as appropriate for producing bread with sodium dodecyl sulphate (SDS) sedimentation values matching to the bread-making threshold value (>60 ml) (Borghi et al., 1996). Wheats with high SDS sedimentation values generally have good bread-making potential because SDS sedimentation value has a high positive correlation with bread volume (Hammed & Simsek, 2014). Corbellini et al. (1999) obtained some einkorn breads with loaf volume changing from 623 to 918 cm³ and crumb scores comparable to those of common wheat, despite dough-handling difficulties. Einkorn flour bread had the lowest volume; however, the addition of common wheat flour to einkorn flour improved bread volume, structure, and crumb elasticity (Piasecka-Józwiak et al., 2015). Similarly, the effect of adding refined and whole einkorn flour on dough rheological and bread baking properties was studied by Keçeli et al. (2021). Increased einkorn flour level in the blends resulted in lowered sedimentation values, which affected baking quality. The authors stated that einkorn had enough potential to be used in making both refined and whole-wheat breads.

Spelt breads have become increasingly popular in many countries (de Sousa et al., 2021). Wieser et al. (2009) evaluated the bread-making properties of several spelt and einkorn genotypes. Both species had similar water absorption values, while einkorn had lower dough development time values compared to spelt. Concerning the baking test, crumb structure and bread volumes are influenced by genotype. Breads with higher volume were produced from spelt and some of the einkorn genotypes. In another study, the bread-making quality of einkorn and spelt refined flour was determined by Abdel-Aal et al. (1997). Einkorn had the lowest bread volumes, while hard red spring wheat had the highest bread volumes. Hard spelt genotypes had intermediate bread volumes, and soft spelt genotypes had comparable results with hard red spring wheat. Additionally, no significant differences were found among the flour sources in terms of crumb firmness, ranging from 91 to 131 kg in the same study. Similar results were determined by Konvalina et al. (2013) who reported that some spelt genotypes had even better baking quality than common wheat genotypes. Spelt wheat genotypes had higher crumb elasticity but lower crumb cell homogeneity than bread wheat, which is likely owing to its unique dough rheological properties (Callejo et al., 2015). Compared to modern wheats, spelt wheat breads had less total starch, less rapidly digested proteins, and more resistant starch (Bonafaccia et al., 2000). Belcar et al. (2021) compared the ancient wheats in terms of technological and baking properties. Breads produced from spelt and einkorn wheats were of good quality; however, those made from emmer wheat didn't meet the required quality parameters related to organoleptic evaluation and bread crumb. Bread-making optimization studies of spelt wheats were carried out by Callejo et al. (2019). The spelt breads had better quality parameters such as higher bread volume, lower crumb density, higher crumb resilience, and lower firmness although they had higher Gli/Glu ratio and weaker rheological characteristics. The

authors concluded that it was critical to include the recommended techniques for the bread-making process utilizing spelt wheat flour.

There are also studies in the literature testing the suitability of ancient wheats for sourdough bread production. Spelt and emmer sourdoughs showed higher titratable acidity, pH values, free amino acid concentrations, and phytase activity than bread wheat sourdough. In the same study, acceptable breads were produced from spelt and emmer wheats according to sensory analysis. Particularly spelt breads had higher similarity to wheat breads in terms of specific volume and crumb than emmer breads (Coda et al., 2010). Spelt flour was also recommended by Abdel-Aal et al. (1998) for producing two-layer flat bread and sourdough bread.

Bread consumption particularly made with flours of whole-wheat and multigrain is increasing in developed countries. This is mainly because of a growing awareness of the required to minimize simple carbohydrates, fats, and cholesterol intake while increasing complex carbohydrates, protein, and dietary fibre consumption (de Sousa et al., 2021). The effect of adding whole-spelt flour on dough rheological and bread baking properties was studied by Kohajdova and Karovicova (2007). Increased spelt flour level in the blend resulted in improved water absorption and dough development time, which influenced baking and sensory quality. In addition, the most acceptable spelt flour substitution ratio was determined as 15% in the same study. A comparison of spelt cultivars revealed acceptable sensory scores with significant differences among the cultivars, leading to the conclusion that spelt could be a good raw material for bread-making; however, it is dependent on the cultivar used (Korczyk Szabo & Lacko Bartosova, 2013). Similarly, Callejo et al. (2019) reported that differences in bread volume between spelt wheat breads may be due to differences in gluten content and Gli/Glu ratio of cultivars used.

Emmer bread is generally available in Switzerland and may also be found in bakeries in Italy as *pane di farro* (Arzani, 2019) and in Turkey as *kavilca ekmeği*, but only in small quantities. Emmer wheat has poor baking quality because of its lower gluten index (23–39%) and Zeleny sedimentation (14.6–17.6 ml) values (Konvalina et al., 2013). Similarly, Piergiovanni et al. (1996) reported that emmer wheat can be used to produce bread with a lower volume. It has been stated that poor baking quality of emmer compared with common wheat that due to lack of the D group in the genome of tetraploids (Rao et al., 2010). However, there is a large variation in the rheological characteristics of emmer wheat (Grausgruber et al., 2004). Therefore, there are controversial results about the bread-making quality of emmer wheat (Kulathunga et al., 2020).

Durum wheat is mostly used for pasta and bulgur productions. However, there is increasing interest in using durum wheat for bread-making. In a study to evaluate the bread-making potential of different wheat species, common wheat cultivars showed the highest bread volume with 850–927 cc followed by emmer (818 cc) and durum wheat (714–769 cc) cultivars (Rao et al., 2010). Kamut® breads had good sensory characteristics and bread volumes and were quite similar to bread prepared from modern wheat (Pasqualone et al., 2011). It reported that Kamut® is more appropriate than durum wheat for fermentation processes in acidic circumstances,

as shown by an increase in bread volume and the metabolic heat production by yeast (Balestra et al., 2015; Boukid et al., 2018).

Consumption of traditional breads, such as pide, lavash, bazlama, and yufka are on the increase in Turkey. Bazlama, leavened flat bread with creamish yellow colour, has a rounded shape with a usual thickness of 3 cm and width varying from 10 to 20 cm (Başman & Köksel, 1999). It has been stated that einkorn wheat can be used in bazlama production according to colour (L, a, b) and textural properties and sensory analysis (Emeksizozoğlu, 2016). Tortillas made with 60% whole Kamut® flour demonstrated a high resemblance with the 100% wheat flour (Carini et al., 2010).

7.3.2 *Other Oven Products (Biscuits, Cookies, Crackers, Water Biscuits, Cakes, Etc.)*

The primary focus on the end-use quality of ancient wheat has been on its baking quality, and only few studies have examined other possible uses of ancient wheat. Hence, there is little information on the usage of ancient wheat in the production of biscuits, cookies, crackers, etc.

Einkorn genotypes were found suitable for cookie production, on average, having thinner and larger in diameter than those produced from soft wheat flour by Corbellini et al. (1999). Similar results were obtained by Nakov et al. (2018) and Brandolini et al. (2009) for einkorn wheat. There is a negative correlation between cookie diameter and protein content produced from einkorn flour (Corbellini et al., 1999). Water biscuits made from einkorn flour preserved the nutritional quality of their flours better than the modern wheats (Hidalgo & Brandolini, 2011). Therefore, einkorn-enhanced biscuits may provide a novel cereal-based product with health benefits (Kulathunga et al., 2020).

In a comprehensive study, the spread ratio values of cookies produced from 6 einkorn and 13 emmer wheats changed from 6.07 to 8.78. However, emmer wheat flour had lower values, while einkorn had higher values which were similar to high-quality cookie flour. The hardness values of cookies were in the range of 40.8–88.4 N. Besides, most of the genotypes had higher hardness values than that of poor-quality cookie flour. The cookie L^*, a^*, b^* values were in the range of 64.3–73.7, 4.87–11.28, and 27.3–36.3, respectively, by Öztürk et al. (2008). Unlike cookie production, Zaparenko et al. (2020) stated that puff pastries prepared from emmer flour were not inferior in quality to wheat flour-baked products, have a bright crust colour and a regular form, and are large in volume.

Spelt offers a good potential for producing cracker. Spelt crackers exhibited less firm textures, a lower tendency toward shape distortion as seen by lower snapback, a higher cookie factor, and a higher ratio between mean width/length and height when compared to common wheat crackers (Filipčev et al., 2012). Kütük et al. (2008) also reported that ancient wheat flour had similar characteristics as

compared to the reference cracker, made by high-quality soft wheat flour, in terms of weight, thickness, length, width, colour, and texture.

7.3.3 *Pasta and Noodle*

Durum wheat is generally preferred for pasta production. However, pasta made from other wheat species is becoming popular in order to meet consumer demand. Emmer wheat is utilized for pasta production in low amounts in Italy at the domestic level (D'Antuono, 2013). The pastas produced from spelt and einkorn wheats are available in countries such as Germany, Italy, France, and the USA (Brandolini et al., 2018).

Einkorn was determined as a potential candidate for pasta production due to its higher carotenoid level which was preserved during kneading and because of the low enzymatic activity (Hidalgo & Brandolini, 2012). However, there are different results in pasta made from einkorn in terms of cooking quality. Einkorn pasta had significantly lower solid losses into the cooking water. Also, cooking and storage had no effect on the lutein content of pasta (Brandolini et al., 2018). Einkorn pasta, on the other hand, had a less compact structure than durum wheat pasta, which resulted in increased cooking losses and a reduced water-binding capacity (Pasini et al., 2015). Pastas made by substituting 50% of semolina mixtures with einkorn wheat flour had a higher gluten aggregation (la Gatta et al., 2017) and a higher firmness and a lower stickiness than pastas made from other types of flour (Agnello et al., 2016).

Emmer semolina was stated suitable for the preparation of pasta with good organoleptic value and represented a source of resistant starch and fibre; however, pasta processing and cooking lowered the tocopherol and carotenoid contents in emmer pasta (Fares et al., 2008). Similarly, pasta produced with emmer was found to be satisfactory because of its lower stickiness, adequate firmness, and darker colour (Cubadda & Marconi, 1996). Gliadin proteins (ω -35 and γ -45), found in two Indian emmer cultivars, are linked to pasta quality (Buvaneshwari et al., 2005). Pastas made from three emmer wheats were evaluated for their sensorial properties. Cultivar Lucille was found to have the best technical performance, while cultivar Red Vernal had the best texture and the most preferred flavour by the pasta chefs (Kucek et al., 2017). Concerning pasta cooking quality, TOM values for 20 emmer accessions with high protein content ranged from 0.7 to 1.4 g/100 g as a result of protein coagulation happening under high-temperature drying conditions (de Vita et al., 2006).

Spelt wheat can be used for producing good-quality pasta compared to durum wheat cultivar provided that high-temperature drying conditions were applied (Marconi et al., 1999) although ancient wheats are generally thought to have poor technological quality (Brandolini et al., 2008). Marconi et al. (2002) used SDS sedimentation, gluten index values, and alveograph and farinograph characteristics to evaluate gluten properties of three spelt genotypes. Spelt pasta dried under

high-temperature conditions exhibited a darker colour than durum wheat pasta, which might be attributed to higher furosine level, which is likely linked to increased reducing sugars or damaged starch levels in the semolina. TOM values of spelt wheat cultivars changed between 1.1 and 2.0 g/100 g of dry pasta, while durum wheat cultivar had 1.2 g/100 g of dry pasta (Marconi et al., 1999).

The food business is responding to increased consumer demand for convenience by significantly expanding pre-prepared meal alternatives such as ready-to-eat meals (Olsen et al., 2010). Therefore, in a study, whole-grain and refined spelt flour have been used to make new types of precooked pasta products. It has been stated that using the extrusion-cooking technique to prepare spelt pasta results in instant products with better nutritional properties, a high degree of gelatinization, attractive quality, and sensory attributes. Additionally, as compared to refined flour, whole-grain spelt precooked pasta had better nutritional value but lower firmness and increased adhesiveness (Wójtowicz et al., 2020).

Erişte, a kind of noodle, is a traditional food product in Turkey. In a study, the cooking time of the noodles produced from ancient wheat was longer, changing from 6 to 22 min. The highest cooking loss and water absorption values of noodles produced from ancient wheat genotypes were obtained as 5.2% and 175%, respectively, while control noodle had 4.2% cooking loss value. The control noodle had 1.12 g/100 g TOM value, and two of them showed close values (1.40 and 1.54 g/100g TOM) to the control sample, indicating medium quality. All ancient wheat noodles except one had similar swelling volume values compared to that of control noodle (Gümüş et al., 2008). A significant positive correlation ($r = 0.77$, $p < 0.01$) was determined between the protein content and the cooking time of erişte by Emeksizoğlu (2016). The same researcher pointed out that cooking losses of erişte in ten einkorn genotypes changed from 6.51% to 14.48%. Regarding erişte colour, L, a, b values of erişte decreased with the cooking process in the same study (Emeksizoğlu, 2016).

7.3.4 Bulgur

Bulgur is one of the world's first wheat-based products. Bulgur has been produced since the beginning of wheat production in Anatolia and the Middle East. It has been called by different names like burghul, cerealis, arisah, and parboiled wheat throughout history (Bayram, 2007). Popularity and consumption of bulgur have recently increased, even in Western nations, due to its ease of preparation, storability, low cost, and increasing interest in traditional and ethnic foods (Giambanelli et al., 2020).

Turkey is the largest bulgur producer and exporter (Kaplan Evlice & Özkaya, 2020). There is no universal milling system or method for manufacturing bulgur (Bayram & Öner, 2005); however, Antep and Mut (Karaman), named after the major bulgur-producing cities in Turkey, are the two main commercial bulgur production techniques (Stone et al., 2020). The main steps of bulgur production are cleaning, cooking, drying, tempering, debranning, cracking, and size classification.

According to particle size defined by Turkish Bulgur Standard (TS 2284), there are six types of bulgur. The first four are coarse bulgur types (pilavlık), while the last two are fine bulgur types (köftelik) (Kaplan Evlice & Özkaya, 2020).

The Antep and Mut (Karaman) systems, the two most popular, are essentially different. The tempering process before debranning is the main difference between the Antep and Mut (Karaman) techniques. The Antep production technique has a relatively short tempering time (15–20 min), resulting in moisture content of 15–17% in the bulgur; however, the Mut (Karaman) method requires a longer tempering time (10–14 h), allowing the moisture content to rise to 20–24% (Bayram & Öner, 2005; Yousif et al., 2018). In either case, tempering the cooked and dried grains helps the separation of bran in the debranning step. Debranning and milling are performed simultaneously in the Mut (Karaman) system traditionally utilizing a stone mill, while the milling is performed on the debranned bulgur in separate processing steps in the Antep technique (Unal & Sacilik, 2011). The colour, shape, and size of the bulgur particles are affected by distinctions in the two bulgur production procedures (Bayram & Öner, 2005). The Antep technique is more widely used (70% of the total world's bulgur) since the Mut (Karaman) method is more difficult to obtain a high-quality end-product, partly owing to the higher moisture requirements (Bayram & Öner, 2005; Stone et al., 2020).

Bulgur can be prepared from different grains such as oat, barley, rye, soybean, corn, triticale, chickpea, and common bean; however, most of the bulgur produced and consumed in the world is wheat bulgur. Bulgur is commonly produced by using durum wheat in Turkey. Einkorn and emmer are used to produce bulgur especially in some parts of Turkey. Processing and consumption of einkorn wheat bulgur in Turkey are still predominantly local, with only preliminary efforts at commercial manufacturing (Giambanelli et al., 2013). Each year in Ihsangazi (Kastamonu, Turkey), a special festival is held to present einkorn bulgur.

Einkorn bulgur is rather different than durum wheat bulgur. Einkorn bulgur has darker colour, specific taste, and flavour compared to durum wheat bulgur (Fig. 7.1a, b) (Hendek Ertop, 2019). Einkorn bulgur production is still being carried out with



Fig. 7.1 (a) Einkorn bulgur produced by traditional method (left) (b) Durum bulgur produced by industrial method (right) (A Kaplan Evlice)



Fig. 7.2 (a) Removing foreign substances and stones from einkorn wheat, in Bolu Seben (left) (HM Yaman) (b) Boiling einkorn wheat in Bolu Seben (right) (HM Yaman)



Fig. 7.3 (a) Ended boiling of einkorn wheat in Kastamonu İhsangazi (left) (HM Yaman) (b) Boiling wheat is poured into the sieve and cooled with cold water (right) (HM Yaman)

traditional methods. The steps of einkorn bulgur production are cleaning (Fig. 7.2a), cooking (Figs. 7.2b and 7.3a), drying (Figs. 7.3b and 7.4a), husking, and crushing (Fig. 7.4b). Einkorn bulgur is ground by a stone mill and has a larger visual size than durum bulgur, and its shape is known “fly wing” in the area (Fig. 7.1a) (Hendek Ertop, 2019).

Images in work areas with bulgur-making for ancient wheat;

There is little information about ancient wheat bulgur in the scientific literature. Giambanelli et al. (2020) reported decreases in carotenoids, tocol, and phytosterol contents and an increase in the free phenolic compound in einkorn bulgur cooked in three different cooking times (2, 3, and 4 h) concerning raw einkorn kernels. Hendek Ertop (2019) compared commercial and traditional bulgurs produced from einkorn and durum wheats. Einkorn bulgur was found to be rich in the contents of ash, fat, and trace and major elements particularly in terms of Fe, Zn, and Al. The bulgur production technique had no effect on the physicochemical properties, while the wheat species had an impact on them. Einkorn bulgur was darker than durum



Fig. 7.4 (a) The wheat is dried for 4–5 hours to be slightly moist (left) (HM Yaman) (b) Separation of milled einkorn, by the electrical stone mill, into parts of chaff, dehulled, and broken grains in Kastamonu, İhsangazi (right) (HM Yaman)

bulgur. The traditional form and size of einkorn bulgur were determined to be unsuitable with the Turkish Bulgur Standard (TS 2284). The einkorn bulgur size was bigger than the durum bulgur size since the einkorn bulgur grain was divided into two from the centre in accordance with its traditional shape by the stone mill. The findings of Yilmaz and Koca (2017) showed that a considerable amount of antioxidant capacity was kept after bulgur production for einkorn and durum wheat. Total phenolic content determined in einkorn was higher. However, it was subjected to higher loss during the production of bulgur than durum wheat. Total yellow pigment content determined was twofold higher in einkorn bulgur than in durum bulgur. Ferulic acid was found the most abundant phenolic acid, while chlorogenic acid was determined as the least phenolic acid for both wheats. During the bulgur production, the maximum losses were recorded in the soluble-conjugated phenolic acid fraction for einkorn and the soluble-free phenolic acid fraction for durum. The same author also evaluated the bioactive components and antioxidant activity of emmer bulgur produced using different cooking (traditional, autoclave, and microwave) and drying (hot air and microwave) methods. When compared to raw emmer wheat, antioxidant activity and bioactive compounds declined in all bulgur cooking and drying methods. In bulgur samples, soluble-free and soluble-conjugated phenolic acids are more decreased than insoluble-bound phenolic acids because of the better thermal stability of insoluble-bound fraction. The microwave cooking method had a positive effect on preserving natural bioactive compounds, while the microwave drying method had a negative effect (Yilmaz, 2019). It is a good option for increasing emmer usage by-product diversity. Messia et al. (2012) used hydrothermal treatment to produce parboiled emmer, which was recognized as a novel product with high nutritional and cooking characteristics.

7.3.5 Novel Products (*Seed Puffing, Flakes, etc.*)

Puffed cereals are ready-to-eat foods that are extensively utilized to produce other breakfast products, snacks, etc. (Hidalgo et al., 2016; Kulathunga et al., 2020). Hidalgo et al. (2016) assessed the technological qualities and chemical compositions of puffed einkorn and common wheat grains. The technological properties of puffed common wheat kernels seem to be more attractive due to their higher expansion rate and related attributes like test weight, surface area, and water absorption compared to puffed einkorn kernels. The puffed einkorn kernels, on the other hand, were rich in protein and bioactive compounds such as tocopherols and carotenoids and low in starch when compared to puffed common wheat kernels.

Hard spelt wheat can be used to produce flaked breakfast cereal with attractive flavour and texture (Abdel-Aal et al., 1998). When compared to commercial products, Kamut® and spelt flakes and muesli exhibited acceptable sensory characteristics (appearance, consistency, and flavour), as well as the highest flavonoid, total phenolic, and crude fibre contents (Sumczynski et al., 2015). Flaked einkorn and spelt products have less lysine and more protein than their flours (Abdel-Aal & Hucl, 2002). The addition of spelt flour (5%, 10%, 15%) to corn grits reduced the fracturability and expansion ratio while increasing the hardness and bulk density of extrudates (Jozinović et al., 2016). Furthermore, colour altered, the hot and cold peak viscosities decreased, and there was less retrogradation.

