Chapter 12
Influence of Genetics on the Secondary Metabolites of Plants

Jyoti Singh Jadaun, Ritesh Yadav, Nisha Yadav, Shilpi Bansal, and Neelam S. Sangwan

Abstract Secondary metabolites are natural products and have been attributed with the diverse roles in plants such as survival, interaction with environment, protection from biotic/abiotic stress conditions and as volatile signalling molecules. Secondary metabolites are broadly classified into chemical classes such as terpenoids, alkaloids, phenylpropanoids, steroids etc., and are known to be modulated by various factors, including physiological, genotypic, and environmental factors. These secondary metabolite compounds are biosynthesized via different pathways specific to each class of chemical entities produced in plants. The biosynthetic pathway operates in a linear or branched fashion where any alteration/modulation in the pathway or single gene or group of gene influences the production and accumulation of these metabolites. It has been well established that up or down regulation of concerned genes brought about by natural or artificial means subsequently leads to alteration in enzymatic activities responsible for secondary metabolite synthesis. There has been considerable progress in literature on recombinant expression of pathway genes and its association in enhancement of secondary metabolites in plants. Equally interesting is to understand the genetic impact of secondary metabolites in plants. It has been shown in some earlier studies that genetics play a key role in defining composition as well overall yield of secondary metabolites in plants. There is impact of genotype and environment interaction in secondary metabolite levels that will determine the overall expression of a distinct chemotype/genotype-based concept. A large number of genotypes with characteristic composition have been identified to be developed as key varieties suited for defined metabolites. Such examples exist in

J. S. Jadaun
Department of Botany, Dayanand Girls Postgraduate College, Kanpur, Uttar Pradesh, India

R. Yadav
National Institute of Plant Genome Research, New Delhi, India

N. Yadav · N. S. Sangwan (✉)
Department of Biochemistry, Central University of Haryana, Mahendergarh, Haryana, India

S. Bansal
ICAR – Indian Agricultural Research Institute, Pusa, Delhi, New Delhi, India

M. Carocho et al. (eds.), *Natural Secondary Metabolites*,
https://doi.org/10.1007/978-3-031-18587-8_12

plants such as *Artemisia* annua, *Mentha* species, *Ocimum* species, *Withania somnifera* and several others, where valuable metabolites were shown to be genetically associated. The aim of the current article is to comprehend this less characterized area of secondary metabolism in plants and develop a knowledge resource involving its biosynthesis, accumulation, regulation with reference to their genetic background which in future could be utilized for commercial and pharmacological applications in aroma and drug industries.

12.1 Introduction

Plant secondary metabolites (PSMs) are one of the most diverse group of compounds as they have tremendous diversity in their structure and functionality. Plants produce a plethora of low molecular weight organic compounds, being classified as primary and secondary on the basis of their direct involvement in growth and development of plants. Earlier PSMs are considered less important for the growth and development, being termed as "secondary." Progressive research in this area revealed their role in interaction with the environment and in defense mechanism, so the term "specialized" will be more appropriate instead of "secondary" (Qari and Tarbiyyah 2021). Overall there is no boundary to distinguish the plant metabolite as primary or secondary because current understanding of plant metabolic engineering extends their area of functionalization (Sangwan et al. 2018; Erb and Kliebenstein 2020). These metabolites are of economically as well as ecological importance, therefore, special attention on their production is needed (Sangwan and Sangwan 2014; Sangwan et al. 2010). Moreover, there is no straight pathway to define their synthesis and degradation and it is often observed that these metabolites are restricted in their distribution and it may be limited up to plant family, genera, or species level and even at the tissue specificity (Fang et al. 2012; Jadaun et al. 2020; Kushwaha et al. 2013a). In some plants, they accumulate in special structures like trichome, vacuole, or specialized glands (Yadav et al. 2014; Yu et al. 2018; Maurya et al. 2019). These metabolites are not only crucial for plant life but also play significant role for humans. Pharmacological activities of these compounds made them attractive for industrial applications. Various biotechnological approaches are followed to enhance their production (Fierascu et al. 2020; Srivastava and Sangwan 2020; Narnoliya et al. 2021).

Secondary metabolites from plants are governed by several factors such as physiological, metabolic, environmental, and genomic (Sangwan et al. 2001a, 2017; Farooqi et al. 2000; Sangwan and Sangwan 2000; Singh et al. 2015; Srivastava et al. 2015; Shukla et al. 2003; Yadav et al. 2015). Conclusively, metabolism of SMs is the result of interaction between gene (G) and the environment (E) (Padilla-González et al. 2019). Generally, these metabolites are produced by plants in substantial amounts but variation in quantity and quality is noticed under fluctuation of biotic and abiotic factors (Sangwan et al. 2001a; Akula and Ravishankar 2011; Mishra et al. 2020). Another major factor responsible for

accumulation pattern of SMs is the genotypic constitution (Maurya and Sangwan 2020). Polyploidy has gained the interest of numerous researchers due to various reasons: induction of polyploidy can be utilized to create genetic diversity in targeted crops, thus it has the potential to create new species. The proliferation of new species and genomes and the possible benefits of compatibility may have an impact on species evolution.

Genetic diversity of plants plays critical role in regulation of their metabolism either its primary or secondary. Quantitative trait locus (QTL) analysis is useful statistical method to employ the relationship between genotype and phenotypes (Miles, C. & Wayne). Metabolic quantitative trait loci (mQTLs) plays an important role to decode the involvement of gene in the production of a metabolite (Alseekh et al. 2015). There are different kinds of molecular markers which are available to record the genetic diversity in medicinal plants such as Simple Sequence Repeats (SSR), DNA polymorphism like Random Amplified Polymorphic DNA, and more specified ones such as Restriction Fragment Length Polymorphism, Amplified Fragment Length Polymorphism, and Single-Nucleotide Polymorphism (Sarwat et al. 2012) (Table 12.1). Now days Expressed Sequence Tag (EST) database is utilized to design the markers (Narnoliya et al. 2017).

In this chapter, we will discuss various factors contributing towards the accumulation of secondary metabolites in plants and also the influence of the genetic variability with respect to their association with the secondary metabolite production.

12.2 Influence of Genetics on Secondary Metabolites

Secondary metabolites are classified into three categories: alkaloids, terpenoids, and phenyl propanoids. We will discuss these compounds in detail with special reference to the effect of genetics on the secondary metabolites using examples of leading and important medicinal and aromatic plants (Table 12.2).

12.2.1 Withania Species *(Solanaceae)*

The genus *Withania* belongs to Solanaceae family and it is used in traditional medicines since ancient times. It has immense importance as an ingredient in different medicinal systems like Ayurveda, Unani, and Siddha medicinal practices. It shows health beneficial effects by controlling the aging process through its rejuvenating properties. Genus *Withania* comprises approximately 23 species, but only few species get attention for their usage and popularity. *W. somnifera and W. coagulans* are two of most studied species (Kushwaha et al. 2013b; Sangwan et al. 2014). Research is carried out significantly on *W. somnifera* followed by *Withania coagulans* (Mishra et al. 2013; Mishra et al. 2016; Jadaun et al. 2017a,

Table 12.1 List of selected plants showing the assessment of genetic diversity by using molecular markers

Plant	Family	Constituent	Marker	Application	Reference
Dioscorea pentaphylla L	Dioscoreaceae	Diosgenin	Start codon targeted	Clonal fidelity in tissue culture raised plants	Manokari et al. 2022
Ocimum spp	Lamiaceae	Citral	RAPD	Genetic diversity	Vieira et al. 2003
O. basilicum	Lamiaceae	Linalool, eugenol, cineole	AFLP	DNA genotyping, authentication	Labra et al. 2004
O. gratissimum	Lamiaceae	Eugenol, thymol, geraniol, xantomicrol, cirsimaritin	RAPD	Genetic diversity	Vieira et al. 2001
O. gratissimum	Lamiaceae	Eugenol, thymol	ISSR	Genetic diversity, Species identification	Kumar et al. 2019
Ocimum spp	Lamiaceae	Aliphatic acids, eugenol, thymol	RAPD	Genetic diversity	Chowdhury et al. 2017
Ocimum spp	Lamiaceae	Caryophyllene, alpha-caryophyllene, and linalool	EST-SSR	Diversity analysis, tagging of traits	Mahajan et al. 2015
O. basilicum and O. tenuiflorum.	Lamiaceae	Ursolic acid and oleanolic acid	duplex PCR assay	Authentication	Travadi et al. 2021
O. africanum	Lamiaceae	–	ISSR	Genetic variability	Makmur et al. 2020
W. somnifera	Solanaceae	withaferin A, withanone, withanolide D or withanolide A	RAPD	Genetic diversity	Chaurasiya et al. 2009
W. somnifera	Solanaceae	withanolides and withaferin A	AFLP	Genetic analysis	Dhar et al. 2006
Withania somnifera	Solanaceae	–	ISSR	Genetic diversity	Hiremath et al. 2021

Withania somnifera	Solanaceae	–	RAPD and ISSR	clonal fidelity, Genetic diversity	Nayak et al. 2013; Tripathi et al. 2012; Khan and Shah 2016
Withania somnifera	Solanaceae	–	EST	Genetic diversity	Parita et al. 2018
Artemisia annua *Artemisia* spp.	Asteraceae	Artemisinin	OPGMA-RAPD	Analysis of artemisinin and chemotypic variants	Sangwan et al. 1999
A. parviflora, *A. vulgaris L.,* *A. myriantha and* *A. nilagarica*	Asteraceae	Artemisinin	RAPD, ISSR, and IRAP	Analysis of artemisinin and chemotypic variants	Nganthoi and Sanatombi (2019)
Boerahvia diffusa	Nyctaginaceae	Rotanoids, punarnavin, steroids	RAPD	Mutant characterization, Genetic diversity	Shukla et al. 2001, 2003
M. cervina	Lamiaceae	Pulegone	ISSR	Morphological traits, EO constituents profile, and chemotypic variants	Rodrigues et al. 2013
Mentha spicata, *M. piperita, M. suaveolens,* *M. longifolia, M. aquatica,* *M. x piperita, M. arvensis* *subsp. Arvensis, M. spicata* *var. crispa,*	Lamiaceae	–	RAPD	Genetic diversity	Momeni et al. 2006; Virginia et al. 2021
M. spicata accessions (C30 and C33, *M. arvensis* accessions (C17 and C18), interspecific hybrid	Lamiaceae	–	RAPD and AFLP	Genetic diversity	Shasany et al. 2005
Mentha species	Lamiaceae	–	RAPD	Genetic diversity	Shinwari et al. 2011

(continued)

Table 12.1 (continued)

Plant	Family	Constituent	Marker	Application	Reference
Bupleurum spp	Apiaceae	Bupleurumol, saikosides	rDNA ITS	Species identification	Xie et al. 2009
Catharanthus roseus	Apocynaceae	vincristine and vinblastine	RAPD, ISSR, EST-SSR	Genetic linkage map	Gupta et al. 2007
Cymbopogon species	Poaceae	Citral, trans, geraniol	RAPD	Genetic diversity, Clonal fidelity	Sangwan et al. 2001, 2003;
Coscinium fenestratum	Menispermaceae	Berberine	18S r-RNA; PCR-RFLP	Authentication	Watthanachaiyingcharoen et al. 2010
Cynara cardunculus	Asteraceae	Cynarin, apigenin, luteolin	AFLP, SSR	Genetic diversity	Mauro et al. 2009
Datura spp	Solanaceae	Atropine, hyoscyamine and scopolamine	sequencing; microarray	Authentication	Carles et al. 2005
Echinacea laevigata Echinacea	Asteraceae	Cichoric acid and tetraene	AFLP	Analysis of population genetic structure and mating system	Peters et al. 2009
Euphorbia spp.	Euphorbiaceae	Jatrophanediterpenes	sequencing	Authentication	Xue et al. 2007
Fragaria vesca	Rosaceae	Biflavansabscisic acid	SCAR	Detection of seasonal control of flowering	Albani et al. 2004
Matricaria chamomilla	Asteraceae	Chamomillol, Chlorogenic acid, Chrysoeriol	RAPD	Genetic variation	Solouki et al. 2008
Origanum spp	Lamiaceae	carvacrol, thymol, limonene, pinene, ocimene, and caryophyllene	RAPD	Quality control	Marieschi et al. 2009
Salvia miltiorrhiza	Lamiaceae	Salvinorin	ISSR, SRAP	Genetic diversity	Song et al. 2010