There have been efforts to use spelt wheat for producing frozen bakery products (Šimurina, 2011). In comparison to the dough without spelt flour and with sucrose, substituting 60% of wheat flour with spelt flour and adding sorbitol as a sucrose substitute increased frozen dough yield and reduced baking loss (Kozłowicz & Kluza, 2009).

Einkorn flour has been found as a promising raw material for producing baby foods (Zaharieva et al., 2010). Einkorn flour has a high nutritional value in terms of protein and antioxidants, as well as excellent pasting characteristics, which makes it appropriate for use in the preparation of specialty products and baby foods (Brandolini et al., 2008).

Spelt wheat, particularly green (immature) grain, might be a valuable substrate for the production of value-added fermented foods. The solid-state fermentation (30 h) of spelt and green spelt with *Rhizopus oligosporus* resulted in value-added tempe with improved nutritional composition and bioactive parameters (Starzyńska-Janiszewska et al., 2019).

7.4 Conclusions

Ancient wheats are very important in terms of food security under low input, low yield, and high-stress conditions. The ancient wheats have a wide genetic diversity, which is used to develop modern wheat cultivars. They are naturally suited to

organic and environmental friendly farming systems. Additionally, they are appropriate for areas with high local demands. However, because of their low yields, ancient wheats would not be suitable for food security and the mass market.

Wheat is a good source of health-related components, especially phytochemicals and antioxidants, as well as the main components of protein, carbohydrate, and lipid. In terms of various nutritional components, there are no significant differences between ancient and modern wheats. The majority of the health-promoting components are mainly located in the germ and aleurone layers of the grain. Consuming wheat as whole-grain products may allow getting maximum benefit from these components. Therefore, it will be more beneficial for health to consume wheat as a whole-grain instead of separating it as modern wheat or ancient wheat.

There are some data currently accessible on the usage of ancient wheat in the production of bread, cookies, pasta, etc. Besides bakery products, ancient wheat seems a promising material for manufacturing new or special foods such as snacks, baby foods, etc. However, further detailed researches are needed to determine the end-product quality of ancient wheat.

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Chapter 8

From Hologenomes to Biofertilizers in Wheat Production



Ömer Can Ünüvar and Ercan Selçuk Ünlü

8.1 Introduction

[...] Then the woman said, “Enkidu, eat bread, it is the staff of life; drink the wine, it is the custom of the land.” So, he ate till he was full and drank strong wine, seven goblets. He became merry, his heart exulted, and his face shone. He rubbed down the matted hair of his body and anointed him- self with oil. Enkidu had become a man [...]. (The Epic of Gilgamesh (Sandars, 1964))

The *Epic of Gilgamesh* has told us that existence of the bread has come to show the differences between barbarous and civilized human beings. The dependence of Enkidu, who is the main character in the epic, on wilderness to live has been ended by bread. According to the epic, bread is a gate that opened the agrarian civilization (Salevurakis & Abdel-Haleim, 2008).

From the existence, feeding of the individuals and/or populations has been important for surviving. Therefore, domestication of plants and animals has played an indispensable role in the development of civilization. The domestication of plants and animals have begun separately in different regions of the earth. The Fertile Crescent, some areas of the Yangzi and Yellow River basins of China, and Mesoamerica have strong arguments for archaeological priority in the development of the domestications (Bellwood, 2019).

The demand for food is expected to increase nearly 40% in 2030 due to the growing population. According to the reports, agricultural zones would stabilize at 1.5 billion hectares, and the yield would increase 2% for each year, but the increase in the yields has been represented as 0.9 over the last decade (Dixon et al., 2009; Movahedi et al., 2009). These statistical predictions and related results are getting worse because of climate change and its effects, especially in Europe. Because of

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increasing food requirement globally, there are many traditional and modern techniques to improve the quality and the quantity of the crops.

In this chapter, we focus on biofertilizers. As known, in parallel with population growth and its demand, the use of fertilizers has been increasing. Fertilizers are one of the most common chemicals. Uncontrolled usage of chemical fertilizers affects the soil negatively by causing the aggregation of heavy metals, nitrogen, phosphorus, etc. The heavy metals can be absorbed by plants and may lead them to enter the food web. High nitrogen-rich fertilizers cause excessive nitrogen accumulation in the soil, and the accumulation may cause production of carcinogenic nitrosamine by plants. Excessive and uncontrolled usage of the chemical fertilizers also affects water supplies by accumulation of the chemicals in water reserves causing related complications such as eutrophication. Fertilizers are reported as one of the main reasons for groundwater pollution (Evans et al., 2019; Savci, 2012; Sönmez et al., 2008). From the ecological perspective, the detrimental impact of chemical fertilizers rises from the ignorance of the communities in interaction among plants and their surrounding habitats before taking any action.

8.1.1 Hologenome Concept and Wheat

Interaction is a well-known phenomenon among living entities. While this interaction is more intense for some species (e.g., humans), its effectiveness becomes more prominent only for certain times of the life cycles for some organisms. The type and extent of the interaction are directly related to the evolutionary position of the species and the necessary environmental conditions for their survival. At this point, the interaction can occur not only between the same or similar species but also between different species. In fact, the importance of this interaction becomes more interesting and important as the evolutionary distance between species widens. The interactions among the same species are likely related with the mechanisms to overcome challenges (e.g., reaching nutritional sources, defending the population or the colonies), while it turns into a symbiotic manner among distant species. The interactions between microorganisms and plants can be accepted as the most fundamental example for symbiotic relationship since the impact of the synergy is not limited to interacting microorganisms and plants but also the whole ecosystem. While the organism can benefit from obtaining essential nutrients or providing extended defense mechanisms, they also contribute to the ecosystem by changing the environmental conditions for all surrounding organisms. Fixation of atmospheric nitrogen or solubilizing several minerals by root-colonized bacteria can improve the nutritional status of the host plants, while other plants can benefit from the changes around them. In addition, since the interaction likely changes the mineral and organic content and the pH of the soil, free-living microorganisms and other plant species found in the niche will have to adapt themselves depending on the outcomes of the interactions. Million years of co-evolution of the species indeed impacted the dynamics of these relationships making it more complex than we probably anticipated (Rosenberg &

Zilber-Rosenberg, 2011). The concept is well defined under the term “hologenome theory of evolution” which basically embraces the plants and associated microorganisms as the holobiont where the net genetic information of the plant host and its microbiota is covered under hologenome (Fig. 8.1) (Zilber-Rosenberg & Rosenberg, 2008).

Selection is one of the driving forces for defining the assembly patterns in the hologenomes. Several factors can influence the dynamics of the selection process determining or shifting the assembly of the hologenome. While these influences can be minimal, some changes may influence the assembly that can lead even to the extinction of some species such as dramatic changes in climate conditions or agricultural practices (Dini-Andreote & Raaijmakers, 2018).

While human influences spread, its impacts on ecology have been rising due to unattended applications of diverse technologies. Agricultural practices undoubtedly have the most influence from this regard especially for domesticated crops. In specific, wheat has been influenced by agricultural practices as being one of the earliest domesticated crops and as it has been planted for generations in diverse continents by diverse agricultural practices.

The study conducted on hologenome of wheat species with diverse genotypes (A, B, D, AB, and ABD) showed that there are minimal differences in core hologenome among different wheat species compared to wild relative species. The data suggest that during the evolution of wheat from ancient species to modern cultivars, wheat species have preserved their core microbiome assemblies. The difference, on the other hand, rises when compared to root-associated genomes (Bulgarelli et al., 2015; Cantor, 2018). Studies suggest that the influence on the stability for certain types of bacteria or fungi is directly related with the impact of the organisms in symbiotic relationship (Banerjee et al., 2019; Cantor, 2018).

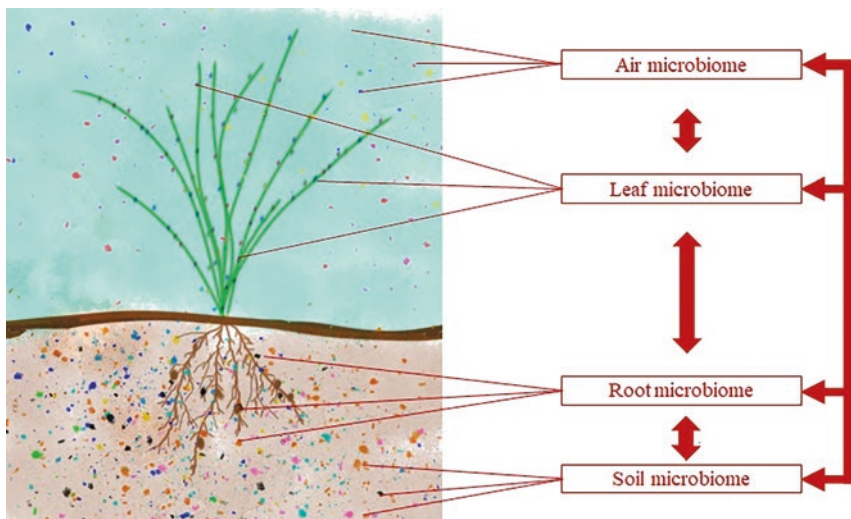


Fig. 8.1 Schematic representation of plant hologenome

Considering the importance of hologenomes in plant evolution, a lot of researchers are studying alternative and sustainable approaches for integrating the crucial microorganisms to agricultural applications. One of them is biofertilizer usage which is based on the implementation of plant growth-promoting microorganisms especially bacterial species to agricultural practices.

8.1.2 A View to Current Status of Biofertilizer Research

Before we start explaining in detail, it is better to provide a precise definition of biofertilizer. If you search the meaning of the term in the dictionaries, you do not find any explanation. There are some confusing explanations about biofertilizers that came from terms of organic fertilizers and biofertilizers. The scientific literature is clear about that. Biofertilizer is defined as the liquid or solid mixtures which involve living microorganism(s) that colonize the rhizosphere or other parts of the plants and promote growth and development of the host plants (Vessey, 2003).

Before giving information about the mechanism of action of biofertilizers, we would like to share the statistical data of the studies conducted over the years. Among the studies about “biofertilizers”, 83.657% were published as articles, 10.304% as reviews, 5.532% as proceeding papers and 4.519% as book chapters until 2021 according to ‘Web of Science’ analysis results. The publication trends are represented in Fig. 8.2 from 2000 to 2021. As it can be seen in Fig. 8.2, publications involving biofertilizers are increasing rapidly from year to year. These data show that scientific interest in biofertilizer-related topics is increasing.

The first 25 countries are represented in Fig. 8.3; we can clearly see that India, Brazil, and China are the more interested countries in the biofertilizer-related studies. This may be one of the indications why India is a pioneer in the global biofertilizer market. Before concluding the general data about the studies, let’s show which “Web of Science” categories were mainly focused in 2368 published studies. In Fig. 8.4, the percent distribution of the published categories is represented.

8.2 Biofertilizers and Wheat Farming

The usage of fire to clean forest areas resulted into the first important anthropogenic effect on the environment. Our ancestors were able to access easily herbivores in the savanna and near lands. Also, they suppressed the growth of less needed plants to use as forage and feeding. But it was the only beginning. Day by day, the usage of the soil as a supply has increased from the Neolithic period (Pyne, 2019; Wrangham, 2009). Increase in the population, climate change, and selecting/protecting desirable plants have caused sharp changes in the relation between the earth and humans. Earth is no longer as fertile as it used to be.

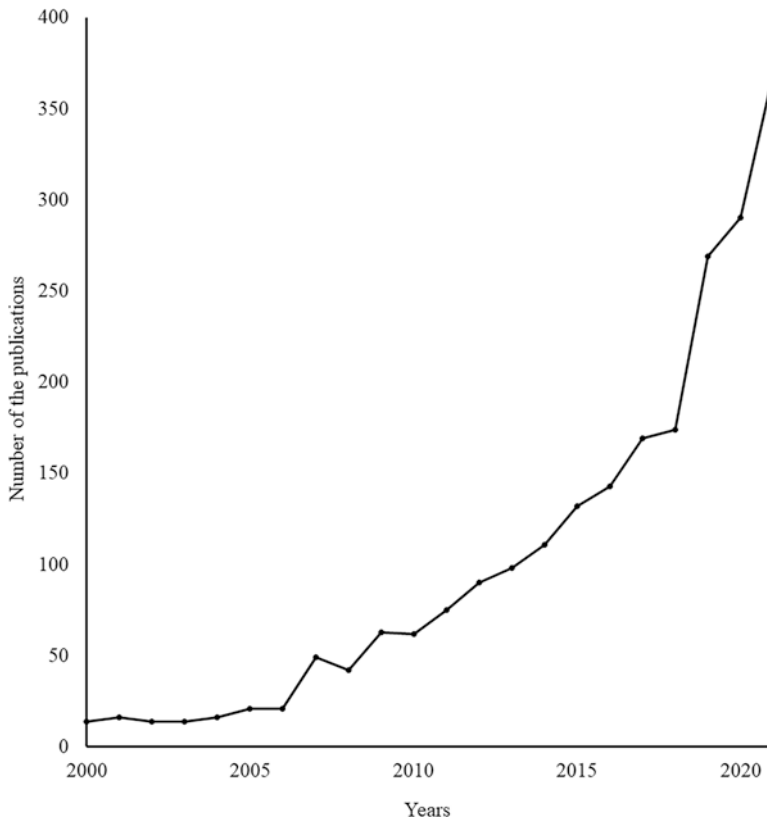


Fig. 8.2 The distribution of the publications between 2000 and 2021. (The data were taken from www.webofscience.com, 28.02.2022)

The fertility of the soil is related with the nutrients used for plant growth. Natural cycling of nutrients plays important roles in this phenomenon. The cycling provides and helps to stabilize the amount of the essential nutrients for plant growth in the soil. The common essential macro- and micronutrients are summarized below.

Primary macronutrients as nitrogen (N), phosphorus (P), and potassium (K); secondary macronutrients as calcium (Ca), magnesium (Mg), and sulfur (S); and micronutrients as iron (Fe), manganese (Mn), zinc (Zn), copper (Cu), boron (B), molybdenum (Mo), chlorine (Cl), and nickel (Ni) are listed as mineral elements for plants. On the other hand, carbon (C), hydrogen (H), and oxygen (O) are non-mineral essential elements used in plant growth (Parikh & James, 2012). Even though these chemical ingredients are analyzed for assessment of soil quality for agriculture, it is usually neglected to monitor whether the soil microbiome is suitable enough for plant adaptation. Along with many plant species, several bacteria have been isolated and characterized with plant growth promoting mechanisms for wheat growth and development which are directly related with the hologenome

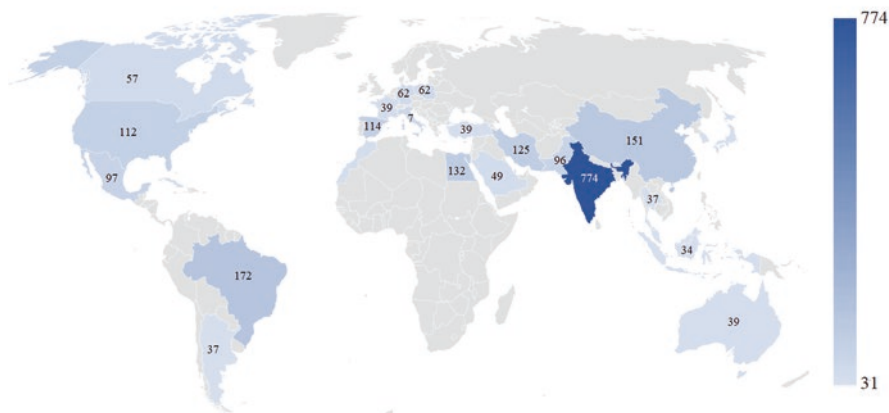


Fig. 8.3 The distribution of the publications by countries. (The data were taken from www.webof-science.com, 28.02.2022; 2368 publications selected from Web of Science Core Collection)

assemblies. Table 8.1 represents some of the most common bacteria that are related with increasing the yield and crop quality in wheat farming.

The following section summarizes some of the parameters related with increasing the quality of soil regarding usage of biofertilizers.

8.2.1 Nitrogen

Especially, nitrogen, which is one of the essential parts of proteins, plays important roles in the catalyzation, transportation, photosynthesis, etc. Promoting the growth of vegetative parts of plants and triggering the growth of roots mainly require the use of nitrogen (Bloom, 2015; Hemerly, 2016). The shortage of nitrogen causes some important effects on plants such as unhealthy growth, color change of leaves from green to yellow (chlorosis), red-purple spots on leaves, and lateral bud growth (Bianco et al., 2015). Deficiency in nitrogen can cause reduction in crop yield and quality, but it is not to be forgotten that excessive amount of nitrogen usage affects plants negatively. In addition, excessive nitrogen usage with fertilizers is an important problem for the environment. Uncontrolled usage of N in agricultural processes may cause accumulation of N in plants which affects human health and ecology as a consequence of methemoglobinemia and groundwater pollution (Elhanafi et al., 2019; Faris, 2014; Peng et al., 2011).

The more dominant inorganic forms of nitrogen in soil are ammonium (NH_4^+) and nitrate (NO_3^-) forms; on the other hand, there are lesser forms as nitride (NO_2) and nitrous oxide (N_2O). Plants commonly use nitrogen in solely ammonium and nitrate forms; other forms especially nitrites are toxic for plants. There are several ways to convert non-available forms of nitrogen to corresponding organic or

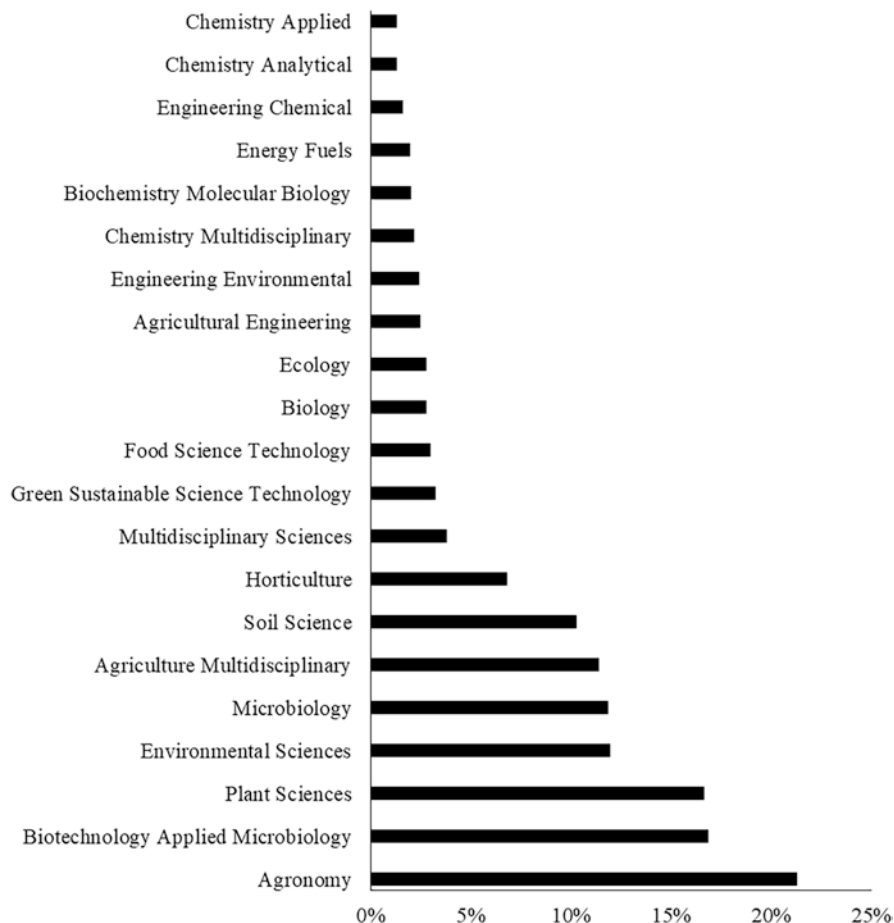
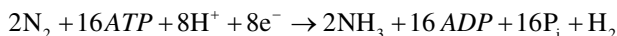


Fig. 8.4 Percent distribution of the publications by fields. (The data were taken from www.webof-science.com, 28.02.2022; 2368 publications selected from Web of Science Core Collection)

inorganic compounds available for plants. Fixation occurs via atmospheric events as lightning, industrially by chemical methods, or biological processes through microorganisms.