Tribulu sterrestris	Zygophyllaceae	Harman	AFLP, SAMPL, ISSR, RAPD	Genetic diversity	Sarwat et al. 2008
Vitex rotundifolia	Lamiaceae	Vitexfolin A, B, and C	ISSR-PCR	Genetic variation and quality control	Hu et al. 2007
Withania somnifera	Solanaceae	Withaniolide	AFLP	Genetic variation and relationship	Negi et al. 2000
Trollius spp.	Ranuculaceae	orientin, vitexinand quercetin-3-*O*neohesperidoside	RAPD	Genetic diversity	Li and Ding, 2010

Table 12.2 Effect of polyploidy on secondary metabolites production

Plant Name	Common name	Family	Classification/ Clade	Ploidy	Name of Molecules	Molecule conc.	Reference
Agastache foeniculum	Blue giant hyssop	Lamiaceae	Angiosperms	Tetraploid	Essential oil (Methyl chavicols)	50% enhancements	Madani et al. 2021; Talebi et al. 2017; Talebi et al. 2016
Arabidopsis thaliana	Arabidopsis	Brassicales	Angiosperms	Polyploid (autotetraploid, allotetraploid)	γ-aminobutyric acid (GABA)	–	Soltis et al. 2015; Madani et al. 2021; Tavan et al. 2022; Van de Peer et al. 2021; Iannicelli et al. 2020
Artemisia annua	Annual wormwood	Asteraceae	Angiosperms	Polyploid (tetraploid)	Artemisinin (Sesquiterpene); essential oil	38% enhancements; 35.6% enhancements	Madani et al. 2021; Yunus et al. 2018; Yadav et al. 2015; Iannicelli et al. 2020
Atropa belladonna	Deadly nightshade	Solanaceae	Angiosperms	Polyploid (allohexaploid)	Tropane alkaloids	68% enhancements	Madani et al. 2021; Volkov et al. 2017; Lavania 2005
Avena sativa	Oats	Poaceae	Angiosperm	Polyploid (allotetraploid)	Avenacins, avenanthramides	–	Tate et al. 2005; Iannicelli et al. 2020; Kennedy et al. 2020
Cichorium intybus	Chicory	Asteraceae	Angiosperms	Tetraploid	Phenolic compound, Chlorogenic acid	10 times	Madani et al. 2021; Street et al. 2013
Coffea arabica	Coffee	Rubiaceae	Angiosperms	Polyploid (tetraploid)	Caffeine	Decrease	Tate et al. 2005; Iannicelli et al. 2020; Lavania 2005
Cymbopogon species	Lemongrass	Poaceae	Angiosperm	Diploid, tetraploid	Citral, geraniol, gernial, geranyl acetate	enhancement, New compositions	Sangwan et al. 2000; Farooqi et al. 2000

Dracephalummoldavica	Moldavian dragonhead	Lamiaceae	Angiosperms	Tetraploid	Essential oil	27.5% enhancements	Madani et al. 2021; Rauf et al. 2021
Galium mollugo	Hedge bedstraw	Rubiaceae	Angiosperm	Autopolyploid	Flavinoids, phenolic acid	–	TATE et al. 2005; Csepregi et al. 2016
Gossypium hirsutum	Cotton	Malvaceae	Angiosperm	Polyploid	Terpenoid aldehyde	–	Tate et al. 2005; Iannicelli et al. 2020; Park et al. 2019
Glycyrrhizia glabra	Licorice	Fabaceae	Angiosperm	Polyploid (tetraploid)	Glycyrrhizic acid, anthocyanins	Upto 12% enhancements	Iannicelli et al. 2020; Moghbel et al. 2015
Hyoscyamus muticus	Egyptian henbane	Solanaceae	Angiosperm	Autotetraploid	Scopolamine, tropane alkaloid,	200% enhancements, 36% enhancements	Madani et al. 2021; Dehghan et al. 2012; Lavania 2005; Iannicelli et al. 2020
Mentha arvensis	Field corn	Lamiaceae	Angiosperm	Autotetraploid	Essential oil	30% enhancements	Madani et al. 2021
Nicotiana tabacum	Tobacco	Solanaceae	Angiosperm	Polyploid	Pyridine alkaloids	–	Tate et al. 2005; Ghasemi et al. 2021
Oenothera lamarckiana	Evening primrose	Onagraceae	Angiosperm	Tetraploid	Ellagitannin	–	TATE et al. 2005; Greiner and Köhl 2014
Panax ginseng	Ginseng	Araliaceae	Angiosperm	Polyploid(tetraploid, octaploid)	Ginseosides (triterpenoid saponin)	22% enhancements	Iannicelli et al. 2020; Kim et al. 2004
Papaver somniferum	Opium poppy	Papaveraceae	Angiosperm	Polyploid (triploid, tetraploid)	Morphine	Upto 50% enhancements	Madani et al. 2021; Iannicelli et al. 2020; Lavania 2005
Salvia miltiorrhiza	Danshen	Lmaamiaceae	Angiosperm	Tetaploid	Tanshinone ⍰, tanshinone⍰⍰ A, Cryptotanshinone	33% enhancements; 25% enhancements; 162% enhancements	Madani et al. 2021; Iannicelli et al. 2020