In the biological processes, atmospheric dinitrogen (N_2) gas is converted to ammonia and hydrogen gas by electron transfer reactions. This biosynthetic process is called biological nitrogen fixation (BNF). The reaction is given as,



The reaction is occurred by several symbiotic and/or free-living bacteria that carry nitrogenase enzyme complex (with two protein components as dinitrogenase reductase and dinitrogenase). Two electrons are donated by dinitrogenase. The

Table 8.1 List of species with growth promoting effects on wheat

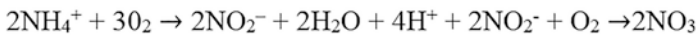
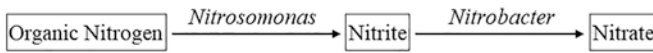
Mechanism	Species name	References
Nitrogen fixation and accumulation	<i>Acinetobacter calcoaceticus</i>	Sarode Prashant et al. (2009)
	<i>Azorhizobium caulinodans</i>	Sabry et al. (1997)
	<i>Azospirillum brasilense</i>	Saubidet et al. (2002)
	<i>Azotobacter chroococcum</i>	Narula et al. (2006)
	<i>Azotobacter chroococcum</i>	Narula et al. (2006)
	<i>Bacillus firmus</i>	Barneix et al. (2005)
	<i>Bacillus megaterium</i>	Abd El-Razek and El-Sheshtawy (2013)
	<i>Bacillus simplex</i>	Barneix et al. (2005)
	<i>Bacillus subtilis</i>	Ünüvar et al. (2022)
	<i>Klebsiella pneumoniae</i>	Iniguez et al. (2004)
<i>Paenibacillus polymyxa</i>	Timmusk et al. (1999)	
Phosphate solubilization	<i>Acinetobacter calcoaceticus</i>	Sarode Prashant et al. (2009)
	<i>Azotobacter chroococcum</i>	Narula et al. (2006)
	<i>Bacillus megaterium</i>	Abd El-Razek and El-Sheshtawy (2013)
	<i>Bacillus subtilis</i>	Ünüvar et al. (2022)
	<i>Pseudomonas fluorescens</i>	Shahzadi et al. (2013)
	<i>Pseudomonas jessenii</i>	Mäder et al. (2011)
	<i>Pseudomonas synxantha</i>	Mäder et al. (2011)
Hormone production	<i>Pseudomonas putida</i>	Egamberdieva and Kucharova (2009)
	<i>Acinetobacter calcoaceticus</i>	Sarode Prashant et al. (2009)
	<i>Azospirillum brasilense</i>	Barbieri and Galli (1993)
	<i>Azotobacter chroococcum</i>	Kumar and Narula (1999)
	<i>Bacillus subtilis</i>	Ünüvar et al. (2022)
	<i>Paenibacillus polymyxa</i>	Timmusk et al. (1999)
	<i>Pseudomonas aurantiaca</i>	Egamberdieva and Kucharova (2009)
	<i>Pseudomonas brassicacearum</i>	Magnucka and Pietr (2015)
	<i>Pseudomonas chlororaphis</i>	Egamberdieva and Kucharova (2009)
	<i>Pseudomonas denitrificans</i>	Egamberdiyeva (2005)
	<i>Pseudomonas extremorientalis</i>	Egamberdiyeva (2005)
	<i>Pseudomonas fluorescens</i>	Egamberdieva and Kucharova (2009)
	<i>Pseudomonas jessenii</i>	Mäder et al. (2011)
	<i>Pseudomonas rathonis</i>	Egamberdiyeva (2005)
<i>Pseudomonas synxantha</i>	Mäder et al. (2011)	

(continued)

Table 8.1 (continued)

Mechanism	Species name	References
Siderophore	<i>Acinetobacter calcoaceticus</i>	Sarode Prashant et al. (2009)
	<i>Pseudomonas jessenii</i>	Mäder et al. (2011)
	<i>Pseudomonas synxantha</i>	Mäder et al. (2011)

Adapted from Çakmakçı et al. (2017)

**Fig. 8.5** Nitrogen conversion via bacteria

dinitrogenase part that contains Fe-S center keeps the electrons before donation. Finally, dinitrogenase catalyzes the reduction of dinitrogen gas (N_2). After the fixation procedure, the fixed form can be oxidized or used by other organisms (Hanrahan & Chan, 2005)

Some microorganisms can convert nitrogen to several organic or inorganic compounds. The conversion of nitrogen to its inorganic forms is known as mineralization. In this process, firstly complex proteins are broken down to their amino acids, amides, and amines by primary heterotrophs. This part is called as aminization. The step which occurs after aminization is called ammonification in which amino groups are converted to ammonium by primarily autotrophic microorganisms.

Other conversion ways of the nitrogen compounds in nature are called nitrification and denitrification. In nitrification process, *Nitrosomonas* and *Nitrobacter* species are responsible. *Nitrosomonas* species convert ammonium to nitrite, and the second part of the process occurs via *Nitrobacter* species that convert nitrite form of nitrogen to nitrate (Fig. 8.5).

Environmental factors such as temperature, soil moisture, aeration, acidity, etc. affect mineralization and nitrification. For instance, nitrification reactions are slow at cold temperatures, and the rate increases with temperature rising until bacterial viability.

In denitrification, nitrates are converted to gaseous forms of nitrogen as nitric oxide, nitrous oxide, and dinitrogen by anaerobic bacteria that use nitrate in respiration under oxygen-limiting conditions. Denitrification changes with temperature and amount of available organic matter in soil. This nitrogen loss is seen clearly in Table 8.2.

The other part of the nitrogen cycle is ammonia volatilization, in which ammonia is lost to the atmosphere because of pH and urea decomposition or hydrolyzation enzymatically to ammonium in soil.

Soluble nitrogen compounds can be leached from soil, and it is called nitrogen leaching in the cycle. In addition to this, there are unavailable forms of the nitrogen to plants. In that process plant residues are converted to inorganic nitrogen forms

Table 8.2 Nitrogen loss changes in saturated soil with temperature

Time (day)	Temperature (°C)	Percent nitrogen loss
3	24–27	6
5	13–16	10
10	13–16	25

that is called immobilization. Environmental situations also affect the reactions especially carbon-to-nitrogen ratio (*Forms of Nitrogen in the Soil | Soils – Part 5: Nitrogen as a Nutrient – Passel, n.d.*)

In several studies, the impact of plant growth-promoting bacteria was studied on wheat. The studies show that nitrogen availability can be promoted via several microorganisms such as *Azospirillum* sp. (Ganguly et al., 1999), *Acinetobacter* sp. (Rana et al., 2011), *Bacillus subtilis* (Ünüvar et al., 2022), and *Azotobacter* (Kumar & Narula, 1999).

8.2.2 Phosphorus

It is the second most important and limiting element for plants with a role in the building of key molecules such as nucleic acids, phospholipids, and adenosine triphosphate (ATP). Plants need phosphorus for survival; it is also used in enzymatic reactions and metabolic pathways. Visual symptoms can be seen after the early stage of growth of related plant in the case of phosphorus deficiency. The deficiency decreases in photosynthesis efficiency and energy investment. On the other hand, the excessive amount of phosphorus causes environmental problems as in excessive amount of nitrogen such as eutrophication (Correll, 1998; Smith, 2003; Theodorou & Plaxton, 1993).

Phosphorus compounds are cycled via rocks, water, soil, sediments, and organisms. Rain and watering cause the release of inorganic phosphate ions into water reserves and soil where it becomes accessible for plants. After the death of the plants and the animals that feed on them, the organic phosphate is returned to the soil. In addition to inorganic forms, organic forms of phosphate are converted to inorganic forms by another mechanism which involves microorganisms. The breakdown process of organic form to inorganic form is called mineralization. Most of the phosphorus is in the sediments and rocks where they are not available for plant intake. Overall, weathering, mineralization, and desorption increase availability of phosphorus for plant, while immobilization, precipitation, and adsorption reduce the plant-available forms of the phosphorus compounds. As aforementioned, several bacterial species have the ability to solubilize inorganic and organic phosphorus forms (Khiari & Parent, 2005; Rodríguez & Fraga, 1999). The released organic acids, which are mainly gluconic and keto-gluconic acids, by phosphate-solubilizing bacteria solubilize phosphate compounds by lowering pH, cation chelation, and

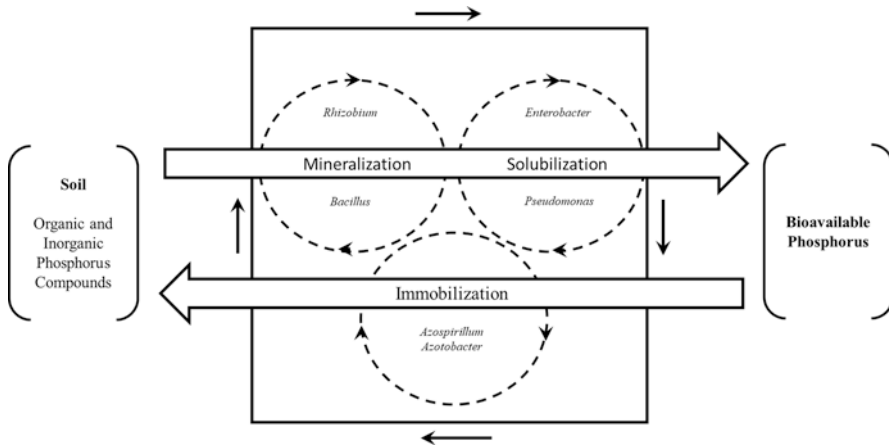


Fig. 8.6 Diagram of phosphorus mobilization in soil by bacteria

adsorption (Nahas, 1996). In Fig. 8.6, phosphorus mobilization is represented with the most common bacterial genera.

One of the most promising PGPB for increasing the availability of phosphorus-related compounds are *Bacillus* sp. and *Pseudomonas* sp. for wheat growth (Afzal et al., 2005; Chen et al., 2006; Üniüvar et al., 2022). These species are usually capable of producing hormones in addition to induction of phosphorus-related compounds' availability. Thus, using PGPB to increase phosphate solubility not only promotes the biosynthesis of several important molecules but also promotes the growth (e.g., root elongation) via the synthesis of auxins that is the one of the major plant hormones.

8.2.3 *Phytohormones*

Growth and development of plants are not only affected by nutrients as nitrogen and phosphorus but also hormones and other metabolites. Most of the communications between plant cells occur via plant hormones that are produced by plant cells or/and rhizobacteria. Indole-3-acetic acid (IAA) is the most abundant phytohormone which is produced in shoots. Production of IAA is common by microorganisms that are living around the rhizosphere of plants. The similarity between the production mechanism of IAA in plants and bacteria is high when they both use tryptophan amino acid as the precursor in IAA biosynthesis pathway.

Two main pathways are responsible for the bacterial production of IAA as tryptophan-dependent and tryptophan-independent. In Fig. 8.7, the pathway of the trp-dependent synthesis of IAA in gram-negative bacterial species is represented. There are five different ways to synthesize IAA as indole-3-acetamide (IAM) pathway, indole-3-pyruvic acid (IPyA) pathway, tryptophan side-chain oxidase (TSO)

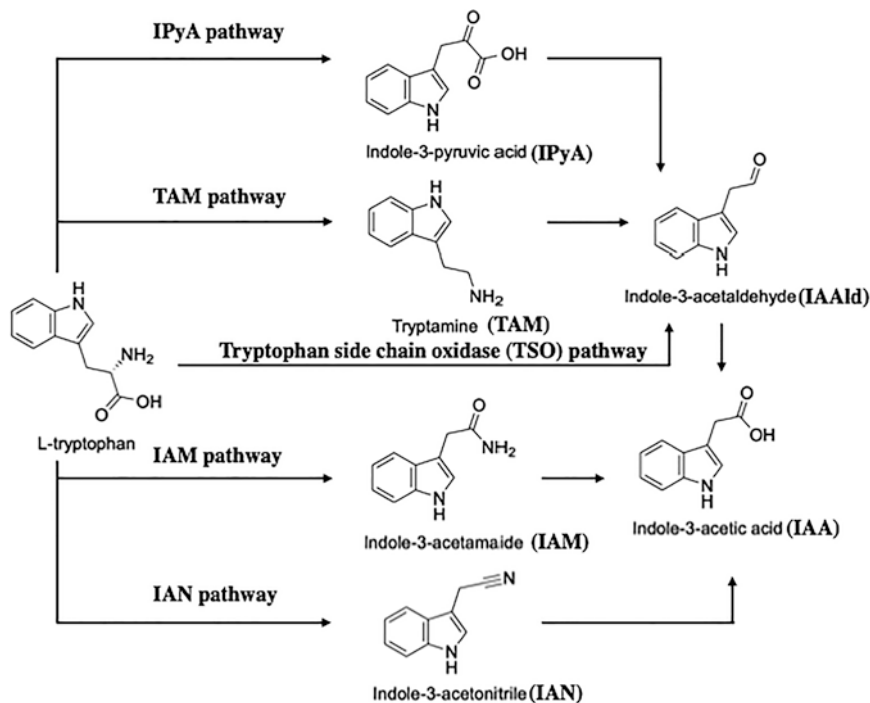


Fig. 8.7 Trp-dependent pathways of the synthesis of indole-3-acetic acid by gram-negative bacterial species

pathway, tryptamine (TAM) pathway, and indole-3-acetonitrile (IAN) pathway (Keswani et al., 2020). On the other hand, trp-independent path is not clear yet (Persello-Cartieaux et al., 2003).

Plant growth-promoting bacteria (e.g., *Pseudomonas* sp., *Azospirillum* sp., *Bacillus* sp. (Akbari et al., 2007; Ünüvar et al., 2022)) improve the yield and effect wheat growth via direct and indirect mechanisms. There are several additional effects of these species on plants such as siderophore synthesis, antimicrobial activity against pathogens, aminocyclopropane-1-carboxylic acid (ACC) deaminase activity, the solubilization of potassium and zinc compounds, etc. Also, they support the host plants against different stress conditions as drought, temperature, salinity, and heavy metals (Çakmakçı et al., 2017; Ramadoss et al., 2013).

Climate crisis and uncontrolled usage of chemicals create a pressure on nature. In conclusion, the importance of optimization of the chemical fertilizer usage has become even more evident. Adapting new approaches by combining conventional methods with biofertilizer applications would provide a reasonable yield increase along with sustainability in agriculture.

8.3 Conclusions

Biofertilizers are one of the most promising approaches for sustainability of agricultural activities. They can be the most eco-friendly and natural alternative for promoting plant growth and increasing crop yield and soil quality in agricultural practices. The impact of chemical fertilizers in soil not only affects the soil but every interacting ecological element connected to farmlands. Applications of biofertilizers containing plant growth-promoting bacteria for wheat farming will provide a sustainable agricultural approach. Using chemical fertilizers for ancient wheat can result into yield loss due to overgrowth of the plant that became vulnerable to environmental phenomena like high wind, rain, hailstorm, and/or frost. Biofertilizers will enhance plant nutrition while providing more control on plant growth at more natural level which in turn reduces the risk of losing yield due to harsh environmental conditions. Hologenome-associated biofertilizers should be the main criteria for PGPB applications since it will increase the efficiencies of biofertilizers, and it will help preserving the place of the wheat in ecology.

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Chapter 9

Wild Relatives and Their Contributions to Wheat Breeding



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9.1 Current Status in Wheat Breeding

Wheat (*Triticum* spp.) is an important stable crop for humankind and a good source of protein and energy consumed by the world population. Since its domestication (ca. 12,000 years ago), wheat has played a critical role to feed the human population. In addition to its importance as food, it can be grown across different regions, from sea level to 4570 m.a.s.l., of the world. Common or bread wheat (*Triticum aestivum* L.) covers the vast majority of the wheat production area at a global level. Durum wheat (*Triticum durum* Desf.) is also grown on approximately 6% of the wheat-growing areas. Additionally, other cultivated wheat forms, such as spelt (*Triticum spelta* L.), einkorn (*Triticum monococcum* L.), and emmer (*Triticum dicoccum* Schrank), are grown in specific areas of the world (Bedő & Láng, 2015; Tekin et al., 2018; Tadesse et al., 2019).

Considering the wheat production data approximately the last 50 years, the harvested area of wheat has ranged from 204 to 239 million hectares; however, it can be clearly said that it has not increased much in general with the decreases in recent years. In contrast, the total wheat production has increased from 222 to 760 million metric tons (Fig. 9.1) due to the increase, from 1.1 to 3.5 tons per hectare, in yield in this process (FAOSTAT, 2022). The increase in wheat production, in general, has resulted from utilizing semi-dwarf wheat cultivars, which are more productive and

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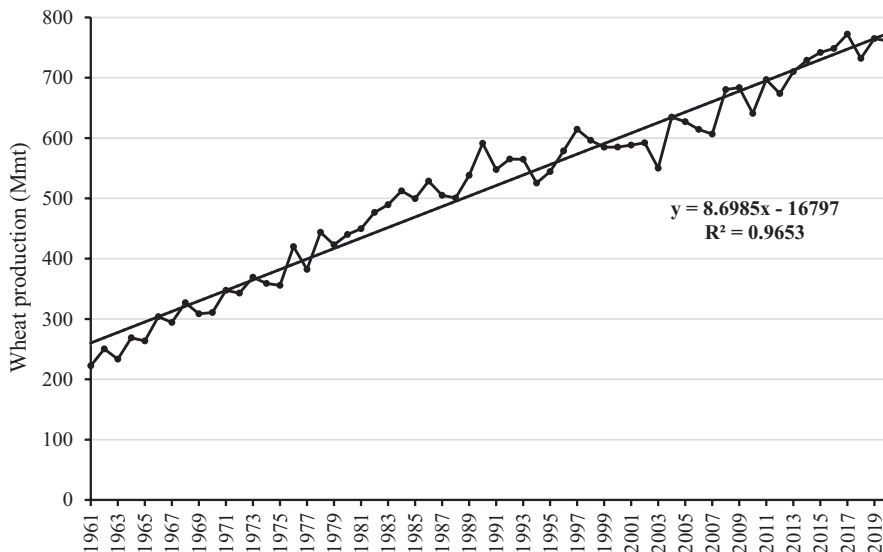


Fig. 9.1 World wheat production. (FAOSTAT, 2022)

have better response to inputs (fertilizers, water, etc.), and resistant varieties to biotic and abiotic stresses. Although wheat production has increased in this period, there is a huge gap between the expected genetic gain and the current genetic gain. An estimation of the global demand for agricultural products by the UN-FAO demonstrates that agricultural production has to be increased by 50% to meet the food supplies of the world population by 2050. To meet this demand on existing cropland, a significant increase will be required especially in grain yield for crops such as wheat. Several studies are also reported that an increase in grain yield of 2.4% is required by 2050 to meet the global wheat demand (Reynolds et al., 2012; Shiferaw et al., 2013). In contrast, the current genetic gain for wheat grain yield is approximately 1% (Crespo-Herrera et al., 2018; Tadesse et al., 2019; Mondal et al., 2020).

Traditional breeding methods have been mostly used to reach the current genetic gain in wheat. Given the many processes of crossing, selection, and testing involved in the traditional production of new wheat varieties, it can take over a one decade to create a new variety. However, both environmental and biotic factors have been changing rapidly, threatening wheat production and making it difficult to reach the expected genetic gain. The Intergovernmental Panel on Climate Change (IPCC) predicts that the average temperature may increase as much as 2 °C by 2050 and nearly 5 °C by 2100.

Breeders, therefore, have constantly been turning to breeding strategies combined with new technologies, providing shortening breeding cycles, and increasing selection efficacy, to keep up with the future wheat production demands and adapt to the changing stress factors. The current genetic gain in wheat to date has heavily relied on the exploitation of variation within the species in the primary gene pool

with narrow genetic variation. However, it can be clearly said that it will be necessary to expand the germplasm base to meet the future global demands. Wild relatives of wheat especially in primary and secondary gene pools have a great potential to expand genetic variation for many useful traits. In recent times, gene/allele introgression from wild relatives into wheat has become one of the popular breeding strategies due to the development of novel strategies to induce introgression and genomic tools to enhance selection efficacy. Existing variation in wild wheat species is able to lead the creation of elite gene pools to achieve the expected genetic gain in wheat.

9.2 Introgression of Useful Traits from Wild Relatives into Modern Wheat

The tribe Triticeae includes over 500 species from 27 different genera (e.g., *Triticum*, *Aegilops*, *Agropyron*, *Ambylopyrum*, *Elymus*, *Hordeum*, *Leymus*, *Secale*, *Thinopyrum*) (Liu et al., 2017a). Interspecies crossing studies have been carried out ever since the beginning of breeding. The first reported fertile hybrids were between bread wheat and rye (Wilson, 1876). Many crossing studies like these were carried out worldwide; however, the findings were not sufficiently used in practice until 1950s. In general, alien gene introgression from wild/cultivated species into wheat depends on the evolutionary distance between the species used in crossing. Harlan and de Wet (1971) proposed a concept of three gene pools as primary, secondary, and tertiary, based on classification of success rate of hybridization, also called as *he sine qua non* of introgression, among the species.