(continued)

Table 12.2 (continued)

Plant Name	Common name	Family	Classification/ Clade	Ploidy	Name of Molecules	Molecule conc.	Reference
Sequoia sempervirens	Coastal redwood	Taxodiaceae	Gymnosperm	Hexaploid	Essential oil (Terpine-4-ol, Eugenol & γ-Terpine)	–	Tate et al. 2005; Youssef et al. 2021
Solanum tuberosum	Potato	Solanaceae	Angiosperm	Tetraploid (allotetraploid, autotetraploid)	Sesquiterpenes	22 folds enhancements	Gantait and Mukherjee 2021; Trojak-Goluch et al. 2021; Tavan et al. 2022; Iannicelli et al. 2020
Tanacetum parthenium	Feverfew	Asteraceae	Angiosperm	Tetraploid	Essential oil	32% enhancements	Madani et al. 2021; Ghasemi et al. 2021
Tolmiea menziesii	Piggyback palnt	Saxifragaceae	Angiosperm	Autotetraploid	–	–	Tate et al. 2005; Nezhadi et al. 2021
Triticum aestivum	Wheat	Poaceae	Angiosperm	Polyploid (Hexaploid)	Lutin	10.8 fold enhancements	TATE et al. 2005; Van de Peer et al. 2021; Iannicelli et al. 2020; Cuttriss et al. 2011
Vaccinium uliginosum	Western blueberry	Ericaceae	Angiosperm	Autopolyploid (tetraploid, hexaploid)	Flavonoids (anthocyanin), phenolic acid	2–17 fold variations	Tate et al. 2005; Lyrene2021; Mengist et al. 2020
Zingiber officinale	Ginger	Zingiberaceae	Angiosperm	Tetraploid	Gingerols, Carotenoids	No change, 1.375 folds enhancements	Iannicelli et al. 2020; Gantait and Mukherjee 2021

2017b, 2020). Although both species are very well studied for their ethnopharmacology and a number of phytomolecules have been isolated and identified from the species. Most of the pharmacologically active compounds belong to the terpenoid category of secondary metabolites and collectively this group of compound is termed withanolides.

Withania somnifera, Ashwagandha, the popular Indian ginseng or gooseberry, is a perennial, evergreen small shrub that is distributed globally in wide geographical regions. Although the whole plant possesses medicinal properties, the roots are preferably used for extraction of "Rasayana" (Sangwan et al. 2007). Many pharmacological activities have been attributed due to this Rasayana like adaptogenic, antisedative, anticonvulsion, anti-inflammatory, immunomodulatory activities (Mukherjee et al. 2021). Interestingly, it is effective against several brain and nervous system-related disorders like Parkinson's disease, Alzheimer's disease, and Huntington's disease (Dar and Ahmad 2020). The neuroprotective activity of *W. somnifera* is mediated through anti- oxidative stress mechanism. Withanolide A, withanolide D, β-sitosterol and stigmasterol are the major metabolites against Alzheimer's diseases (Hannan et al. 2020). There is transcriptomic data of *W. somnifera* available, which also reflects the tissue-specific accumulation of different withanolides (Gupta et al. 2013; Sangwan et al. 2014). There is a repository of transcription factors which are involved in regulation of secondary metabolites metabolism (Tripathi et al. 2017). There is variation in ploidy level of *W. somnifera*. Plants may have diploid ($2n = 24$), tetraploid ($2n = 48$), and hexaploid ($2n = 72$) levels, although the predominating ploidy is diploid ($2n = 24$). *W. somnifera* shows significant variation in morphogenetic characters. By using the EST-SSR markers, genetic diversity in 36 genotypes of *W. somnifera* is accessed (Parita et al. 2018). Application of ISSR marker showed genetic diversity in *W. somnifera* species collected from diverse geographical locations (Hiremath et al. 2021). Sangwan and his coworkers have carried out several experiments to explore more and more about the genetic and chemotypic diversity of *W. somnifera* plants collected from different locations in India (Sangwan et al. 2004). Different types of chemotypes (NMITLI-101, 110, 133, 104, 128, 141, 108, 109, 118, 135, and 144) have been identified (Tuli and Sangwan, 2009). The phytochemical analysis includes polypeptide polymorphism, isoenzymes, along with withanolide profiles. RAPD analysis reveals that they cluster together on the basis of genetic similarity. In another study, thirty cultivars are selected from eight different states and cultivated for approaching their stability and adaptability. Out of thirty, cultivars W 20, W 1 (cv. Pratap), W 2, W 3 (cv. Chetak), W 4 and W 6 were found to be highly stable for the root yield (Lal 2015). Five different varieties of *W. somnifera* (Jawahar, Nimithli, Chetak, Pratap, Poshita and) were examined for the withanolide content. There is a significant variation in all the varieties, with the highest content of alkaloids (withaferin A and withanolide A) found in Poshita, followed by Jawahar-20 (Singh et al. 2018; Mishra et al. 2020). In another study performed in Sri Lanka, withaferin A content was estimated in two varieties of *W. somnifera*, LC1 and FR1, these varieties were grown under the same climatic conditions. Different tissues (leaf, bark stem and root) of both varieties are screened for alkaloid content and results showed highest

content of withaferin A in leaves of LC1, with minimum content found in the root of FR1 (Siriwardane et al. 2013). Srivastava et al. (2018) reported genetic variability in 53 genotypes of *W. somnifera* collected from different geographical locations. According to pathway coefficient analysis, there is positive correlation in fresh root weight, total alkaloid (%) in leaf and deoxywithastramonolide in the root (%). Effect of genotype-environment interaction on withanolide content was studied on 16 genotypes of *W. somnifera*. A total of 12 characters were considered along with alkaloid content and a variation was observed in characters of all the genotypes grown under different locations (Kumar et al. 2020). Mirjalili et al. (2009) reported the genetic (by using the RAPD anlysis) and withaferin A diversity in Iranian natural populations of *W. somnifera* and *W. coagulans*.