In this concept, crop species are classified into primary (GP-1), secondary (GP-2), and tertiary (GP-3) gene pools. Species in the primary gene pool include durum and bread wheat varieties, and their landraces, cultivated emmer, einkorn, and spelt, wild *T. dicoccoides*, and diploid donors (*Triticum boeoticum*, *T. urartu*, *Ae. tauschii*) of the A and D genomes. Genetic transfers between these two genomes can occur with direct hybridization due to homologous recombination. Although embryo rescue can be used in some cross combinations, no cytogenetic manipulation procedures are required in general. The secondary gene pool includes the polyploid *Triticum* and *Aegilops* spp. (*T. timopheevii*, *T. zhukovskiyi*, *Ae. speltoides*, etc.), and they commonly share partial or at least one genome among the three genomes of hexaploid wheat. The hybridization within this gene pool illustrates reduced chromosome pairing. Genetic transfers among them can occur with direct hybridization, breeding protocols, and partial homologous exchanges or using special manipulation strategies among the nonhomologous genomes. Embryo rescue can be used a complementary to obtain fertile hybrids. On the other hand, the diploid and polyploid species belonging to mostly Triticeae species, including the two domesticates [rye (R genome) and barley (H genome)], are in the tertiary gene pool, and they have nonhomologous genomes. Therefore, gene transfer from a species to

other one cannot be achieved by homologous recombination. As a necessity, somewhat complex protocols, assisting homoeologous exchanges, are needed to transfer related genes/alleles controlling useful traits.

Although there were some barriers to exploit introgression materials in wheat improvement in old times, such as obtaining defective genotypes with low grain yield and quality likely caused by a combination of linkage drag and an inadequate level of genetic complementation (Hao et al., 2020), many successful introgressions from wild wheat species into modern wheat have been made using new breeding strategies combined with genomic tools in recent times.

9.2.1 *Introgressions for Resistance to Abiotic Stresses*

Several useful genes/alleles providing resistance to abiotic stresses have also been successfully introgressed from wild relatives into durum/bread wheat germplasm although genes from wild relatives are often introgressed to enhance resistance to biotic stresses. Some examples of successful introgressions are summarized below.

Especially useful genes in accessions, belonging to *T. monococcum*, *T. dicoccoides*, and *T. dicoccum*, have been introgressed into durum/bread wheat, so far. Successful overwintering is a preliminary requirement to guarantee high winter wheat yields, and frost tolerance is one of the most critical factors in this overwintering. Dorofeev et al. (1987) reported that some RILs, obtained from crosses between *T. turgidum* spp. *durum*/*T. monococcum* spp. *monococcum* amphiploids and bread wheat, were found to have enhanced frost tolerance. Recently, Knox et al. (2008) also identified candidate *CBF* (*C-repeat binding factor*) genes for the locus *Fr-A^m2* controlling frost tolerance in *T. monococcum*. In addition to frost, soil salinity also causes a significant yield reduction, and it is known that sodium exclusion is one of the key mechanisms providing salt tolerance in wheat (Munns et al., 2006). Additionally, durum wheat is more sensitive to salinity than bread wheat in general. However, a durum wheat line (Line 149), derived from a cross between an accession of *T. monococcum* (C68-101) and a durum wheat cultivar (Marracos), has a tolerance to salinity stress. In this genotype, two major loci (*Nax1* and *Nax2*) (Munns et al., 2003; Davenport et al., 2005) were determined to control the Na⁺ exclusion trait and transferred into durum wheat. Munns et al. (2012) reported that *TmHKT1;5-A*, a gene in the *Nax2* locus, increased the grain yield of durum wheat by 25% in comparison to NILs without the *Nax2* locus in field trials on saline soils.

T. dicoccoides also harbors rich allelic diversity for many important traits such as resistance to biotic and abiotic stresses. Merchuk-Ovnat et al. (2016) introgressed some QTLs from *T. dicoccoides*, via marker-assisted selection, into durum and bread wheat cultivars and reported that obtained NILs enhanced grain yield and yield stability across environments with water-limited conditions. Bacher et al. (2021) also conducted a study, using an elite durum wheat background derived from crosses between a durum wheat (cv. Svevo) and *T. dicoccoides* (acc. Zavitan), under water-limited conditions and reported that a line among introgression lines had a

significant shift in root-to-shoot ratio and incorporating wild gene/alleles into elite durum wheat has a great potential to enhance the range of water stress adaptation. Additionally, Ullah et al. (2021a) highlighted that cultivated emmer (*T. dicoccon*) has a potential to enhance heat stress tolerance in hexaploid progeny. In this study, diverse *T. dicoccon* genotypes were crossed and backcrossed with hexaploid wheat genotypes, and approximately a thousand DH lines were obtained from BC₁F₁s of these crosses. These emmer-based hexaploid lines were tested with commercial checks in a multi-environment, multi-season strategy, and the results demonstrated that direct emmer introgression can positively affect wheat performance under heat stress (Ullah et al., 2021a). Apart from these species, in recent times, many studies have been conducted to transfer adaptive traits of *Aegilops tauschii*, the ancestral donor of the D genome, into hexaploid wheat, and preliminary results illustrate that *Ae. tauschii* has a great potential to introduce drought and heat adaptive traits into modern wheat genotypes (Itam et al., 2020; Molero et al., 2022).

9.2.2 Introgressions for Resistance to Biotic Stresses

9.2.2.1 Resistance to Rust Diseases

Three stem rust resistance genes, *Sr21*, *Sr35*, and *Sr22*, have been introgressed from einkorn into bread wheat (Kerber & Dyck, 1973; McIntosh et al., 1984; Olson et al., 2010). When *Sr21* was introgressed from einkorn to tetraploid and hexaploid background, a progressive dilution of resistance provided by *Sr21* was observed (McIntosh et al., 1984), and later, Jin et al. (2007) claimed that the expression of *Sr21* can be affected by polyploidy or genetic background. *Sr22* resistance gene, first identified in the *T. boeoticum* Boiss. accession G-21 (Gerechter-Amitai et al., 1971) and *T. monococcum* L. accession RL5244 (Kerber & Dyck, 1973), was introgressed from wild and cultivated A genome diploid wheats into bread wheat (The, 1973; Olson et al., 2010). The et al. (1988) reported that there were no significant differences between *Sr22* carriers and non-*Sr22* controls. Later on, it was determined that this gene provides effective resistance to the TTKS lineage (known as Ug99) (Jin et al., 2007). In addition to these three resistance genes, *SrTm4* (Briggs et al., 2015) and *Sr60* were introgressed from *T. monococcum* into hexaploid wheat. Chen et al. (2020) introgressed *Sr60* gene, providing a race-specific resistance, from *T. monococcum* into common wheat breeding line (UC12014-36) and reported that the effect of this gene for resistance to stem rust was successfully validated. Wheat wild relatives except for einkorn have been previously used for introgression of resistance genes, providing resistance to stem rust, into wheat varieties including *Sr32*, *Sr33*, *Sr39*, *Sr45*, *Sr46*, *Sr47*, *SrTA10171*, *SrTA10187*, and *SrTA1662* from *Aegilops* spp. (Kerber & Dyck, 1979, 1990; Marais et al., 1998; Faris et al., 2008; Yu et al., 2015) and *Sr36*, *Sr37*, and *Sr40* from *T. timopheevii* (Zhuk.) Zhuk. (McIntosh & Gyrfas, 1971; Dyck, 1992).

Similar to stem rust, many leaf rust resistance genes have been transferred from wild relatives into wheat varieties. A dominant resistance gene, located on chromosome 3A, was introgressed from *T. monococcum* to common wheat by Valkoun et al. (1986). Hussein et al. (1997) introgressed some leaf rust resistance genes from *T. monococcum* ssp. *monococcum* and spp. *aegilopoides* into wheat. Singh et al. (2007) also mapped a number of leaf and stripe rust resistance genes in *T. monococcum* and transferred to bread wheat. Recently, the gene *Lr63* from *T. monococcum* was transferred onto chromosome 3AS of hexaploid wheat by Kolmer et al. (2010). Other *Lr* genes from *Triticum* spp. such as *Lr1*, *Lr9*, *Lr24*, and *Lr47* from “Thachter” isogenic lines (Nocento et al., 2007); *Lr50*, *QLr.icg-5B*, and *QLr.icg-2A* from *T. timopheevii* (Brown-Guedira et al., 2003; Leonova et al., 2008); and *Lr53* and *Lr64* from *T. dicoccoides* (Marais et al., 2005a; Kolmer et al., 2010) were introgressed into wheat. Additionally, the resistance genes *Lr21*, *Lr22a*, *Lr32*, *Lr41*, and *Lr42* were introgressed from *Aegilops tauschii* into wheat (Dyck & Kerber, 1970; Rowland & Kerber, 1974; Kerber, 1987; Cox et al., 1994). Many leaf rust resistance genes, such as *Lr76* from *Ae. umbellulata* (Bansal et al., 2017); *Lr47*, *Lr51*, and *Lr66* from *Ae. speltoides* (Helguera et al., 2000, 2005; Marais et al., 2009a); *Lr54* from *Ae. kotschyi* (Marais et al., 2005b); *Lr56* from *Ae. sharonensis* (Marais et al., 2010); *Lr57* from *Ae. geniculata* (Kuraparthi et al., 2007a); *Lr58* from *Ae. triuncialis* (Kuraparthi et al., 2011), *Lr59* from *Ae. peregrina* (Marais et al., 2008); and *Lr62* from *Ae. neglecta* (Marais et al., 2009b) were identified or transferred in *Aegilops* spp. (except for *Ae. tauschii*) into wheat.

Although not as much as stem and leaf rust, stripe rust resistance genes were also characterized in wild wheat germplasm and transferred into wheat. However, several major genes identified in *T. dicoccoides* have been intensely used in breeding programs for resistance to stripe rust worldwide. Gerechter-Amitai and Stubbs (1970) reported that accession G-25 of *Triticum dicoccoides* was resistant to many races of *Puccinia striiformis* (pathogen causing stripe rust disease) from different geographical origins, and Gerechter-Amitai et al. (1989) subsequently transferred resistance gene *Yr15* from this accession into durum and bread wheat. This gene has still provided an important resistance to *Puccinia striiformis* races worldwide (Cat et al., 2021; Wang et al., 2022). Another gene, *Yr36*, identified in *T. dicoccoides* was mapped on chromosome 6B and found to be tightly linked to grain protein content locus (*Gpc-B1*) (Uauy et al., 2005), and this gene, providing non-race-specific resistance at high temperature, was successfully transferred into durum and bread wheat varieties (Hale et al., 2012). Stripe rust resistance gene *Yr5*, derived from *Triticum spelta* “album” (Macer, 1966), has carried very sound resistance to all isolates in the world (Chen et al., 2021) except for recently emerged races in China (Zhang et al., 2020) and Turkey (Tekin et al., 2021) and was introgressed into bread wheat germplasm for enhancing resistance to stripe rust (Kema, 1992; Sun et al., 2002; Keilwagen et al., 2022). Apart from these major genes, Chhuneja et al. (2008) detected adult plant resistance to stripe rust in genotypes of *T. monococcum* and *T. boeoticum*, and resistance loci were mapped onto chromosome 2A and 5A in these genotypes. Then, the resistance gene characterized from *T. boeoticum* was successfully transferred to hexaploid wheat (Chhuneja et al., 2008). Additionally, many

stripe rust resistance genes were identified or transferred from *Aegilops* spp., such as *Yr28* from *Ae. tauschii* (Singh et al., 2000), *Yr8* from *Ae. comosa* (Riley et al., 1968), *Yr17* from *Ae. ventricosa* (Bariana & McIntosh, 1993), *Yr37* from *Ae. kotschyi* (Marais et al., 2005b), *Yr38* from *Ae. sharonensis* (Marais et al., 2010), *Yr40* from *Ae. geniculata* (Kuraparthy et al., 2007a), and *Yr70* from *Ae. umbellulata* (Bansal et al., 2017).

9.2.2.2 Resistance to Powdery Mildew

There are very few examples for introgression of powdery mildew (*Pm*) resistance genes in comparison to rust resistance genes. Approximately 20 resistance genes to date have been introgressed from diploid and tetraploid wheat relatives, including *T. monococcum*, *T. carthlicum*, *T. timopheevii*, *T. dicoccoides*, *T. dicoccum*, and several *Aegilops* spp. (McIntosh et al., 2009; Schmolke et al., 2012). Only four known powdery mildew resistance genes/alleles, *Pm1b*, *Pm25*, *pm2026*, and *Pm4*, have been transferred from *T. monococcum* (Hsam et al., 1998; Shi et al., 1998; Xu et al., 2008; Schmolke et al., 2012). *T. dicoccoides* accessions have also been reported to be resistant to powdery mildew. Several *Pm* genes, such as *Pm16*, *Pm30*, *Pm26*, *Pm42*, *Pm36*, *Pm41*, and *Pm64*, were identified and transferred from *T. dicoccoides* into durum and/or bread wheat (Reader & Miller, 1991; Rong et al., 2000; Liu et al., 2002; Chen et al., 2005; Blanco et al., 2008; Hua et al., 2009; Li et al., 2009; Zhang et al., 2019). Two dominant powdery mildew resistance genes (*PmPS5A* and *PmPS5A*) were also introgressed from an accession PS5 of *T. carthlicum* into common wheat by Zhu et al. (2005a). Apart from *Triticum* spp., several genes were introgressed from *Aegilops* spp., including *Pm34*, *Pm35*, and *Pm58* from *Ae. tauschii* (Miranda et al., 2006, 2007; Wiersma et al., 2017), *Pm53* from *Ae. speltooides* (Petersen et al., 2015), and *Pm57* from *Ae. searsii* (Liu et al., 2017b).

9.2.2.3 Resistance to Fusarium Head Blight

Fusarium head blight (*Fhb*) has become one of the most serious wheat diseases especially in temperate regions of the world in addition to rust diseases. Its epidemics can result into dramatic yield and quality losses and most importantly contamination of mycotoxins such as deoxynivalenol (DON), nivalenol, and zearalenone (Prat et al., 2014). It is known that durum wheat (*T. durum*) is particularly susceptible to this disease. At the same time, mycotoxin contamination poses a great concern since semolina and pasta produced from durum wheat are used directly for human consumption. Therefore, introgression studies have been focused on enhancing resistance of durum wheat to *Fhb* in general (Prat et al., 2017).

On the other hand, resistance sources to FHB have been identified especially in hexaploid wheat. The Chinese wheat cultivar Sumai-3 and its derivatives have provided the highest levels of resistance (Prat et al., 2014). Major QTLs, *Fhb1* and *Fhb2*, from Sumai-3 derivatives were identified on chromosome 3B (Liu et al.,

2006) and 6B (Cuthbert et al., 2007). A recent study conducted by Prat et al. (2017) reported that the major gene *Fhb1* was successfully introgressed from bread wheat into durum wheat. Additionally, although several studies showed that moderate FHB resistance was found in accessions of *T. monococcum*, *T. dicoccoides*, *T. dicoccum*, and *T. carthlicum* (Buerstmayr et al., 2003, 2012; Oliver et al., 2007, 2008), to our knowledge, no successful introgression from these species into durum/bread wheat has been reported. However, wild species in tertiary gene pool of wheat have shown promise for FHB resistance and introgression of resistant genes/alleles (Qi et al., 2008; Jauhar, 2014; Haldar et al., 2021).

9.2.2.4 Resistance to Other Biotic Stresses

Several introgression studies to enhance resistance of wheat against minor diseases and insects have also been conducted using wild wheat relatives. However, the majority of developed lines has not been widely used, so far. Some important studies among them are summarized below.

Vasu et al. (2000) identified resistance genes to karnal bunt (caused by *Tilletia indica*) in *T. monococcum* and transferred into bread wheat. Additionally, synthetic hexaploid wheats, developed using direct crosses between *Ae. tauschii* and durum wheat genotypes, have an important genetic diversity to biotic stresses. These include resistance to karnal bunt (Mujeeb-Kazi et al., 2008), Stagonospora blotch (caused by *Stagonospora nodorum*) (Loughman et al., 2001), Septoria leaf blotch (caused by *Septoria tritici*) (Arraiano et al., 2001), tan spot (Xu et al., 2004; Tadesse et al., 2007), soil-borne cereal mosaic virus (Hall et al., 2009), cyst nematode (*Heterodera avenae*) (Eastwood et al., 1991), and root-knot nematode (*Meloidogyne* spp.) (Kaloshian et al., 1990).

On the other hand, diploid and tetraploid wheat species have a rich genetic variation for resistance to insect pests such as hessian fly (*Mayetiola destructor*) and greenbug (*Schizaphis graminum*). Many hessian fly and greenbug resistance genes were introgressed from *Aegilops* spp., *T. carthlicum*, *T. araraticum*, *T. dicoccum*, and durum wheat into bread wheat germplasm (Nsarellah et al., 2003; Martin-Sanchez et al., 2003; Liu et al., 2005; Sardesai et al., 2005; Weng et al., 2005; Zhu et al., 2005b; Friesen et al., 2008; Singh et al., 2020).

9.2.3 Introgression of Other Useful Traits

9.2.3.1 Bread-Making Quality

The bread-making quality is a vital breeding aim for bread wheat. In general, some variations in bread quality are due to high molecular weight glutenins (HMWGs) (Boehm Jr et al., 2017; Hernandez-Espinosa et al., 2019; Morris, 2021). It is known that the *Glu-D1* genes, especially the *Dx5* + *Dy10*, contribute superior dough

strength and bread quality compared to the *Glu-A1* and *Glu-B1* genes. In contrast, *Dx2 + Dy12* in *Glu-D1* is more suitable for pastry end uses. Therefore, *Dx5 + Dy10* has been an important breeding target for wheat breeders. On the other hand, utilization of durum wheat is somewhat hampered by especially low gluten strength caused by the lack of *Glu-D1* loci. During the last two decades, there has been growing interest in the utilization of D genome synthetic hexaploid wheats (SHWs) developed from crosses between *Ae. tauschii* and durum wheat. Recently, Delorean et al. (2021) sequenced 273 *Ae. tauschii* accessions and discovered 45 haplotypes in *Glu-D1* and reported that *5+10* allele originated in lineage 3 showing a unique origin of this important allele. Bibi et al. (2012) characterized an SHW subset (*T. turgidum* × *Ae. tauschii*) and claimed that favorable allelic variants (*Dx5 + Dy10* and *Dx1.5 + Dy10*) in *Glu-D1* were frequently observed in SHWs. Rasheed et al. (2012) also indicated that these superior alleles in SHWs can be used as the priority selective sieve to improve wheat quality.

9.2.3.2 Grain Protein Content

Avivi (1978) reported that *T. dicoccoides* has great potential to improve grain protein content of modern wheat. Later, Joppa and Cantrell (1990) crossed *T. dicoccoides* with durum wheat and claimed that obtained substitution lines had higher grain protein content than checks. Joppa et al. (1997) discovered a QTL, controlling high Gpc trait, which explained 66% of total variation in these substitution lines. Distelfeld et al. (2004) named this QTL as *Gpc-B1*, and then it was positionally cloned and renamed as *NAM-B1* by Uauy et al. (2006). In addition to *T. dicoccoides*, two spelt wheat (*T. aestivum* var. *spelta*) accessions have wild-type allele of this loci (Asplund et al., 2010). Many studies have shown that wild-type allele of this locus can be easily introgressed into wheat by marker-assisted selection. Considering conducting studies, lines carrying this wild-type allele of *Gpc-B1* averaged an increase of 21.8 g/kg in grain protein content compared to other lines without this allele (Kumar et al., 2011; Vishwakarma et al., 2014, 2016). Tabbita et al. (2017) reported that this functional allele also positively impacts on grain Fe and Zn in addition to protein content.