12.2.2 Artemisia (Asteraceae)

Artemisia is a rich genus of aromatic plants with around 500 species including both herbs and shrubs (Valles and Arthur 2001). Several *Artemisia* species are perennial. However, few *Artemisia* species are annual or biennial (Vallès et al. 2003) because of its greater population and morphological complexities, the taxonomy of genus *Artemisia* is not clear (Hayat et al. 2009). Based on recent molecular studies, the genus *Artemisia* has been divided into five deferent subgenera. These subgenera are *Absinthium, Artemisia, Dracunculus, Tridentata,* and *Pacifca* (Vallès et al. 2011). The diversity of *Artemisia* is mainly found in Asia, Europe, and North America. China (around 150 accession) and Europe (174 accession) are main regions of *Artemisia* diversity. Japan (50 species), Pakistan (38 species), and Iran (35 species) also add diversity to the genus (Hayat et al. 2009). Only few *Artemisia* species (*Artemisia* annua, *Artemisia Absinthium*, and *Artemisia* vulgaris) are commonly utilized as traditional medicine in Asia and Europe from ancient time.

Artemisia annua (sweet wormwood or qinghao in China) is the most popular among *Artemisia species* due to its antimalarial properties from ancient times (Hong et al. 2015). *Artemisia annua* is native to Asian, North American, and European countries. The antimalarial properties of *Artemisia annua* are due to an important compound, artemisinin. The acceptance of artemisinin as antimalarial drug was gained due to the emergence of chloroquine resistance in the 1950s (Ma et al. 2020). Artemisinin, the sesquiterpenoid-based therapy has so far proved efficient in the treatment of millions of people in developing countries (Ma et al. 2020). The Chinese scientist Youyou Tu was honored with the 2015 Nobel Prize in Physiology or Medicine for her discovery of artemisinin, which became a promising drug (Liu and Liu 2016). Recently, artemisinin-derived compounds such as artesunate and artemether are basically used in medicine preparation for malarial drugs (Tu et al. 1981; Wright 2002).

The main chemical constituents of *Artemisia* species are polyphenols and essential oils, but also coumarins, acetylenes, sesquiterpene-lactones, alcohols,

flavonoids, monoterpenes, and sesquiterpene derivatives. The chemical diversity of constituents attributes *Artemisia* species with different pharmacological bioactivities including antibacterial, antimalarial, anticancer etc. (Padalia et al. 2016; Nigam et al. 2019; Sangwan et al. 2010). Vast research is ongoing with *A. annua* for the development of drugs to treat COVID-19 (Bisht et al. 2021).

Artemisinin, the endoperoxide sesquiterpene-lactone synthesized in the eight-celled trichome glands of *A. annua*, is an active ingredient in the artemisinin combination therapy-based treatment of malaria. The content of artemisinin varies among the varieties of *A. annua* from different origins. Even Indian population of *A. annua* plant exhibited very high level of genetic variation in RAPD analysis (Sangwan et al. 1999). Advances in genetics and metabolic engineering approaches widen the path to *in planta* artemisinin production. Microbial-based metabolic engineering of artemisinic acid (the precursor of artemisinin) had demonstrated some potential, but the further chemical synthesis of artemisinin are costly (Ro et al. 2006). Thus, the agricultural production of artemisinin is the only viable source for ACT. *A. annua* is an undeveloped crop and plant-based production of artemisinin is a challenging task. Development of improved varieties for artemisinin production purposes is the realistic target. Different approaches have been applied to improve *in planta* artemisinin yield.

Several plant breeding-based techniques are used to develop improved variety of *A. annua* in terms of higher artemisinin yield. Conventional breeding approach includes identification of superior parental lines with desired traits and crossing them to develop hybrid lines with improved artemisinin yield (Delabays et al. 2001; Cockram et al. 2012; Townsend et al. 2013). Artemisinin biosynthesis in *A. annua* is controlled by genetic factors (Ferreira et al. 1995b; Delabays et al. 2001, 2002). The vegetative phase of *Artemisia* plant can be maintained under long photoperiods, while flowering can be induced under short days. Flowering time of different varieties varies under field conditions, but can be induced in greenhouse. Delabays et al. (1992) had crossed artemisinin (1.1%, w/w) rich, late-flowering Chinese clone with European plants and developed new variety with 0.64% to 0.95% artemisinin content and dry leaf yields between 14 to 21 t/ha (Delabays et al. 1992). Banyai et al. (2010) have developed the tetraploid cultivar of *A. annua* by doubling the chromosomes number with higher level of artemisinin content. Graham et al. (2010) studied genomics of *A. annua* where all the genes of the pathway are shown in genetic linkage with marker traits. Artemis (1.4% artemisinin) is commercially important variety of *A. annua* developed by cross between two genetically different heterozygous parental genotypes C4 and C1 (Delabays et al. 2001). Graham et al. (2010) have developed genetic linkage and QTL maps for Artemis and validated positive QTL for artemisinin yield. 34,419 SNPs with mean SNP frequency of 1 in 104 base pairs were identified from the five EST databases of Artemis F1 hybrid material (Graham et al. 2010). Further, polymorphism confirmation with 19 AFLP primer combinations revealed 322 polymorphic markers. 49 SSR markers which are segregated in the Artemis F1 population were also identified through *in silico* approach (Graham et al. 2010). The mapping population of the Artemis F1 exhibited variation in plant phenotype, artemisinin content (0.93 to 20.65 mg/mg dry weight), glandular

trichome density (4.89 to 19.11 mm^2), leaf area (508.76 to 4696.08 mm^2) and plant fresh weight (160 to 4440 g). Graham et al. (2010) found strong segregation distortion of the advantageous alleles QTL on C4LG1 in favor of artemisinin yield trait (a product of artemisinin concentration and plant fresh weight). Although, artemisinin is present in *A. annua* plant leaf and stem tissues, but artemisinin yield depends on plant genetic potential (Ferreira et al. 1995a). Heterozygous nature of *A. annua* is also a challenge for plant breeders because developed plant exhibit varying degrees of artemisinin content (Delabays et al. 2001; Graham et al. 2010; Larson et al. 2013).