9.3 Quantitative Trait Loci (QTL) Mapping in Wild Relatives of Wheat

Most of the characteristics of economic significance to breeders and consumers are quantitative traits that are controlled by polygenes or QTLs. The genome regions including the genes encoding the important feature have been determined more specifically utilizing markers and QTL analysis. Thus, a new door has opened for more complex applications. To get better information the genetic basis of complex traits,

QTL mapping has been broadly performed in various crop species (Zhu et al., 2008). Wild ancestors of crops harbor great genetic diversity, with highly beneficial potential to improve new cultivars having different traits such as resistance to biotic and abiotic stresses for crop breeding, compared to modern cultivars (Tanksley and McCouch 1997). Hence, for breeders, reintroducing beneficial wild alleles “left behind” throughout plant domestication and modern breeding is very crucial to enhance the existing gene pool (Merchuk-Ovnat et al., 2016). Numerous QTL works responsible for different traits have been conducted on various traits in crops, containing wheat which is one of the world’s major food sources. Up till now, many desirable genes for resistance to different stresses have been transferred from relatives of wheat which have as a potential source for favorable alleles, such as *Aegilops tauschii* and durum wheat into bread wheat (Knot, 1989; Lutz et al., 1995), and discovered new QTLs using various populations. In addition, significant associations were determined between RFLP (restriction fragment length polymorphism) markers by Dubcovsky et al. (1996), constructed a map (length: 1.079 cM) in *T. monococcum* composed of 335 markers. Through a population from “*T. dicocoides* accession Hermon H52 × durum variety Langdon,” Peng et al. (2000) constructed another map in which the total map length exceeded 3.000 cM. AFLP markers revealed that genetic variations between parental genomes were controlled by the B genome. Segregation-distorted markers were mainly found on chromosomes (Chr) 4A, 5A, and 5B. In a BC₂F₂ population produced from “Prinz variety × W-7984 line,” Huang et al. (2003) studied QTLs associated with yield and yield components. A synthetic hexaploid wheat, W-7984, was from “Altar 84 (*Triticum turgidum* L. cultivar)” and “WPI 219 (*Triticum tauschii* accession),” as defined by Nelson et al. (1995). Out of 298, the polymorphism rate of 210 microsatellite markers was found as 70.5% for yield and yield components. They concluded that QTLs for these traits were mostly separated on Chr 7B, 6A 5B, 4D, 3B, and 2D. Another study, Lohwasser et al. (2005), used 75 RILs from ITMI (International Triticeae Mapping Initiative) for mapping. They indicated a major QTL (LOD>3.0) for dormancy on Chr 6DL. Singh et al. (2007) constructed a genetic linkage map using a set of 93 RIL lines from *T. boeoticum* × *T. monococcum*. They determined that the amounts of polymorphism between parents were 50% (RFLP markers) and 73% (SSR markers) and the 188 polymorphic loci were totally mapped. In earlier works, Landjeva et al. (2008) reported a number of QTLs linked to coleoptiles, shoots, and roots under drought stress using 114 RILs from “Opata 85 (*Triticum aestivum* L.) × W7984 (the synthetic hexaploid wheat).” It was proved that the 35 QTLs linked to shoot length, coleoptile, root, and root/shoot ratio were found on Chr 7D, 6D, 6B, 5B, 3D, 2D 2B, 2A, 1B, and 1A. With another approach, Liu et al. (2014) mapped 148 putative QTLs related to 9 yield traits using RILs from “NongDa3331 (common wheat line) × Zang 1817 (Tibetan semi-wild wheat accession)” and QTLs separated on 19 Chr (except for 2D and 1A). They indicated that Zang 1817 had some stable QTLs determined in more than four environments, such as *QSI-7A1* (spike length); *QPh-4B1*, *QPh-3A1*, and *QPh-4D* (plant height); *QGws-4D* (grain weight per spike); *QEp-4B2* (ears per plant); and *QTgw-4D* (thousand grain weight). Another map reported by Milner et al. (2016) used the *Triticum turgidum* ssp. *durum*

multiparental RIL population from crossing four elite cultivars (Colosseo, Neodur, Rascon/2*Tarro, and Claudio) from various origins. They determined QTLs associated with maturity and heading date, plant height, and grain yield. Among the QTLs, a major QTL (*QGy.ubo-2B*) linked to grain yield per se across environments was determined on Chr 2B. Findings demonstrated that this multiparental population supplies important possibilities for the genetic dissection of agronomic significant characters of breeding in durum wheat. Merchuk-Ovnat et al. (2016) explored the potential of selected QTLs from wild emmer wheat, introgressed by marker-assisted selection. Three of the introgressed QTLs (*7AS QTL*, *2BS QTL*, and *1BL QTL*) were successfully confirmed in NILs (near-isogenic lines) BC₃F₃ and BC₃F₄. These QTLs can be supported to enhance drought resistance in *T. turgidum* ssp. *durum* and *T. aestivum* cultivars. Through wheat–*Agropyron cristatum* 7P deletion lines and translocation lines, the high thousand grain weight locus from *Agropyron cristatum* Chr 7P onto 7PS1-3 was mapped by Lu et al. (2017). During the same period, Ma et al. (2017) released another map. They used a total of 186 RILs (Tibetan semi-wild wheat Q1028 × Zhengmai 9023) and found 15 QTLs related to root traits (total root length, maximum root length, root average diameter, root volume, root surface area, root/shoot ratio, and number of root tips) on 8 chromosomes. Q1028 had positive alleles of six QTLs. QTLs controlling flag leaf width, flag leaf length, flag leaf angle, and flag leaf area using RIL population (ND3331 × Zang1817-Tibetan semi-wild wheat) were identified works of Liu et al. (2018). The 23 QTLs related to these traits were found on different chromosomes (7D, 7B, 6B, 5A, 4B, 3D, 3A, 2B, and 1B). Additionally, multiple linked QTLs were determined to be responsible for pleiotropic effects on Chr 5A, 4B, and 1B. Zhang et al. (2019) mapped the enhanced GNS locus from *Agropyron cristatum* 6P onto the 6PL (0.27–0.51) by using five wheat–*Agropyron cristatum* 6P translocation lines, five deletion lines, and genetic populations of these lines, resulting in an improvement of nearly ten grains per spike. Worth noting, a recent map was assembled from 208 RILs from Svevo × WEW accession Y12-3 by Fatiukha et al. (2020). This map identified several QTLs responsible for thousand kernel weight (TKW) and grain protein content (GPC). Totally, 12 and 11 QTLs were determined in the population for GPC (LOD= 3.6–27.8) and TKW (LOD= 2.2–15.7), respectively. Significant GPC QTLs (*QGpc.uhw-7B.2*, *QGpc.uhw-6B*, *QGpc.uhw-6A*, and *QGpc.uhw-5A.1*) with favorable alleles from WEW were detected on Chr 7BL, 6BS, 5AS, and 4BS. In another very recent report, QTLs responsible for grain yield-related features were determined using four RILs from “a Chinese domesticated cultivar; Yanzhan 1 × 4 donor parents containing Hussar (a British domesticated cultivar) and three semi-wild wheats in China” (Hu et al., 2020). They determined 161 QTL associated with different traits: spike-related traits (49 QTLs), grain yield per plant and yield components (10 QTLs), plant height (22 QTLs), flag leaf-related traits (43 QTLs), heading date, and flowering date (37 QTLs). Result of the QTL validation, for yield-related traits, *QSPS-2A.4* and *QSL-4A.1*, revealed important marker–trait associations. These QTL regions will be valuable for yield improvement in wheat.

In addition to these examples, various QTL studies using wild progenitors/relatives of wheat were also performed to explore novel variants/alleles responsible for

quality traits of wheat such as grain protein content (GPC), grain zinc content (GZn), grain iron content (GFe), and yellow pigment content (Yp) (Blanco et al., 1996, 2002; Joppa et al., 1997; Börner et al., 2002; Gonzalez-Hernandez et al., 2004; Nelson et al., 2006; Peleg et al., 2009). The QTLs for GPC, for instance, Avivi (1978) detected a QTL for Gpc-B1 in a wild accession (FA-15-3) of tetraploid wheat (*Triticum turgidum* var. *dicoccoides*). Afterward, Joppa and Cantrell (1990) assessed the same accession to produce a complete set of chromosome substitution line. Olmos et al. (2003) determined Gpc-B1 gene on Chr arm 6BS using substitution lines. Gonzalez-Hernandez et al. (2004) identified three QTLs linked to GPC on Chr 5B of *T. turgidum* var. *dicoccoides*. In another study, through RILs from “a synthetic hexaploid (W7985) × bread wheat (Opata 85),” Nelson et al. (2006) determined QTLs in the wheat genome affecting baking quality and milling and claimed that GPC associated with the Gli-D2 gliadin locus on Chr arm 6DS. In an earlier report, Peleg et al. (2009) studied the phenotypic effects and chromosomal location of QTLs linked to wheat grain mineral nutrient concentration using 152 RILs from “Langdon (durum wheat) × G18-16 (wild emmer).” A total of 82 QTLs related to 10 minerals were mapped. For grain protein content and other minerals, some genomic regions on Chr 7A, 6B, 5A, and 2A were determined. They concluded that these regions offer unique advantages for breeding studies to improve various mineral nutrients in wheat grain, synchronously. These QTL studies about GPC confirmed the potential role of wild emmer germplasm for breeding programs.

In addition to GPC, wild relatives of wheat also had novel grain Fe and Zn contents. For this aim, many studies using wild relatives of wheat were performed to identify QTLs for both Fe and Zn (Peleg et al., 2009; Tiwari et al., 2009; Pu et al., 2014; Srinivasa et al., 2014; Crespo-Herrera et al., 2017; Krishnappa et al., 2017; Velu et al., 2017). For instance, using RILs from “*T. boeoticum* accession pau5088x *T. monococcum* accession pau14087,” Tiwari et al. (2009) determined QTLs related to grain Fe (two QTLs) on Chr 2A and 7A and grain Zn (1 QTL) on Chr 7A. Another map was constructed by Srinivasa et al. (2014) using 185 RILs. They detected QTLs for Zn content (QZn.bhu-2A, QZn.bhu-2B, QZn.bhu-3D, QZn.bhu-6A, and Zn.bhu-6B) and Fe content (QFe.bhu-1A.1, QFe.bhu-1A.2, QFe.bhu-1A.3, QFe.bhu-2A, and QFe.bhu-3B) of the grain. Another RIL population produced from “*T. spelta* L. × synthetic hexaploid wheat” used by Crespo-Herrera et al. (2017) explored QTLs for Zn and Fe: *nQGZn.cimmyt-7B_IP2* and *QGFe.cimmyt-4A_P2* on Chr 7B and 4A, respectively. They reported that for wheat biofortification, the regions determined on Chr 1B, 3B, 6A, and 7B are of special attraction. Velu et al. (2017) used 105 tetraploid RIL populations (Saricanak98 × *T. dicoccon*; MM5/4) and 127 hexaploid RIL populations (Adana99 × *Triticum sphaerococum* line 70,711) for QTL mapping related to high GZn and GFe contents. In tetraploid population, they detected QTLs for GZn and GFe on Chr 6B, 5B, 1B, and 3A while eight major QTLs for GZn and four for GFe found on Chr 7B, 7A, 6B, 3D, 3A, 2B, 1D, and 1B in hexaploid population. For GZn and GFe, colocalization of QTLs offers the advantage to use only one MAS program to upgrade the contents of both GFe and GZn, simultaneously. Identified genomic regions related to grain Zn and Fe

contents explained above will support crop improvement to develop micronutrient density in wheat grain.

Besides these information, Se concentration of the grain is also an essential mineral for human and animal nutrition (Kieliszek & Błazejak, 2016). Up to now, some publications have reported QTLs linked to accumulation of Se in wheat grain using wild relatives (Pu et al., 2014, 2018; Yan et al., 2018), for instance, a total of 39 QTLs determined by Pu et al. (2014) using two RIL populations: (i) SHW-L1 × Chuanmai 32 and (ii) Chuanmai 42 × Chuannong 16. A synthetic hexaploid wheat (SHW-L1) used in this research was produced from “*Triticum turgidum* ssp. *turgidum* AS2255 (AABB) and *Aegilops tauschii* ssp. *tauschii* AS60” (Zhang et al., 2004). For Se concentration in wheat grain, in the first population, authors found four QTLs on Chr 3D, 4A, 5B, and 7D while one QTL on Chr 4D found in the second populations. In another research, 15 QTLs for GSeC (grain Se concentration) and GSeY (grain Se yield) on Chr 7B, 7A, 6A, 5A, 4B, 3A, 2B, 1B, and 1A explaining phenotypic variation between 1.4 and 18.6% were determined by Yan et al. (2018) using RILs from *T. dicoccoides* (accession G18-16) and Langdon (durum wheat). Pu et al. (2018) also used a RIL population generated from “SHW-L1 × Chuanmai 32.” For Se concentration, they detected QTL on 1B, 3D, and 7D in the grains, roots, and leaves. They emphasized that synthetics are potentially beneficial germplasm sources for improving Se contents of the grains, as well as for increasing K, Mn, P, and Fe concentrations. These above QTL findings can be evaluated in the MAS programs for Se biofortification of wheat grain.

In the last two decades, during the selection of new wheat cultivars, wheat breeders have not only focused on protein, zinc, iron, and selenium content of grain but also on high yellow pigment content of grain which is predominantly linked to carotenoid compounds. Up to now, numerous QTL mapping studies have been performed to map QTLs responsible for grain yellow pigment content in wheat (Elouafi et al., 2001; Pozniak et al., 2007; Zhang et al., 2008; Patil et al., 2008; Blanco et al., 2011; Crawford et al., 2011). For example, Elouafi et al. (2001) determined three QTLs on Chr 7 explaining 62% of the total phenotypic variation using a RIL population (*T. turgidum* L. var. durum cultivar × *T. dicoccoides* (acc.600545)).

In another study, Pozniak et al. (2007) explored four QTLs underlying phenotypic variation in endosperm color on Chr 7B, 6B, 4B, and 2A. This study became the first report of mapping *Psy* (phytoene synthase) genes in durum wheat and indicated the role of this gene for endosperm color. *Psy-A1* gene on Chr 7A was determined by Zhang et al. (2008). They also found a QTL on 1RS demonstrating 31.9% of the phenotypic variance. In addition to these examples, Patil et al. (2008) also determined five QTLs linked to yellow pigment content on Chr 7B, 5B, 3B, and 1A using RILs from “variety (PDW 233) × landrace (Bhalegaon 4).” Another RIL population from “line UC1113 × the variety Kofa” was used by Roncallo et al. (2012) to determine main QTLs responsible for yellow pigment content and flour yellow color on Chr arms 7BL, 7BS, 7AL, 7AS, 4AL, and 6AL. They also determined that a novel minor QTL found 7AS affected flour yellow color with an epistatic effect on yellow pigment content. To take a general look at the various QTL information described above about wild relatives of wheat, the closely related markers/marker

Table 9.1 List of QTLs determined for grain protein content (GPC), grain zinc (GZn) content, grain iron (GFe) content, grain selenium (GSe) content, and yellow pigment content

Cross	Population type and size	Total QTLs	Chr	Marker	References
<i>Grain protein content</i>					
Durum wheat (Messapia) × <i>T. turgidum</i> L. var. <i>dicoccoides</i> (MG4343)	RILs (65)	6	4BS, 5AL, 6AS, 6BS, 7BS	–	Blanco et al. (1996)
<i>T. turgidum</i> (L.) var. <i>dicoccoides</i>	RICLs (85)	1	6BS	Xabg387-6B-Xmwig79-6B (66)	Joppa et al. (1997)
Durum wheat (Messapia) × <i>T. turgidum</i> var. <i>dicoccoides</i> (MG4343)	RILs (65)	7	4BS, 5AL, 6A, 6BS, 7AS, 7BS	Xpsr627 (10.2), Xutv913 (12.6), Pan2 (14.8), Xcdo412 (14.9), Xpsr167 (18.4), Gai-1 (31.7)	Blanco et al. (2002)
<i>T. aestivum</i> (Opata 85) × synthetic hexaploid wheat (W7984)	RILs (114)	2	2DS, 7AS	–	Börner et al. (2002)
<i>T. turgidum</i> (L.) var. <i>dicoccoides</i> [LDN(Dic-5B)] × LDN	RICLs (133)	3	5B	Xbcd1030–Xgwm604 (32), Xcdo584–Xabc310 (33)	Gonzalez-Hernandez et al. (2004)
<i>T. aestivum</i> (Opata85) × synthetic hexaploid wheat (W7984)	114 RILs	4	2AL, 2DS, 5AL, 6DS	Xbcd152-Xfbb329 (15), Xfba85-Xgwm469 (16), Xcdo1312-Xabg391 (19), Xbcd102- Xbcd18 (32)	Nelson et al. (2006)
Durum wheat (Langdon) × wild emmer accession (G18–16)	RILs (152)	10	2AL, 2BL, 3BL, 4AL, 5AS, 5BL, 6AS, 6BL, 7AL, 7BS	–	Peleg et al. (2009)
<i>Grain zinc (GZn) and grain iron (GFe) contents</i>					
<i>T. boeoticum</i> (Tb5088) × <i>T. monococcum</i> (Tm14087)	RILs (93)	GZn-2; GFe-3	7A; 2A, 7A	Xcfd31-Xcfa2049 (18.8) [GZn]; Xwmc382-Xbarc124 (12.6); Xgwm473-Xbarc29 (11.7) [GFe]	Tiwari et al. (2009)

(continued)

Table 9.1 (continued)

Cross	Population type and size	Total QTLs	Chr	Marker	References
Durum wheat (cv. Langdon) and wild emmer (accession G18-16)	RILs (152)	GZn-6; GFe-11	2A, 5A, 6B, 7A, 7B; 2A, 2B, 3A, 3B, 4B, 5A, 6A, 6B, 7A, 7B	wPt8216 (15.4), wPt 9555 (15.7), gwm445 (15.4) [GZn]; gwm154 (14.6), wpt-9555 (10.7), gwm473 (11.7); gwm1054 (11.7) [GFe]	Peleg et al. (2009)
Synthetic hexaploid (SHW-L1) × <i>T. aestivum</i> (Chuanmai 32)	RILs (171)	GZn-4; GFe-4	2D, 3D, 4D, 5D; 2B, 5B, 5D, 7D	-	Pu et al. (2014)
<i>T. spelta</i> (PI348449) × <i>T. aestivum</i> (HUW 234)	RILs (185)	GZn-5; GFe-5	2A, 2B, 3D, 6A, 6B; 1A, 2A, 3B	989092IF10-1101425IF10 (16.46) [GZn]; 1708014IF10-1000008IF10 (16.55), 3022954IF10-1102324IF10 (25.95) [GFe]	Srinivasa et al. (2014)
<i>T. aestivum</i> (SeriM82) × <i>T. dicoccoides/Ae. Tauschii</i> (SHW CW176364)	RILs (140)	GZn-3; GFe-5	4BS, 6AL, 6BL; 2BL, 2DS, 4BS, 6AL, 7DS	TP91631-TP81797 (17.3) [GZn]; TP91631-TP81797 (10.7), TP43715-TP37547 (14.5) [GFe]	Crespo-Herrera et al. (2014)
<i>T. aestivum</i> (Adana99) × <i>T. sphaerococum</i> (70711)	RILs (127)	GZn-10; GFe-7	1B, 1D, 2B, 3A, 3D, 6A, 6B, 7A, 7B; 1B, 2A, 2B, 3A, 6B, 7B,	rPt-6561 (12), wPt-2698–wPt-0398 (14), wPt-2083–wPt-6083 (15), wPt-733,112 (25), wPt-7161–wPt-9812 (31) [GZn]; wPt-667,798–wPt-7065 (14), wPt-1394–wPt-7864 (17), wPt-5922 (18) [GFe]	Velu et al. (2017)
<i>T. spelta</i> (Bubo) × resynthesized hexaploid wheat (Turtur)	RILs (188)	GZn-4; GFe-3	1B, 6A, 7B; 3A, 4B, 5B,	3934172-3934936 (15.10), 3945822-1132640F0-5CG (16.75) [GZn]; 1234521;3034169F0-11AG (10.35) [GFe]	Crespo-Herrera et al. (2017)

(continued)

Table 9.1 (continued)

Cross	Population type and size	Total QTLs	Chr	Marker	References
Synthetic hexaploid wheat (Louries) × <i>T. spelta</i> (Bateleur)	RILs (188)	GZn-12; GFe-7	1A, 1B, 3B, 3D, 4A, 5B, 6A, 7B, 7D; 2A, 2B, 3B, 4A, 4D, 5B	4543935-3937719 (10.78), 4991478-3937490 (11.25), 4394657-3947677 (10.93), 1079651;1262636 (32.79) [GZn]; 4407677-1129284 (11.62), 4262668-1226245 (14.23), 2363822-3961236 (14.62), 3385350-1211533 (21.14) [GFe]	Crespo-Herrera et al. (2017)
<i>T. aestivum</i> (WH542) × synthetic derivative (<i>Triticum dicoccon</i> PI94624/ <i>Aegilops squarrosa</i> [409]//BCN)	RILs (286)	GZn-5; GFe-4	2A, 4A, 5A, 7A, 7B; 2A, 5A, 7A, 7B	–	Krishnappa et al. (2017)
<i>Grain selenium (Se) content</i>					
Synthetic wheat (SHW-L1) × <i>T. aestivum</i> (Chuanmai 32)	RILs (171)	24	1B, 3D, 5A, 6A, 6B, 6D, 7D,	wPt-3566~wPt-8168 (11.58), wPt-667,315~wPt-741,543 (14.34), wPt-7273~wPt-9792 (15.64), wPt-733,447~wPt-667,315 (21.15), wPt-741,599~wPt-742,156 (25.57), wPt-733,447~wPt-667,315 (25.58), wPt-733,447~wPt-667,315 (28.38)	Pu et al. (2018)
<i>Triticum dicoccoides</i> (Langdon) × wild emmer wheat (acc. G18-16)	RILs (152)	15	1A, 1B, 2B, 3A, 4B, 5A, 6A, 7A, 7B	–	Yan et al. (2018)
<i>Yellow pigment content</i>					
<i>T. turgidum</i> L. var. durum (Omrabi5) × <i>T. dicoccoides</i> (acc.600545)	RILs (114)	3	7AL, 7BL	Xgwm63e (13), Xgwm34 (53)	Elouafi et al. (2001)
<i>T. turgidum</i> L. var. durum (W9262-260D3 × <i>T. turgidum</i> L. var. durum (Kofa)	DH lines (155)	4	2A, 4B, 6B, 7B	Xgwm495 (14-20), Xgwm425 (15-21), Xgwm193 (15-21), Psy1-1 (20-23)	Pozniak et al. (2007)

(continued)

Table 9.1 (continued)

Cross	Population type and size	Total QTLs	Chr	Marker	References
<i>T. turgidum</i> L. var. durum (PDW 233) × <i>T. turgidum</i> L. var. durum (Bhalegaon 4)	RILs (140)	5	1A, 3B, 5B, 7A, 7B	Xscar3362 (22.61-55.22)	Patil et al. (2008)
<i>T. turgidum</i> L. var. durum (UC1113) × <i>T. turgidum</i> L. var. durum (Kofa)	RILs (93)	1	7A	Xcfa2293-7A - Xwmc116-7A	Zhang et al. (2008)
<i>T. turgidum</i> L. var. durum (Latino) × <i>T. turgidum</i> L. var. durum (Primadur)	F2:F3 families (121)	5	2A, 3B, 5A, 7A	Xgwm372-wPt_9797 (11.1-24.5), Xbarc84-Xgwm299 (11.5-16.2), Xgwm282-wPt_4345 (19.8-30.4), D_304196-PsyA1 (42-53.2)	Blanco et al. (2011)
<i>T. turgidum</i> L. var. durum (UC1113) × <i>T. turgidum</i> L. var. durum (Kofa)	RILs (93)	15	1BL, 2AS, 4AL, 5AS, 5AL, 5BL, 6AL, 7AS, 7AL, 7BL	BE443797_436–barc302 (10.8), Lpx-A3–wmc617 (12), wmc219–psr573.2 (12), cfa2040–barc1073 (15), wmc311–wmc276 (16.9), barc146–gwm132 (16.8-42.7)	Crawford et al. (2011)

intervals for the important QTL for yellow pigment and protein, zinc, iron, and selenium contents of the grain were presented in Table 9.1.

To sum up, unveiling the genetic source of yield-related traits in wheat relatives which are higher genetic diversity, many alleles, and potency of special variants of alleles compared to modern wheat varieties is crucial to provide global food security. Information about QTLs explained above improved our understanding on the genomic background of wild relatives. It was revealed that future wheat growing would advantage from the incorporation of alleles from wild relatives of wheat. QTLs linked to novel genes controlling crucial adaptive and yield-linked traits are now presented and could be performed through molecular breeding to develop new wheat cultivars.

Resistance to biotic stresses such as leaf rust, stripe rust, Fusarium head blight, powdery mildew, viruses, and insects which cause globally significant yield losses in plants (Imren et al., 2017; Mutlu et al., 2017; Yeken et al., 2018, 2019; Cat et al., 2019; Mutlu et al., 2020; Kavak & Celik, 2021; Celik & Ertunc, 2021; Celik & Morca, 2021; Nadeem et al., 2021; Palacioğlu et al., 2021; Saleh et al., 2021; Tekin et al., 2021; Celik et al., 2022; Celik, 2022; Morca et al., 2022) and abiotic stresses will be a future breeding aim to mitigate the world's rapidly improving food demand. Throughout history, the host and the pathogen in the wild co-evolved together due to having co-lived in mutual habitats. Therefore, it was reported that among the landraces and wild

relatives of wheat, the sources of resistance can exist often at these centers of origin (McIntosh et al., 1995; Nadeem et al., 2021).

In literature, using molecular markers, many investigations were carried out to explore the resistance properties of wild relatives of wheat for different biotic stresses. For instance, Singh et al. (2004) determined leaf rust (*Lr*) resistance genes *Lr39* and *Lr41* closely linked to Xgwm210 and Xgdm35 markers, both produced from *T. tauschii*. Marais et al. (2005a) transferred a *Lr* (caused by *Puccinia triticina*) resistance gene from *T. dicoccoides* to bread wheat. Another map was constructed by Salina et al. (2006) for *Lr* using “*T. timopheevii* var. *timopheevii* × *T. timopheevii* var. *typical*” and “*T. timopheevii* K-38555 × *T. militinae*”, and they determined 121 and 103 loci, respectively. Kuraparthi et al. (2007b) determined and mapped the cryptic wheat–*A. triuncialis* rust-resistant translocation using molecular and cytogenetic mapping in a segregating population. They designated the wheat–*A. triuncialis* translocation as T2BS·2BL–2tL (0.95). A doubled haploid population produced from synthetic hexaploid wheat line TA4152-60 and the North Dakota breeding line ND495 used by Chu et al. (2009) determined four QTLs on Chr arms 6BL (*QLr.fcu-6BL*), 5BL (*QLr.fcu-5BL*), 3AL (*QLr.fcu-3AL*), and 3BL (*QLr.fcu-3BL*) responsible for *Lr* resistance, marker intervals Xbarc5–Xgwm469.2, Xgdm116–Xbarc59, Xcfa2183–Xgwm666, and Xbarc164–Xfcp544, respectively. Another work, using introgression lines from Saratovskaya 29 (spring wheat cultivar) × *T. timopheevii*/*T. tauschii* (a synthetic allopolyploid wheat), Leonova et al. (2007) determined QTLs responsible for *Lr* in line BC5 on Chr 2B (Xgwm257–Xgwm120) and 2D (Xgwm1099–Xgwm484) and in line BC9 on Chr 2B (Xgwm257–Xgwm117) and 6B (Xgwm518–Xgwm889). McCallum et al. (2012) reported a total of 67 *Lr* resistant genes in wheat. Out of 67 genes, 35 genes were derived from alien sources, and five of them were introgressed from *T. dicoccoides* or *T. turgidum*. Ullah et al. (2016) constructed introgression lines (*Triticum aestivum*–*Triticum turgidum*) and determined loci related to *Lr* resistance on Chr 1A (Xgwm 219–Xgwm 24), 5A (Xgwm327–Xgwm24), and 2B (Xgwm451–Xgwm115) using microsatellite markers. They also emphasized that *Triticum aestivum*–*Triticum turgidum* introgression lines can be evaluated as a source of resistance genes and to the mapping of genes for pathogen resistance in breeding programs. Very recently, for the first time, Pourkhorshid et al. (2022) developed SSR markers (Xgwm88 and Xcfd13) which gave clearly scorable bands and categorized resistant/susceptible and heterozygotes for the *Lr* resistance gene *Lr36*.

Another damaging disease of wheat is stripe rust caused by the fungal pathogen *Puccinia striiformis* West. f.sp. *tritici* (Pst) (Chhuneja et al., 2008). Different researchers cataloged and designated the 40 stripe rust (*Yr*) resistance genes; out of 40, 11 genes were transferred from wild relatives of wheat (McIntosh et al., 2005; Uauy et al., 2005; Marais et al., 2005a; Kuraparthi et al., 2007b; Chhuneja et al., 2008). The *Yr* resistance gene *Yr36* was mapped on Chr 6B (Uauy et al., 2005). A set of 121 RILs from *T. monococcum* (acc. pau14087) and *T. boeoticum* (acc. pau5088) for *Yr* used by Chhuneja et al. (2008) explored QTLs on Chr 2A (Xwmc407–Xwmc170, *QYrtm.pau-2A*) in *T. monococcum* and on Chr 5A (Xbarc151 and Xcfd12, *QYrtb.pau-5A*) in *T. boeoticum*. Rosewarne et al. (2013) reported that

140 QTLs for *Yr* resistance has been identified to be spread over 49 Chr regions on wheat maps.

In addition to rust diseases in wheat explained above, Fusarium head blight (FHB) is also a serious disease of wheat cultivated in many regions of the world constraining the production. There are many reports about genes/QTL for FHB. For instance, Stack et al. (2003) found resistance QTL on Chr 7A, 5B, 3A, and 1A. A doubled haploid population of 85 lines was generated from F1 hybrids of “Strongfield” and “Blackbird” (*T. carthlicum* Nevski (*T. persicum* Vav.) (synonym *T. turgidum* L. (Nevski) group *carthlicum* Bowden) line REB68421) used by Somers et al. (2006) and explored QTLs for FHB one on 2BL from Strongfield and on Chr 6BS from Blackbird. Chen et al. (2007) determined the main FHB resistance QTL in tetraploid wheat maps to Chr 3AS. Kumar et al. (2007) found QTLs for FHB resistance on Chr 7A in tetraploid wheat. In another study, in *T. macha*, resistance QTLs were mapped to Chr 5B, 5A, 2B, and 2A (Buerstmayr et al., 2011). Based on mapping population from *T. dicoccum* donor line with three Austrian recipient cultivars, Buerstmayr et al. (2012) reported resistant QTL on Chr 7B, 6B, 6A, 4B, and 3B. *T. dicoccum* contributed all QTLs (except for 3B). Another RIL population from AC Brio (a Canadian bread wheat cultivar moderately susceptible for FHB) × TC67 (resistant for FHB cultivar produced from *Triticum timopheevii*) used by Malhipour et al. (2017) found a novel QTL for FHB resistance on Chr 5AL in the marker interval of *cfid39-cfa2185* using microsatellite molecular markers. Zhao et al. (2018) used 205 RILs from Joppa (a durum wheat cultivar) × 10Ae564 (a durum wheat introgression line for FHB resistance produced from the hexaploid wheat line PI 277012) and determined two QTLs (*Qfhb.ndwp-5A* and *Qfhb.ndwp-7A*) from 10Ae564QTL and one QTL (*Qfhb.ndwp-2A*) from Joppa for FHB resistance. Wild relatives of wheat are not only included QTLs for FHB but also are contained exotic genes for powdery mildew (*Pm*) (caused by *Blumeria graminis* f.sp. *tritici*) resistance. Using the tetraploid and diploid relatives of wheat, containing *T. carthlicum*, *T. timopheevii*, *T. monococcum*, *Aegilops* spp., and *T. turgidum* var. *dicoccoides* and var. *dicoccum*, some resistant genes were transferred (McIntosh et al., 2008, 2009; Schmolke et al., 2012).

In addition, using wild emmer, various researchers identified *Pm* resistance genes: *Ml3D232 PmG16*, *Pm42 Pm41*, *Pm36*, *MlIW72*, *MlZec1*, *Pm30*, *Pm26*, and *Pm16* on Chr 5BL, 7AL, 2BS, 3BL, 5BL, 7AL, 2BL, 5BS, 2BS, and 5BS, respectively (Reader & Miller, 1991; Rong et al., 2000; Liu et al., 2002; Chen et al., 2005; Mohler et al., 2005; Ji et al., 2007; Blanco et al., 2008; Li et al., 2009; Hua et al., 2009; Ben-David et al., 2010; Zhang et al., 2010). In another study, resistant gene for *Pm* was transferred from *Triticum urartu* Tum. accession UR206 to hexaploid wheat (*Triticum aestivum* L.) via crossing and backcrossing methods (Qui et al., 2005). They mapped resistant gene on the Chr 7AL (Xwmc273-Xpsp3003). *Pm1b* (*T. monococcum* to hexaploid wheat) and *Pm25* (*T. boeoticum* into hexaploid wheat) were determined using *T. monococcum* accessions on Chr 7AL and 1AL, by Hsam et al. (1998) and Shi et al. (1998), respectively. In another realm, Yao et al. (2007) determined *Pm* resistance genes (*Mlm 2033* and *Mlm80*) close to the *Pm1* locus in two different accessions of *T. monococcum* on Chr 7AL. In addition, Schmolke

et al. (2012) reported *Pm4d* (marker intervals; Xgwm526-Xbarc122) resistance allele for *Pm* using *Triticum monococcum* accession. Using a RIL population from *T. boeoticum* acc. pau5088 (resistant) × *T. monococcum* acc. pau14087 (susceptible), Chhuneja et al. (2012) explored *Pm* resistance genes: *PmTb7A.1* (DARt marker wPt4553, SSR marker Xcfa2019) and *PmTb7A.2* (STS markers MAG2185 and MAG1759) on Chr 7A. From *T. dicoccoides* to hexaploid wheat, *PmAS846* gene for *Pm* resistant was transferred by Xue et al. (2012). They indicated that *PmAS846* gene is located on Chr 5BL of wheat. Liu et al. (2012) found *MIW170* gene in the distal region of Chr 2BS3-0.84-1.00 (SSR markers; Xcfd238 and Xwmc243) using durum wheat line 81086A × IW170 (wild emmer).

On the other hand, using wild relatives of wheat, several studies were also performed to discover insect resistance genes. For this purpose, Hessian fly (Hf) [*Mayetiola destructor* (Say)] resistance gene, *H24*, was mapped to 3DL by Ma et al. (1993). Sardesai et al. (2005) determined *H32* gene on the Chr 3D in synthetic wheat “W-7984” produced from *Ae. tauschii* × a durum wheat “Altar 84.” In another study, the first emmer-produced Hf resistance gene has been mapped and characterized by Liu et al. (2005). They reported five SSR markers (Xbarc263, Xcfa2153, Xpsp2999, Xgwm136, and Xgwm33) responsible for Hf resistance gene (*Hdic*) on Chr 1A in the same region as the H11, H10, and H9 loci. They suggested that *Hdic* is a novel allele of a known *H* gene or a new gene on Chr 1A. The *H26* locus on Chr 3D was determined by Wang et al. (2006) in SW8 and SW34 with the previously determined resistance genes produced from *Ae. tauschii*. They also released SSR (Xcfd211) and TRAP markers, 7.5 cM proximal and 2.9 cM distal to *H26*, respectively. These markers will be beneficial to support the selection of the *Hf* resistance in wheat breeding studies. A glimpse of the diverse applications of QTLs/genes for different biotic stresses such as *Pm*, *Hf*, *FHB*, *Yr*, and *Lr* in wild relatives of wheat was summarized in this section. Although different researchers have discovered QTLs with the same chromosomal regions for diverse features of wheat, for breeders, further screening is required to pinpoint the candidate genes. Overall, compiling all valuable genomic information associated with QTL traits herein can be evaluated to improve cultivars having resistance to different biotic stress for revolutionizing agriculture and meeting the diverse demands of humankind.

9.4 Genome-Wide Association Studies (GWAS) in Wild Relatives of Wheat

To improve modern wheat cultivars able to adapt to changing climatic situations, investigating the genetic control of features of interest is highly important. After QTL mapping, GWAS is thought to be the next step to investigate the genetic basis related to desirable features. During the last decade, numerous GWAS have been carried out in various wheat genetic resources to uncover the genetic basis managing diverse features such as rust resistance, drought resistance, heat tolerance,

protein content, glutenins, carotenoid, color, cooking, grain micronutrients, plant height, heading date, thousand kernel weight, grain architecture, etc. In an earlier report, Laido et al. (2014) genotyped a panel of 230 tetraploid wheat lines (*Triticum turgidum* ssp.) according to the 26 SSR and 970 DArT (Diversity Arrays Technology) markers and determined 89 QTLs for plant height (PH), heading date (HD), protein content (PC), and thousand kernel weight (TKW). They detected novel QTLs on Chr 3A, 5B, and 6B for TKW; on Chr 1B, 3AL, 3BL, and 7AL for PC; on Chr 1AS for HD; and on Chr 1B, 3A, and 7B for PH. Liu et al. (2015) performed a GWAS for 29 morphological traits. They used 7,185 SNP (single nucleotide polymorphism) markers across 322 different accessions of *Ae. tauschii*. They identified 18 SNPs linked to 10 morphological traits by the general linear model (GLM) and mixed linear model (MLM). Using 7,185 SNP markers, Qin et al. (2016) conducted a GWAS for drought resistance traits in 373 *A. tauschii* accessions. They determined 25 SNPs markers by GLM and MLM analysis. Another GWAS performed by Liu et al. (2017c) using 196 emmer wheat accession for stripe rust resistance. The panels were genotyped by SNP array and detected 14 loci linked to field resistance in various environments. Arora et al. (2017) conducted GWAS using 114 non-redundant *Ae. tauschii* accessions and 5,249 SNP markers. They determined 17 SNPs linked to grain size characters separated over all the 7 chromosomes. Bhatta et al. (2018) performed another GWAS in 123 synthetic hexaploid wheat using 35,648 genotyping-by-sequencing (GBS). The results indicated that novel marker-trait associations (MTA) were determined on the A genome (45 MTA), on the B genome (11 MTA), and on the D genome (34 MTA) or haplotype blocks linked to grain yield and yield-related features in drought-stressed conditions. A panel of 243 consisting cultivars and advanced lines were evaluated by Johnson et al. (2019) for GWAS using high-density 90k Infinium SNP marker data for 24 traits. They determined 179 MTAs which are found in 95 genomic regions of all 14 durum wheat chromosomes. Moreover, significant QTLs for polyphenol oxidase activity on Chr 1A, 2B, 3A, and 3B and for gluten strength on Chr 1A and 1B were defined. In another realm, for the first time, Arora et al. (2019) performed a GWAS for micronutrients (Fe, Zn, Cu, and Mn) on *Ae. tauschii* germplasm using 5,249 GBS markers, and 19 SNP MTAs were reported for all traits. The accessions having higher Fe and Zn reported here and genomic regions can provide a jumping board for developing biofortified wheat varieties. Liu et al. (2019) reported 141 markers using 13116 DArT-seq markers for GPC in 161 wheat lines from wild emmer by GLM and grouped them into 48 QTL regions. They identified four major markers that were placed into two novel QTL regions on Chr 7BL (*QGpc.cdl-7B.2*) and 2BS (*QGpc.cdl-2B.1*). These genes and QTLs can be beneficial for improved GPC of wheat in marker-assisted breeding. Alemu et al. (2020) reported 46 QTLs for L (brightness), b (yellowness), a (redness), grain width, and grain length traits. They detected a significant QTL controlling both brightness and redness characters on chromosome arm 2AL. In another study published the same year, Marone et al. (2020) genotyped 184 durum wheat (*Triticum turgidum* subsp. *durum*) genotypes with DArT-seq markers and performed GWAS to recognize loci defining prostrate/erect growth habit. They detected two MTAs on Chr 2B (D1202558) and unmapped (D2277949)

considering LSmeans from all environments. Using 441,327 SNPs markers in a world collection of 299 *T. urartu* ex situ accessions, Talini et al. (2020) performed GWAS and reported 25 QTN for flour quality and agronomic traits. Liu et al. (2021) conducted GWAS for characterizing Zn, Fe, and Mn contents of grain in 161 advanced lines produced from *Triticum turgidum* ssp. *dicoccoides*. They reported six MTAs on Chr 3B, 4A, 4B, 5A, and 7B for Fe, three MTAs on Chr 1A and 2A for Zn, and five MTAs on Chr 1B for Mn. Considering grain shapes, Zhao et al. (2021) evaluated 221 *Ae. tauschii* accessions, utilizing 4282 SNP markers. They determined six putative candidate genes associated with grain shapes. Another GWAS for morpho-physiological characters was performed by Mahjoob et al. (2021) using 34,829 DArT-seq markers in 343 *Ae. tauschii* accessions. They identified 23 MTAs in all accessions. Ullah et al. (2021b) identified 125 MTAs for optimum condition and 142 MTAs for heat stress condition. Wang et al. (2021) found SNPs for kernel length (15 SNP), kernel width (28 SNP), kernel volume (22 SNP), kernel surface (14 SNP), kernel width-to-length ratio (21 SNP), and hundred kernel weight (13 SNP) and candidate genes (*AET4Gv20799000*, *AET2Gv20774800*, *AET5Gv20084100*, *AET5Gv20005900*, *AET5Gv21111700*, and *AET7Gv20644900*) controlling kernel traits. Aoun et al. (2021) determined 56 QTLs related to all-stage stripe rust resistance placed on all 14 durum wheat chromosomes. Recently, a HarvestPlus Association Mapping panel (HPAMP) of 293 wheat lines including landraces, *T. dicoccon*-based synthetic derivatives, *T. durum*-based synthetic derivatives, *T. Spelta* derivatives and pre-breeding derivatives produced from diverse progenitors genotyped by Baranwal et al. (2022), and determined 53 QTLs for 10 grain mineral (Ca, K, Mg, P, S, B, Fe, Cu, Mn and Zn) accumulation and 12 significant QTL for *YR* and *LR*. To the best of our knowledge, the many GWAS studies conducted in recent years using wild relatives of wheat are summarized in this section. These important breakthroughs conducted for various traits of wheat will provide a new framework for breeders to accelerate the wheat breeding by genome editing technology and MAS.