Besides *A. annua*, several other *Artemisia* species such as *A. absinthium, A. aff. Tangutica, A. apiacea, A. bushriences, A. campestris, A. cina, A. diffusa, A. dracunculus, A. dubia, A. indica, A. japonica, A. lancea, A. moorcroftiana, A. parviflora, A. roxburghiana, A. scoparia, A. sieberi, A. sieversiana, A. spicigeria,* and *A. vulgaris* are reported to have artemisinin (0.05–0.034%) in their aerial parts (Mannan et al. 2010; Singh and Sarin 2010; Rashmi et al. 2014). Nganthoi and Sanatombi (2019) had studied artemisinin contents in four *Artemisia* species (*viz., A. parviflora, A. vulgaris L., A. myriantha, and A. nilagarica*) of Manipur region in India and also established the genetic relationship among the four *Artemisia* species by using RAPD (15), ISSR (11) as well as IRAP (3) markers. Out of total 267, 203 and 58 reproducible fragments, 240, 187 and 51 were polymorphic with high average polymorphism (89.88% for RAPD, 92.5% for ISSR and 87.93% for IRAP) (Nganthoi and Sanatombi 2019). The highest similarity index in RAPD (0.5), ISSR (0.5) as well as IRAP (0.79) markers was found between *A. nilagarica* and *A. myriantha* (both belongs to Sub-genus *Artemisia*) with similar artemisinin content per gram DW in leaf (0.031%–0.044%), young flower (0.042%–0.052%), mature flower (0.031%–0.047%) and stem (0.008%–0.01%) (Nganthoi and Sanatombi 2019). While *A. parviflora* (belongs to the Sub-genus Dracunculus) depicted least similarity index with rest three *Artemisia* species and high level of artemisinin content in leaf (0.078%–0.087%). *A. nilagarica* and *A. myriantha* had depicted low genetic variability and have a closer evolutionary relationship, while *A. parviflora* had depicted higher genetic variability.

12.2.3 Ocimum *(Lamiaceae)*

Ocimum genus belongs to the Lamiaceae family and holds immense importance in the plant word due to its medicinal and aromatic properties. The plant is well adapted to survive under tropical warm and temperate conditions and is an inhabitant of tropical Africa, tropical Asia, and tropical America (Paton et al. 2004). The genus contains approximately 150 species out of which 66 species have been reported online (Chowdhury et al. 2017; Gurav et al. 2021). Several Ocimum species are studied in great details for their pharmaceutical constituents being the major ones *O. tenuiflorum, O. basilicum*, *O. Americanum*, O. africanum, *O. gratissimum*, *O. kilimandscharicum*, and *O. citriodorum* (Gupta et al. 2018; Gurav et al. 2021;

Maurya et al. 2019). In India, nine species, including three exotic species namely *O. adscendens*, *O. basilicum*, O. canum, *O. gratissimum*, *O. kilimandscharicum*, O. tenuiflorum, *O. americanum* L., *O. minimum* L., and *O. africanum* Lour have been reported (Chowdhury et al. 2017). Ocimum is rich in chemical diversity which makes it extremely useful in culinary, fragrance, therapeutic, and the cosmetic industry (Pandey et al. 2016). Ocimum is a repertoire of compounds like terpenoids, alkaloids, phenols, tannins, anthocyanins, lignins, quinones, saponins, flavonoids etc. Essential oils present in Ocimum contain linalool, chavicol, methyl chavicol, eugenol, camphor, 1,8-cineole, α-terpineol 6.5%, β-pinene, camphene, sabinene, cis-ocimene, trans-ocimene, ß-caryophyllene, geraniol, limonene, 10-Heptadecen-8-ynoic acid; Cyclopentane, thymol, etc. (Maurya et al. 2019; Enegide and Charles 2021). These compounds impart hepatoprotective, anti-diabetic, antioxidant, neuroprotective, anti-inflammatory, antiseptic, antidiarrheal, antispasmodic, and antimicrobial properties. They also play a role in the treatment of skin, respiratory tract infections, stomach ache, kidney malfunctions etc. (Prakash and Gupta 2005; Dhama et al. 2021; Enegide and Charles 2021). Basil seeds are a rich source of proteins (11.4–22.5 g/100 g), carbohydrates (40–63.8 g/100 g), dietry fiber (7.1–22.6 g/100 g) along with calcium, magnesium, and phenolic compounds (orientine, vicentine, and rosmarinic acid) (Calderón Bravo et al. 2021). Microencapsulation of essential oil from *O. basilicum* was conducted to develop a packaging material which could control the bacterial growth in packaged food and also increase the shelf life of food by inhibiting change in pH (Amor et al. 2021). Similarly, seed mucilage from *O. basilicum* was used with montmorillonite (MMT) as nanofiller to synthesize bionanocomposite films to be used as packaging material (Rohini et al. 2020). Its use as a seasoning, sauces, salads is well accounted for. These traits of Ocimum makes its essential oil very important in the national and international market and thereby an 8% compound annual growth rate is expected from 2019 to 2023 (Gurav et al. 2021).