9.5 Future Perspectives

Classical breeding, consisting of many processes heavily based on crosses and phenotypic selection, has been the mostly used breeding method to date and is still the main method to develop a new cultivar. However, the impact of breeding studies, conducted using next-generation technologies combined to classical ones, has been gradually increased in recent years especially in developed countries. In a short time, it is likely that widespread use of these technologies will be also replaced to a certain extent in developing countries. In this way, a related gene can be directly transferred through gene editing and then phenotypic selection. Although it is still early for the widespread use of this genomic selection strategy, the combined approaches, like integration of speed breeding and high-throughput phenotyping with classical method, are more promising to accelerate the current breeding progress to reach the current genetic gain in wheat.

In particular, speed breeding accelerates the transition between generations by shortening the vegetation times of plants. Most of all, the generation time of long-day plants and neutral-day plants are reduced, and these plants are taken advantage of positively by obtaining more products in less time. The speed breeding was conducted on different plants such as barley (Hickey et al., 2017; Ghosh et al., 2018), oat (Ghosh et al., 2018; González-Barrios et al., 2021), quinoa (Ghosh et al., 2018), different *Brassica* species (Ghosh et al., 2018), pea (Mobini & Warkentin, 2016), soybean (Fang et al., 2021), chickpea (Ghosh et al., 2018), grass pea (Ghosh et al., 2018), peanut (O'Connor et al., 2013), amaranth (Stetter et al., 2016), *Brachypodium distachyon* (Ghosh et al., 2018), and bread/durum wheat (Alahmad et al., 2018; Ghosh et al., 2018; Watson et al., 2018; Vikas et al., 2021; Yayla et al., 2021). However, to our best knowledge, this technique has not been conducted using wild relatives of wheat. New wheat cultivars having high yield, enhanced grain quality, and resistant to stress factors can be developed using wild relatives of wheat with speed breeding via combining different modern technologies such as high-throughput phenotyping/genotyping (sensors, robotics, cameras, and computers), genomic selection, and genome editing (CRISPR/Cas9). In addition, the future pre- and main breeding studies can be led by considering the other suggestions below.

1. Crop wild relatives are great assets for human beings, and wheat wild relatives played significantly in wheat durum and bread wheat evolution. Huge numbers of wheat wild relatives are still present and need to be collected and should be conserved.
2. Thousands of wheat wild relatives are conserved in gene banks and waiting to be characterized at both phenotypic and molecular levels. It is suggested that conserved germplasm should be characterized to investigate novel variations that will be helpful for wheat breeding.
3. Efforts are required for the *in situ* and *ex situ* conservation of wheat wild relatives.
4. Wheat is one of the crops that are intensively studied, and literature reported huge numbers of marker–trait association. There is a need to validate identified genomic regions using wild relatives and convert them into KASP markers for their efficient utilization in marker-assisted breeding.
5. Genome editing emerged as the most important and powerful tool, and significant number of studies should be performed in wheat.

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Chapter 10

Socioeconomic Evaluation of Einkorn Wheat Production



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10.1 Introduction

Wheat is a cultivated plant that is the most produced in the world and indispensable in the nutrition, trade, and crop rotation systems of many countries. The cultivation area and production of wheat, which is an important plant in human nutrition, increase in parallel with population growth (Serpi et al., 2011). Today, it is accepted as a constant food by the people living in the geography from the west of Europe to the north of India, from Scandinavian and Russia to Egypt. It is one of the products with a strategic feature along with products such as corn and soy in the world food markets.

It is reported that there are historical remains in the vicinity of the Red Sea that wheat was used by hunter-gatherer human societies approximately 19,000 years ago (Tanno & Willcox, 2006; Feldman & Kislev, 2007). However, the first wheat cultivation, that is, the beginning of the cultivation of wheat, is dated 10–12 thousand years ago. The first wheat farming in the world started in the Mesopotamian region, which is located in the Fertile Crescent region, 10–12 thousand years ago in the Neolithic period, and has enabled the transition of human societies from a foraging and a sedentary lifestyle to a settled lifestyle. It is reported that the first cultivated wheat forms were diploid, einkorn ($2n = 14$, AA genome), and tetraploid, emmer ($2n = 28$, AABB genome) wheats (Shewry, 2009; Peng et al., 2011a). It has been reported that einkorn and emmer wheats are instrumental in the development and spread of wheat agriculture in the world and are an important food source for the nutrition of the people of the world in the few thousand years until the emergence of more productive polyploidy wheats (Shewry, 2009).

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Wheat and its products can be examined in two sections: hulled wheat and its products, which are known as the ancestor of another word called einkorn (*Triticum monococcum* L. ssp. *monococcum*), and other wheat and its products. The first of these, einkorn wheat, also known as “Siyez” or “Iza,” is the cultivated form of the wild wheat species *Triticum boeoticum* that grows in France, Morocco, Yugoslavia, and Türkiye. It is one of the wheat species cultivated in the early period, together with *Triticum dicoccum* Schrank wheat. It is thought to have been domesticated for the first time in Karacadağ, located in the Southeastern Anatolia region (Heun et al., 1997). Today, this wheat is still cultivated in Northern Anatolia, the Balkan countries, Germany, Switzerland, Spain, and Italy. As a result of examining the genetic relationships of einkorn and emmer wheats, it has been revealed that the gene center of these wheat species is the Southeastern Anatolia Region of Türkiye (Diyarbakır-Karacadağ region) (Heun et al., 1997; Dubcovsky & Dvorak, 2007; Shewry, 2009; Özkan et al., 2010). These wheat species are among the first domesticated plants among thousands of plant species in the Fertile Crescent (Zohary & Hopf, 2000) and grow naturally in the region (Özkan et al., 2010; Peng et al., 2011b).

Wheat farming, which started in the “Fertile Crescent” region, reached Greece and Europe (8000 BC) through Anatolia, the Balkans along the Danube, Italy, France, and Spain (7000 BC), and finally England (5000 BC), in different time periods. Likewise, it reached Central Asia and China (3000 BC) via Iran and Africa (3000 BC) via Egypt. It was brought to Mexico by the Spanish in 1529 and to Australia in 1788 (Shewry, 2009; Peng et al., 2011a). It is reported that the first wheat cultivation in the United States was made in the 1830s, but the variety known as Turkish red wheat, which was brought to the state of Kansas in the United States by the Mennonites who migrated from the Crimea, began to be grown intensively in 1874 (Quisenberry & Reitz, 1974). Until the 1940s, this variety was largely grown in the United States. Wheat was a wild character plant that was only grown/cultivated in the Middle East geography 10–12 thousand years ago, but it has spread all over the world in this time period and has become an important cultivated plant. Today, wheat farming, which has spread throughout the world, is carried out in more than 120 countries according to FAO data (FAO, 2021). It is reported that wheat cultivation is carried out between 67° north latitudes in Scandinavian countries and Russia, and 45° south latitudes in Argentina (Shewry, 2009).

According to USDA, as of 2020, a total of 775.9 million tons of wheat is produced in an area of 220.9 million hectares in the world (USDA, 2021). 44.12% of production is in Asia and 34.75% is in Europe (FAOSTAT, 2021). Roughly 95% of the production is common wheat and the remaining 5% is durum wheat (Dubcovsky & Dvorak, 2007). However, einkorn wheats (such as einkorn and Iza), emmer wheats (germik), and spelt wheats, which are the oldest forms of wheat, are not produced in a statistically significant manner today (Nesbitt & Samuel, 1995; Zohary & Hopf, 2000; Kan et al., 2016a).

Wheat is one of the important building blocks of food security. Shocks such as the food crisis in the 2000s and the COVID-19 process we are in, as well as the negativities caused by climate change and modern technology, have shown the strategic importance of wheat in food security. Although the effects of the crises are

different in every country, when it comes to food, the fact that humanity is faced with the danger of not being able to feed itself is important for all countries of the world to understand the importance of basic food sources. For this reason, genetic resources are important in ensuring sustainable production and building resistance against shocks.

Another issue as important as food security is food safety. Developing welfare levels pushes people to question food safety more. At this point, people turn to organic products, local products and tastes where traditional methods are used, where less technology and input are used, and they describe this production and products as reliable in terms of food safety. Local varieties and populations considered as genetic resources are also considered in this segment. Reliability also creates a willingness to pay, and these products are sold at higher prices in the market. Contrary to mass production, limited production creates a supply gap, and excessive demand increases prices. In fact, this can be called localization in the face of globalization, or in other words, activating local dynamics. For example, consumers regard organic products as reliable food. It is reported that the farm gate price of organically produced wheat is 50–200% higher in many European countries (Offermann & Nieberg, 2000). For this reason, genetic resources such as local wheat have an economic dimension as well as food and nutrition.

While wheat is seen only as food in many countries, it is among the sociocultural values of many countries such as Türkiye (Karabak & Kan, 2021). Both beliefs and traditions add a different value to wheat and its products and create a bond between the area where it is grown and the people. Sociologically, seeing wheat as a representative of the nutrients necessary for the continuity of human life and that bread made from wheat is attributed as sacred for this reason is a phenomenon that is not foreign at all in Turkish and Middle Eastern cultures (Bjørnstad, 2016). For this reason, wheat and its products are included in many traditions and are used symbolically, with the effect of the historical process in geographies that are the gene center of wheat.

In the historical process of wheat development, it is seen that many wheat species gain importance during certain periods. Ancient wheats such as einkorn, emmer, and spelt are used in human nutrition in many regions, primarily in Anatolia, but with the help of later developments and modern agricultural techniques, it is seen that hexaploid wheats that we use today are more widely produced and traded. Especially the fact that these newly improved wheat varieties have great advantages such as yield compared to the varieties and populations in other wheat species is one of the main reasons for the disappearance of these spelts (Kan et al., 2016a). However, studies conducted in recent years show that the nutritional content of einkorn, emmer, and spelt wheat is very rich compared to other wheats, and they are more beneficial for human health (Shewry & Hey, 2015; Arzani & Ashraf, 2017). This situation shows that local wheats, which are considered as genetic resources, are exposed to genetic erosion. In order to both limit genetic erosion and ensure the sustainability of these wheat species, it is necessary to determine the potential for commercialization of these wheats by revealing the factors that affect the producer

decisions about the production of local wheat populations in order to provide economic gain for the producers growing these types.

10.2 Economics of Einkorn Wheats

It is important to evaluate genetic resources from an economic point of view both in terms of protection and sustainability. There is an increasing interest in local products and local varieties/populations in the world (Kan et al., 2016a; Petropoulos et al., 2019; Blare et al., 2020; Lodhi et al., 2020). There are many international initiatives that try to activate local dynamics based on wheat and its products. It is important to add added value to these products, especially in studies aimed at protecting genetic resources in situ (in situ conservation). In this context, the most used applications are organic production of these products (Varia et al., 2021), protection within the geographical indication system (Raggi et al., 2021), and marketing them as finished goods in niche markets by creating a healthy food perception (Kan et al., 2021).

When einkorn wheats are evaluated, which constitute the scope of this study, Turkiye, the homeland of wheat, is one of the countries with the widest examples in this regard. There are important studies in Turkiye on Siyez and Iza wheats, which are einkorn wheats. Siyez wheat is grown in Turkiye, especially in the Kastamonu region, and is consumed by processing bulgur, as it is not suitable for making bread (it is very hard and mills cannot convert it into flour). Einkorn bulgur is a product obtained by drying Siyez wheat, which has single-grain spikelets and a husked structure, after being soaked in water and grinded in stone mills with traditional methods and sieved (Wikipedia, 2021). There are three geographical indications (PDO) belonging to Siyez wheat, which gained added value within the geographical indication system, especially in Turkiye after 2017. These are (TURKPATENT, 2021)

- Kastamonu Siyez bulgur (C2017/201)
- Kastamonu Siyez wheat (C2019/001)
- Kastamonu Siyez flour (C2018/154)

There are many local development approaches especially for Siyez bulgur in Turkiye. Siyez bulgur produced around Kastamonu is also on the agenda of many NGOs and is a wheat product supported by the Slow Food organization as a local product (Slow Food, 2021). This product is produced not only in the Kastamonu region but also in many regions of Turkiye and has been converted into economic activity in a few known places. It is used as animal feed in other areas (Karabak et al., 2019). In their study on Siyez wheat in İhsangazi district of Kastamonu province, it was determined that the producers of Siyez wheat used 68% of the Siyez wheat they produced for animal feeding and 32% for making bulgur. They stated that 11% of the produced Siyez bulgur is consumed at home and 85% is marketed.

Another example of einkorn wheat in Turkiye is Iza wheat. It is produced in Bolu and Bilecik provinces in Turkiye. It can be said that Iza wheat, which is consumed

as bulgur, is tried to be evaluated within the geographical indication system, especially with the aim of gaining added value. This wheat has two applications for geographical indication registration in Türkiye, one in 2018 (Bolu Seben Iza Wheat) and the other in 2020 (Seben Iza Bulgur) (TURKPATENT, 2021). In studies on this subject, Yaman et al. (2020) tried to make a socioeconomic analysis of production in the Black Sea region (Bolu, Karabük, Kastamonu, Sinop, and Samsun provinces) where emmer and einkorn wheats are grown intensively. As a result, they determined that 86% of the producers growing these wheats use the products for their own consumption, not for commercial purposes. It has been stated that while einkorn wheats are primarily used to meet household needs such as bread making by obtaining bulgur and flour, emmer wheat is mostly used for animal feed. In their studies, it is stated that this type of wheat should be transformed into value-added products to protect and ensure its sustainability.

In addition, there are other examples in the world where einkorn wheat is produced and sold as a local product and an economic value is created. For example, emmer, einkorn, and spelt wheats, known as spelt in Italy, are called “farro.” It is stated that new markets based on the products obtained from these wheats have developed in Italy, although it is subject to a large amount of genetic erosion (D’Antuono & Bravi, 1996). For example, it is stated that an Italian company uses spelled by-product (i.e., husks) to produce high-quality baby products (pillows, mattresses) that do not cause allergies. In addition, it is emphasized that these products are preferred with the healthy and nutritious aspects of these wheats (Laghetti et al., 2009).

Another example of einkorn wheat is in Greece known as “Kaploutzas.” This wheat is grown organically in Aridaia, Kilkis, and Thessaloniki (Region of Central Macedonia, Greece) mostly on flat and sloping lands. Papadakis (1929) reports that “Kaploutza-Einkorn Landrace” was brought to Thrace and from there to Central Macedonia with Greek refugees from Anatolia in the early 1920s. This wheat has a local niche market with its special flavor and rich nutritional content. Most farmers sell produce directly at organic public or farmer’s markets or using local distributors. Its producers are supported by the Greek Ministry of Rural Development and Food, and AEGILOPS NGO (Network for Biodiversity and Ecology in Agriculture) provides support for the protection and sustainability of this wheat species. (ECP/GR, 2021a). In this structure, it is seen that this wheat is sustainable and creates added value, especially with organic product certification, government, and NGO support.

Another einkorn wheat sample is from Romania known as “Bözödi.” This einkorn wheat was first grown in Hungary and then in Romania in Tata, Héreg, Ócsa, and Badacsony regions. “Bözödi einkorn” is traditionally cultivated in “Bözödújfalú” valley (Central Romania) on several hectares. “Bözöd” landrace on separated several hectare fields are marketing the dehulled seeds or the products (flour, pasta). The farmers and researchers working on Bözöd landrace established the “Bözöd Einkorn Society,” but now there is no activity within the Society. There is the increasing demand for the products of Bözöd landrace by the pasta and bread business, and this ensures to conserve the diversity over the long period (ECP/GR,

2021b). In this example, it is seen that Bözöd einkorn wheat is not supported by any competent authority, and the element that ensures sustainability is the demand from the private sector.

In a study conducted in Croatia, an alternative production system consisting of emmer, einkorn, and spelt wheats was compared with the wheat produced in the conventional and organic farming system by making an economic analysis. As a result of the research, it is stated that these grains are more profitable than the conventional and organic farming system. In this context, it is recommended to expand the production of these grains in the country, especially within the organic farming system (Čop et al., 2019).

Konvalina et al. (2010) evaluated the performance of einkorn, emmer, and spelt wheats in an organic farming system in the Czech Republic. The local varieties evaluated as a result of the research findings show that they can be grown under the organic farming system, especially in marginal areas for agricultural production. It is stated that producers can increase their product variety and expand their market opportunities by producing these local varieties in these areas. Thus, it has been reported that they can contribute to both sustainable development and the conservation and sustainable use of plant genetic resources.

Longin et al. (2016), in their study with the aim of agronomic comparison of emmer, einkorn, and spelt wheats with bread and durum wheats in Germany, determined that spelt, emmer, and einkorn wheats yielded 37, 55, and 62% less yield than conventional bread wheats, respectively. Despite this, they state that these wheats will be preferred to produce high-quality bread, breakfast cereals, and special breads.

10.3 Case Study from Turkiye: Einkorn Wheat (Iza Buğdayı-Iza Wheat)

Turkiye is in a very special position in terms of plant genetic resources. Among the centers of diversity and origin explained by Vavilov (1994), the Mediterranean and Near East Centers overlap in Turkiye. In terms of plant gene resources, Turkiye is one of the richest countries in the world. According to J. Harlan, there are five micro-gene centers in our country where more than 100 species show wide variation (Demir, 1990). Turkiye's rich biodiversity stems from being an important gene center and home to many plant and animal species. The most important of these plant species is "wheat."

Wheat, which has a history of 10,000 years in Anatolia (Harlan, 1995), is a cultural heritage as well as being a strategic product. Turkiye is one of the important regions where wheat is cultivated (Zohary & Hopf, 2000). In Turkiye, wheat has an economic, social, cultural, historical, and even archaeological value and importance. The history of wheat in Turkiye goes back before all civilizations. Wheat is the value at the center of the revolutionary changes that determine the way of life that man has reached today, and moreover, the changes that have taken place in the

geography we live in today. There are historical remains showing that wheat is the homeland of the Fertile Crescent, especially in the Southeastern Anatolia Region of Turkiye (Nesbitt & Samuel, 1995; Tanno & Willcox, 2006).

Wheat is the most important agricultural product in Turkiye, and when products such as bread, phyllo, and bulgur are considered, it can be said that Turkish cuisine is one of the indispensable foodstuffs. When the statistical data of institutions such as TURKSTAT and FAO are examined, it is seen that Turkiye's annual wheat production is approximately 20 million tons, which corresponds to an agricultural production value of 7 billion dollars. In terms of added value, it can be said that the agricultural industry based on wheat and wheat products is one of the main sectors in the food industry and economy.

10.3.1 Research Area

In this case study, the socioeconomic evaluation of Iza wheat production in *Triticum monococcum* subspecies, which is one of the einkorn wheats belonging to *Triticum monococcum* L. ssp. *monococcum* wheat species, and effective management strategies on the decisions of the operator in wheat production in the enterprise are emphasized.

The main material of the research consists of the data obtained through the survey conducted with the agricultural enterprises producing Iza wheat (einkorn wheat) in Gölpazarı and Pazaryeri districts of Bilecik province and Göynük, Merkez, Mudurnu, and Seben districts of Bolu province. The research area is shown in Fig. 10.1. Bolu and Bilecik provinces, which are defined as research areas, are the places where Iza wheat is widely cultivated in Turkiye. Especially in Bolu province, there are two geographical indication registration applications under the names of "Bolu Seben Iza Wheat" and "Seben Iza Bulgur" (TURKPATENT, 2021). For this reason, this wheat is one of the hulled wheats known in both regions and has been produced for a long time.

10.3.2 Sampling Method

Since it is not possible to reach the records and quantitative data of the producers using local wheat varieties, the "Snowball Sampling Method," which is one of the non-probabilistic sampling varieties, and the chain transportation principle were used in this study. In the selection of this method, due to the difficulty of determining the producers using unregistered local wheat varieties, attention was paid to reaching the other one through the first producer. Snowball sampling has been particularly effective in identifying individuals or situations that can be a rich source of information. In snowball sampling, it is aimed to establish a relationship with the sample event that can be included in the universe and suitable for the purpose of the

research, and then with the help of the person contacted, with the help of another case, and then with another case study in the same way, and in this way, it is aimed to enlarge the sampling like the snowball effect (Coşkun et al., 2017; Kuş, 2012). This approach is particularly effective in identifying individuals or situations that can be a rich source of information regarding the researcher’s problem (Fig. 10.2).