A number of different approaches have been used to increase secondary metabolites in terms of concentration and components. With the aim of incorporating desired traits and increase the genetic stock; plant breeding has been practiced for many decades (Ahmar et al. 2020). In *Ocimum basilicum var. glabratum* Benth chemotypes which were morphologically similar but contained different chemical compounds, i.e., eugenol, methylchavicol, and camphor were established. It was assumed that the variation in accumulation of compounds with different origins (eugenol, methylchavicol are phenylpropanoids while camphor is terpenoid) was due to the presence of a multi-allelic gene which expresses differentially (Gupta et al. 2005). Similarly, a eugenol rich and methyl chavicol rich line had also been developed by breeding (Gang et al. 2001). In another study, a cross between basil varieties "Perrie" (rich in eugenol) and "Cardinal" (rich in methyl chavicol) on segregation generated eugenol chemotype (23%–25%), methyl chavicol chemotype (23%–25%, and an intermediate mixture of the two compounds (~50%). The segregation followed 1:2:1 pattern, thereby suggesting involvement of a single bi-allelic gene with incomplete dominance (Dudai et al. 2018). Other than these a large number of chemotypes have been developed by conventional breeding such as

CIM ayu; eugenol rich variety of *O. sanctum*, CIM Angana; high yielding variety of *O. sanctum* (Shyam tulsi); CIM Saumya; methyl chavicol and linalool rich short duration variety of *O. basilicum* (Lai 2003; Lal et al. 2004, 2008). Higher ploidy level is associated with higher essential oil content (Lavania 2005). Attempts were made to induce autotetraploidy by colchicine treatment at different plant stages and at different concentrations. Polyploidy resulted in large stomata, pollen grains and increased chloroplast number (Omidbaigi et al. 2010).

12.2.4 Mentha *Species*

The *Mentha* species (Lamiaceae) plants evolved through natural hybridization and selection, consisting of about 25 to 30 species (Brickell and Zuk 1997). *Mentha* essential oil is important for pharmaceutical, cosmetics, food, confectionery, liquor, and pesticide industries (Sangwan et al. 2000). *Mentha* is a rich source of plant secondary metabolites such as terpenes (Sangwan et al. 2000). Phylogenetic relationships and genetic diversity among *Mentha* species have been widely studied mainly by using molecular markers such as RAPD and amplified fragment length polymorphism (AFLP) primers-based genetic analyses (Gobert et al. 2002; Shinwari et al. 2011; Rodrigues et al. 2013; Virginia et al. 2021). Rodrigues et al. (2013) had studied the morphological, phytochemical, and genetic variation in 12 populations of *Mentha cervina* L. (Portuguese endangered medicinal plant) to access the level of diversity through ISSR markers. One hundred and twenty-one individuals of ISSR amplification gave a total of 175 bands (average: 82.4 fragments per individual) with 97.7% (171 band) of them being polymorphic. Out of twelve, four populations clustered together in morphological parameters as well as molecular studies, but the secondary metabolites, i.e., essential oils basis created different clustering (Rodrigues et al. 2013). Rodrigues et al. (2013) interpreted the observation as essential oils evolve more rapidly than the morphological traits, so the morphological traits were found more correlated with the genetic variation. Shinwari et al. (2011) studied the RAPD profile based similarity and diversity of two mint species (*Mentha royleana and Mentha spicata*) including 15 accessions of each. The polymorphism percentage observed in all the 15 accessions of *M. royleana* was 27.44% while in *M. spicata,* it was 47.2%. A high level of genetic polymorphism was noticeable among population, thereby indicating genetic richness and heterozygosity (Shinwari et al. 2011). RAPD profiles of different mint species/accessions *Mentha arvensis* L., two of *Mentha spicata* L., one each of *Mentha spicata* L. cv. viridis, *Mentha* × *piperita* L., *Mentha* × *piperita* L. cv. citrata and *Mentha* × *gracilis* Sole cv. RAPD markers could clearly identify various Mentha genotypes, accessions as well as hybrids. Fenwick and Ward (2001) identified 17 accessions of *Mentha* (three species) belonging to different geographical origins from the USA by using RAPD-based profiling. Momeni et al. (2006) had studied the genetic variation and taxonomic relationship between 17 accessions out of four *Mentha* species *viz., M. spicata, M. piperita, M. suaveolens,* and *M. longifolia* by RAPD fingerprinting.

In another study, Virginia et al. (2021) had established the genetic variations between ten varieties of *Mentha* (*Mentha longifolia, Mentha aquatica, Mentha x piperita, Mentha arvensis subsp. arvensis and Mentha spicata var. crispa* and five commercial varieties) by using eight arbitrary RAPD primer based profiling and recorded 100% polymorphism. Hundred percent polymorphism was interpreted due to out-breeding and the wide dispersal of seed and pollen grains (Virginia et al. 2021).

12.2.5 Geranium sp.

Geranium (family geraniaceae) is a very popular aromatic crop of commercial importance. Essential oil of geranium is used in a variety of formulations of commercial, pharmaceutical, and agricultural importance (Gallardo et al. 2012; Narnoliya et al. 2019). Transcriptome information of *P. graveolens* proposed the pathways involved in primary and secondary metabolite biosynthesis (Narnoliya et al. 2017, 2018). Geranium essential oil has several terpenes as constituents majorly geraniol, citronellal, linalool, geranyl acetate, limonene, pulegone, citronellyl format, citronellylacetate, menthone isomenthone, 10-epi-γ-eudesmol, etc. (Rana et al. 2002; Jadaun et al. 2017a, 2017b). A large number of species diversity (~ 200 speices) is reported in the *Geranium* genera in reference to morphological features and essential oil composition (Tyagi et al. 2003; Yi et al. 2018). Commercially, four species: *graveolens: odoratissimum: radens, and P. capitatum* were grown for harvesting the essential oil. On the basis of variation in oil yield and oil constituents, different chemotypes of *Pelargonium capitatum* are reported (Demarne et al. 1993). Production of somaclonal variations may be a good source of induction of the genetic variability in plants. Kulkarni et al. (1997) reported about the intra clonal variations in *Pelargonium* sp. as they differ significantly in essential oil composition (isomenthone is major constituent instead of geraniol and linalool). Somaclonal variations were studied in an Indian cultivar, "Bourbon" of rose-scented geranium (*Pelargonium graveolens*), they differ in the phenotype (height, leaf shape, leaf size, leaf dentation) and phytochemical composition (Ravindra et al. 2004). Glass house grown somaclones, highly dentated leaves (HDL) and less dentated leaves (LDL) show variability in leaf dentation pattern, herb yield as well as variability in the oil yield also (Saxena et al. 2008; Tyagi et al. 2003).