In the study, the opinions of experts (Provincial Directorates of Agriculture and Forestry, District Directorates of Agriculture and Forestry, Chambers of Agriculture and University) were taken to determine the number of producers in the provinces



Fig. 10.1 The map of research area

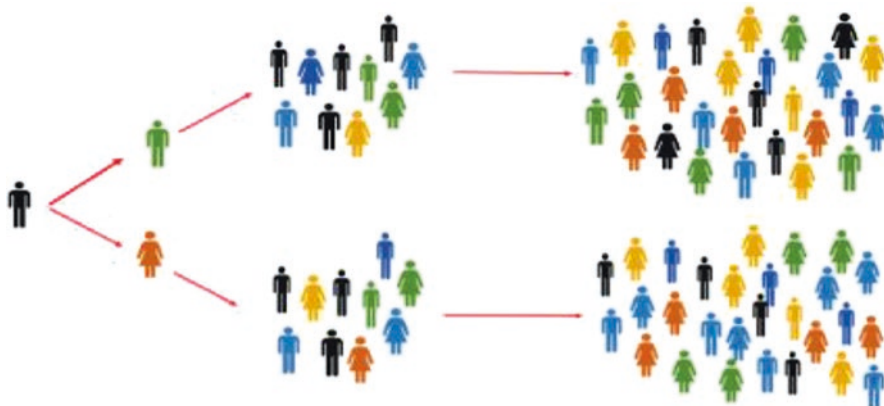


Fig. 10.2 Schematic of the snowball sampling model

in the research area. As a result of the study, a total of 51 Iza wheat producers were reached and the questionnaires were filled in face to face.

10.3.3 Research Aims

Structured questionnaire questions were used in the study, and the following information was tried to be reached with the study:

- Socioeconomic and demographic characteristics of Iza wheat growers
- Agricultural structure of Iza wheat growers and determination of the place of Iza wheat in production
- Iza wheat production strategies and reasons for preference
- Determination of production cost of Iza wheat
- Evaluation of Iza wheat

10.3.4 Analyze Methods

In the study, producers were divided into two groups. These are “only einkorn (Iza) wheat producers – EWP” and “both einkorn (Iza) wheat and improved wheats producers – BWP.” Statistical analyses were done on these two groups. In the study, *t*-test statistics, which is one of the parametric tests, was used when the assumptions of the normal distribution were met in the comparison of the means of two independent groups. In cases where the assumptions of the normal distribution were not met, the Mann–Whitney *U*-test, which is one of the nonparametric tests, was used. The chi-square test of independence was used to test the interdependence of two discrete variables (Kesici & Kocabaş, 2007).

10.3.5 Research Findings

Iza wheat, one of the important einkorn wheats grown in Türkiye, is produced especially in the provinces of Bolu and Bilecik. These provinces are in TR4 East Marmara Region according to the Türkiye Statistical Regional Units Classification. In addition, Iza wheat, which is produced in other provinces, is called by different names according to the regions (Kan et al., 2016a; Zencirci et al., 2020; Yaman et al., 2020). For example, it is known as Siyez wheat in and around Kastamonu province. IZA wheat, which set out from Karacadağ, reached the province of Bolu and its districts thanks to migrations and trade routes, and gained a unique genetic structure compatible with this environment by being cultivated here for many years. Siyez and Iza wheats are often confused with each other. According to Zencirci

et al. (2020), the name Siyez is the species name (*Triticum monococcum* ssp. *monococcum*), and Iza wheat is a village variety (wheat landraces) under this species.

In this study, there are two types of production strategies of the producers. While 41.18% of the producers produce only Iza wheat, the remaining 58.82% produce Iza wheat (einkorn wheat) and other improved wheats (*Triticum aestivum* and *Triticum durum*) together (Fig. 10.3). These behaviors of producers developed for both wheat species, which have advantages and disadvantages relative to each other, can also be called risk management strategy.

The demographic characteristics of the Iza (einkorn) wheat producers interviewed in the research region according to their production strategies are given in Table 10.1. When the table is examined, it is seen that the age of the producers dealing with the production of Iza is over 50. In particular, it was determined that the producers that implement the BWP strategy are younger than those that implement the EWP strategy and this difference is statistically significant at the 99% confidence level. Kan et al. (2016a) stated in their study with wheat landrace producers in Turkiye that the average age of wheat landrace producers was over 50 years old. They reported that the average age of producers producing only wheat landraces was even higher. When both research findings and other studies are examined, it can be said that age is an important variable in both decision-making and production behaviors in wheat landrace production. Especially the older population has a higher tendency to produce only wheat landrace, and as a result of the research, similar results were obtained in the production of Iza wheat. In recent studies on the age of producers engaged in agriculture in Turkiye, it is stated that the average age varies between 46 and 51 years (KKB, 2020). This situation shows that the population dealing with Iza wheat production is above the average farmer age of Turkiye.

Another demographic factor is the education level of the producers. It has been determined that more than 85% of Iza wheat producers in the research area have education between primary–secondary and high school. The average education year is 6.80 years. The academic year was higher especially in producers who implemented the BWP strategy, and the difference was statistically significant at the 95% confidence level. As the education level rises, producers tend toward more

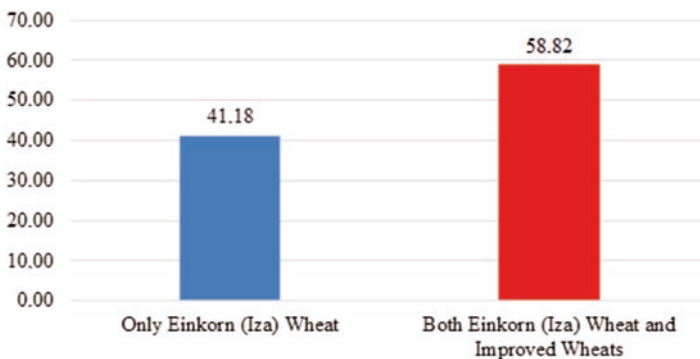


Fig. 10.3 Production strategies of the Iza wheat producers

Table 10.1 Demographic statistics of the farmers by their production strategies

Variables		Production strategy					<i>t</i> -test/ chi-square test	
		Only einkorn wheat (Iza wheat) – EWP		Both einkorn (Iza) and improved wheat varieties – BWP		Farm average		
		Mean	%	Mean	%	Mean		%
Household heading age		58.90		50.07		53.71		2.87***
Education	Illiterate		0.00		0.00		0.00	3.80
	Literate		9.52		3.33		5.88	
	Primary– secondary school		71.43		53.33		60.78	
	High school		14.29		36.67		27.45	
	Vocational school		0.00		0.00		0.00	
	Higher education		4.76		6.67		5.88	
Education (year)		5.48		7.73		6.80		–2.24**
Household number (total)		3.00		3.77		3.45		–2.30**
Household number (male)		1.48		1.90		1.73		–2.05**
Household number (female)		1.52		1.87		1.73		–1.59
Man power unit (total)		2.07		2.79		2.50		–2.63**
Man power unit (male)		1.19		1.58		1.42		–2.09**
Man power unit (female)		.88		1.22		1.08		–2.28**

Statistically significant at *90%, **95%, and *** 99% confidence levels

commercial wheats. Kan et al. (2016a) stated in their study that as the education level increases the producers tend to produce more improved wheat.

Household size and man power unit (MPU) size are the other factors in demographic factors examined in the research. It makes sense when both factors are evaluated together. It has been determined that the size of the household and, accordingly, the MPU size is higher, especially in the producers who carry out the BWP strategy. This is since producers tend to meet their labor needs from the family. The increase in MPU leads producers to different products, not to a single product. As can be seen in the table, there is a statistically significant difference between the producers producing both production strategies in terms of these two factors.

The land assets and land patterns of the producers in the research area are shown in Table 10.2. When the table is examined, it has been determined that the average land size in the average of agricultural holdings is 13.09 ha and 14.52% of this land is allocated for Iza wheat production. Iza wheat is produced in the rainfed farming system. It has been determined that the producers following the EWP production strategy are engaged in agricultural activities on a smaller scale than the producers applying the BWP production strategy. The difference between the two different

Table 10.2 Land assets of the Iza wheat producers

Variables	Production strategy			<i>t</i> -test/Mann–Whitney <i>U</i>
	EWP	BWP	Farm average	
	Mean	Mean	Mean	
Total cultivated area (ha)	6.01	18.05	13.09	−4.80***
Own area (ha)	2.92	7.66	5.71	−3.91***
Rented area (ha)	3.09	9.31	6.75	−2.84***
Sharecropping area (ha)	0.00	1.08	0.64	−0.83
Own irrigated area (ha)	0.39	0.96	0.72	−0.63
Rented irrigated area (ha)	0.63	0.51	0.56	0.22
Sharecropping irrigated area (ha)	0.00	0.00	0.00	–
Einkorn (Iza) wheat production area (ha)	1.38	2.26	1.90	1.63*
				188.00** (M-W-U)

Statistically significant at *90%, **95%, and *** 99% confidence levels

types of farms in terms of both the size of the cultivated land and the size of the land allocated to the production of Iza wheat was found to be statistically significant. It has been determined that the producers following the EWP production strategy allocate 22.96% of the total cultivated land to Iza wheat production, while the producers following the BWP production strategy allocate 12.52% of the total cultivated land to Iza wheat production. The latest data on the scale of agricultural holdings in Turkiye show that they have an average size of 7–8 ha (TURKSTAT, 2018). Considering the size of the owned land in the research region, it is seen that the size of the agricultural holdings is below the Turkiye average. It has been determined that the producers following the BWP production strategy rented a significant amount of land for agricultural production. According to Kan et al. (2021), they determined the average land size of wheat landrace producers in Turkiye as 7.16 ha and as 6.11 ha in Eastern Marmara Region in their study. Yaman et al. (2020), in their study on emmer and Einkorn wheats, reported that the average farm size was 7.97 ha. As can be seen from the studies on wheat landraces producing agricultural holdings, as the size of the agricultural holdings increases, the agricultural holdings tend to combine the wheat landrace production with the commercial (improved) wheat production. In other words, the production strategies of the producers change with the size of the agricultural holdings. There is a positive relationship between agricultural holding size and the adoption of modern varieties (Perrin & Winkelmann, 1976; Feder et al., 1985; Kan et al., 2016b). These research findings also support this result.

One of the most important aspects of Iza wheat production is the economic return of production. In this context, expense items were calculated according to the inputs and practices used by the producers in the production of Iza wheat and are presented in Table 10.3. Cost items are determined only on variable costs and fixed costs were not considered. As a result of the analysis, the average variable cost per hectare of agricultural holdings was calculated as \$641.96, and the gross agricultural production value was \$156.56. There was no statistically significant difference between the

Table 10.3 Cost items of Iza wheat production

	Production strategy						t-test value
	EWP		BWP		Farm average		
	TL	\$ ^a	TL	\$	TL	\$	
Soil preparation cost (TL/ha)	511.43	72.85	440.00	62.68	469.41	66.87	1.50
Planting cost (TL/Da) (including bottom fertilizer)	1899.05	270.52	1920.00	273.50	1911.37	272.28	-0.27
Fertilizer and fertilization cost (TL/ha)	1044.76	148.83	1159.67	165.19	1112.35	158.45	-0.72
Plant protection cost (TL/ha)	138.10	19.67	92.00	13.11	110.98	15.81	1.89*
Harvest cost (TL/ha)	307.14	43.75	326.67	46.53	318.63	45.39	-3.22***
Threshing cost (TL/ha)	365.71	52.10	371.67	52.94	369.22	52.59	-0.84
Revolving fund interest (5%)	213.31	30.39	215.50	30.70	214.60	30.57	
Total variable cost	4479.50	638.11	4525.50	644.66	4506.56	641.96	0.18
Straw yield (kg/ha)	2438.10		2477.78		2461.44		-0.84
Grain yield (kg/ha)	2467.62		2633.33		2565.10		-0.79
Gross agricultural production value (TL/ha)	1082.53	154.21	1110.57	158.20	1099.02	156.56	-1.08
Gross profit (TL/ha)	634.58	90.40	658.02	93.73	648.37	92.36	-0.71

Statistically significant at *90%, **95%, and *** 99% confidence levels

^a1\$:7.02 TL (Central Bank of the Republic of Turkiye, 2021-year average Exchange rate data)

variable costs of Iza wheat and the gross agricultural production value of the producers following both EWP and BWP production strategies. One of the most important subjects is related to the yield of Iza wheat. According to TURKSTAT 2020 data, Turkiye's average wheat yield was 2920 kg/ha, Bolu province was 2970 kg/ha, and Bilecik province was 2600 kg/ha (TURKSTAT 2021). Considering these figures, the yield of Iza wheat is below the average wheat yield. The other subject in the region is that Iza wheat is supported by the Bolu Municipality, especially in the province of Bolu, and contracted agriculture with a purchase price above the market price. It was determined that Bolu Municipality made a purchase from contracted producers for 512.82 \$/ton in 2020. According to TURKSTAT 2020 data, the average wheat price in Turkiye is 213.68 \$/ton, Bolu is 186.61 \$/ton, and Bilecik is 226.50 \$/ton (TURKSTAT, 2021). Given these data, the producers consider wheat production economically sustainable and continue to produce.

Studies on wheat landraces show that the need for family consumption is an effective factor in the sustainability of the wheat landrace production (Frison et al., 2011; Kan et al., 2021; Jaradat, 2017; Karabak et al., 2019; Yaman et al., 2020). In the research, the evaluation methods of Iza wheat and improved wheat were examined. In the research area, 15.69% of the Iza wheat producers do not sell the Iza wheat anywhere, and they use it only for family needs. The ways of using Iza wheat of the remaining 84.31% are given in Fig. 10.4 in detail. When the figure is examined, it has been determined that 78.63% of the Iza wheat production was sold to the "Köroğlu Cooperative," which was established under the leadership of Bolu Municipality. The Cooperative, supporting Iza wheat producers in Bolu province,

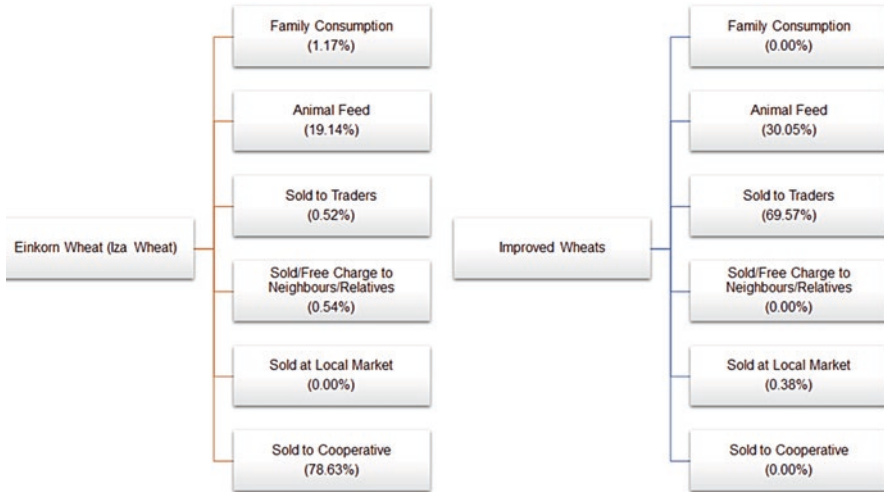


Fig. 10.4 Evaluation of einkorn (Iza) wheat and improved wheats

supports the farmers by contracted production method and purchases the product from the producer at a higher price than commercial wheats in the market. For this reason, commercialization potential in Iza wheat production in Bolu province is higher than Bilecik province. In Bilecik, 63.64% of the producers stated that they produce only for their own family needs. In addition, animal feeding is an important factor in the production of Iza wheat, and it has been determined that the producers use 19.14% of the production for animal feeding. Regarding improved wheat, traders have a significant share in product evaluation (69.57%). As can be seen from the figure, NGOs play an important role in turning wheat landraces into economic advantages. The sample in Bolu is an important initiative for both the expansion of the production of Iza wheat and the conservation and sustainable use of genetic resources. In addition, the Cooperative’s attempts to create a brand by transforming Iza wheat into value-added products give hope that this initiative will contribute significantly to regional producers and local economic development in the future.

In Fig. 10.5, the variables that can be effective in the production decision of the Iza wheat and commercial wheat farmers are shown comparatively. The variables were evaluated with 5-point scoring (5 – very effective factor in decision-making in production). When the figure is examined, it has been determined that the factors “drought tolerance,” “cold tolerance,” “tolerance to diseases,” “tolerance to pests” are effective in decision-making, especially in the production of Iza wheat. These factors show that Iza wheat has an advantage over commercial wheats under biotic and abiotic stress conditions. Studies on this subject report that wheat landraces are preferred by producers as they are more tolerant to biotic and abiotic conditions (Meng et al., 1998; Williams, 1989; Jarvis et al., 2000; Bardsley & Thomas, 2005; Kan et al., 2021; Jaradat, 2017) and has greater adaptability to a range of soil types (Bellon & Taylor, 1993).

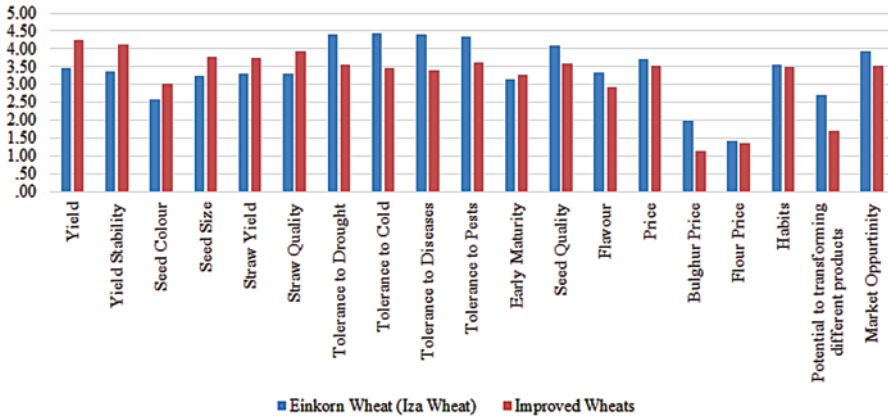


Fig. 10.5 The reasons for preference by the farmers for einkorn (Iza) wheat and improved wheats

10.4 Conclusion

Considering both its importance in human nutrition and its indispensability in animal production with its grains and stems, wheat is one of the strategic products. With the development of modern technology and science, we can say that today there is a quantitative increase in wheat, thanks to plant breeding methods. With this development, it is seen that modern wheat varieties have entered our lives more and the industry based on this also demands these varieties. However, the crises experienced lead people to become more conscious about healthy consumption and to produce higher quality products by using more environmentally friendly techniques. In this regard, the increasing perception turns into more attitudes and behaviors, that is, there is a shift in preference toward the products that create the quality perception of the consumers. In this context, concepts such as local product, niche product, geographically indicated product, traditional product, and organic product have become more audible in the market, and these concepts have begun to be brought together with the perception of healthy products. In this context, local seeds and local varieties/populations have become preferred in more production and consumption.

In this new market created, the concept of healthy products has gained an important place in an increasing trend today. The COVID-19 pandemic we are currently experiencing has an important contribution to the development of this new trend. This situation also provides a positive contribution to the conservation and sustainable use of local varieties (landraces) important in terms of genetic resources. Spelled wheat is one of these genetic resources (einkorn, emmer, spelt). The results of the research on these wheats showed that these wheat types have quantitative advantages when compared with the existing wheat varieties, and this new trend led to the inclusion of spelt. In addition to the studies carried out in Türkiye in the Fertile Crescent, which is known as the homeland of wheat, findings such as the

positive effects of these wheat species on human health in Europe, their resistance to climate change, and their resistance to biotic and abiotic stress conditions have made these wheats more popular.

Iza wheat, which is one of the einkorn wheats and grown around Bolu and Bilecik provinces in Türkiye, is one of the spelled wheats that has been mentioned frequently in recent years. Studies in the world focus on organic agriculture, geographical indication systems, government and NGO supports, as well as marketing them into value-added products in order to protect and sustainably use such genetic resources. In the case study examined within the scope of this study, the geographical indication system and the contribution of NGO were emphasized. In this way, a local development strategy based on this wheat has been tried to be created.

The involvement of the municipality, cooperatives, and the private sector in supporting Iza wheat is one of the best examples of how other sectors can achieve this without just creating a public policy. Especially in the province of Bolu, which is discussed within the scope of the study, purchasing the products of the producers at high prices and making them into finished products ensure that the difference created by the loss of efficiency is arranged and they remain in production. A shift from household consumption to commercial production is seen as a positive development in local economic development in the region. Local actions are important for the protection and sustainable use of wheat landraces, which is now in danger of disappearing in Türkiye. In this way,

- It contributes to regional recognition by allowing products with high added value to enter the market more, while providing more economic return.
- It develops social cooperation between producer, public, NGO, and private sector.
- It increases more tendency toward organization and improves organizational commitment.
- It strengthens social capital.
- By supporting environmentally friendly production systems (organic agriculture, geographical indication system, etc.), it contributes to combating climate change, reducing environmental damage from the use of inputs, and evaluating marginal areas.
- It ensures the preservation of cultural values and the continuity of ancestral information.

Considering the importance of wheat in the nutrition of the people of the country and in terms of farming, this strategic product is indispensable even if the geographical and climatic conditions are difficult, and it reveals the necessity of creating product-based development strategies. The fact that local wheat varieties find buyers, especially in niche markets, shows that the advantages of niche marketing will be effective for wheat. Local development strategies that can be created will allow in situ conservation of wheat landraces, as well as provide economic gain for the producers of the product.

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