For identification of genetic diversity in *Geranium* species different types of DNA markers are also used. RAPD and ISSR analysis in cutting (diploid) and polyploidy seedlings revealed geraniol, geranyl formate, and linalool concentration was quite higher in polyploid plants (Yi et al. 2018). RAPD analysis was performed to trace the phylogenetic relationship among the following species of Geranium: *G. dissectum* L. (sec. Dissecta); G. persicum Schönb.-Tem., *G. tuberosum* L., G. kotschyi Boiss., *G. platypetalum* Fisch. & C. A. Mey., G. gracile Ledeb. Ex Nordm., *G. ibericum* Cav. (sec. Tuberosa subsect. Mediterranea R. Knuth). *G. stepporum* P.H.Davis (sec. Tuberosa subsect. Tuberosa (Boiss.) Yeo);

G. columbinum L., *G. rotundifolium* L., *G. sylvaticum* L., *G. collinum* Stephan ex Willd, G. pratense (Yin et al. 2021).

12.2.5.1 Molecular Markers in Association to Secondary Metabolism

Molecular markers have been used extensively in characterization of accessions, germplasm as well as mutant lines. Along with the progress that has markedly taken place during past two to three decades in molecular techniques in developing markers such as RAPD, RFLP, ISSR, microsatellites etc., the markers have been associated with various traits of interest. The plants with secondary metabolites have also been studied, and the co-relation with various markers to assist the analysis related to association between secondary metabolites and markers. RAPD markers were quite popular owing to ease in establishment and also not requiring genomic information (Sangwan et al. 1999, Shukla et al. 2005). Medicinally important plant punarnava (*Boerhavia diffusa*) which is used in herbal and medicinal puoposes. The plant grows in wild and majorly two varieties, white flowered and red flowered are in the trade (Shukla et al. 2005). The attempts to develop better varieties through conventional breeding approaches and also mutant generation through mutagens of physical and chemical nature were attempted. The genetic differences were well differentiated by the RAPD marker as well as by isozymic pattern analysis. The performance of RAPD markers in associating the trait was much better as compared to isozymes. Also the mutant which had unique characters, including flower color, exhibited a unique RAPD pattern.

Similarly, in Cymbopogon species, the fragrant members of Poaceae family were examined with RAPD, isozyme and protein polymorphic markers basis. Cymbopogons are aromatic species and differ in their ploidy status as well. *Cymbopogon martinii* is one of the most prominent species preferred by the farmers for cultivation owing to its rose like aroma. The essential oil contains geraniol and geranyl acetate in elite varieties of sofia and motia. The genetic score, gene diversity, heterozygosity, etc., were computed. Such genetic marker analysis could distinguish elite genotypes of Cymbopogon species (Sangwan et al. 2001a, 2007; Sangwan Neelam et al. 2003). Furthermore, information related to the genetic basis of wild and collected germplasm as well as requirement for trait introgression can be envisaged from the diverse growing plant types.

Similarly in another study, important trade type of Cymbopogon species were studied using RAPD marker to distinguish diversity of trade type varieties and species differing substantially in their oil types (Sangwan et al. 2001a, 2003). The study correlated essential oil metabolic diversity with the genetic diversity parameters. Markers also clustered together *Cymbopogon flexuosus*, *C. pendulus* and *C. khasianus* in one cluster, owing to the similarity in their essential oil constituent profile. Diagnostic markers and stand-alone markers were identified and associated with constituents (Sangwan et al. 2001a, 2003). In another study, genetic variation was created through somaclonal variation in Cymbopogon hybrid Jamrosa. Large numbers of growth parameters were impacted including essential oil content (Nayak

et al. 2003). Gross genetic variations in RAPD pattern of parent and somaclones of Jamrosa indicated the extent of variation in several physiological parameters as well secondary metabolites (Nayak et al. 2003). Superior clones with respect to higher terpinol geraniol content were evaluated in field trials for consistency in performance and characterization through RAPD molecular marker (Nayak et al. 2003).

Withania somnifera is one of the traditional resources of medicinal herb. Molecular markers including RAPDs, and SSRs have been used in studying the genetic diversity and characterizing the genotypes in metabolic core groups (Chaurasiya et al. 2009; Sangwan et al. 2017). Such markers provided clues in establishing genetic diversity of collection at intra- and interspecies levels. The levels of secondary metabolites such as wihanolidal diversity in terms of qualitative as well as quantitative variations were also elucidated. In vitro clonal plants were also established for their clonal stability and also evaluation for distinctness of secondary phytochemicals (Sabir et al. 2007). High throughput analytical techniques of metabolomics and a combination of markers together further substantiated large numbers of core collections for metabolites in an integrated manner (Dhar et al. 2006; Chaurasiya et al. 2009).

12.3 Status of Polyploidy and Its Impact on Secondary Metabolites

Polyploidy is a phenomenon of two processes, i.e., endomitosis and endoreduplication. Other processes like fusion of nuclei, ineffective mitosis, or the emergence of multinucleated cells are also involved in creating polyploid cells. Unlike normal mitosis, the endomitosis is a process in which the cell membrane is not destroyed, and mitosis takes place within the nucleus, chromosome numbers double and sister chromatids are likely to separate and return to the interphase mode, except those chromosomal spindles are not created and as a result sister chromatids are not stretched to the side of the cell and polyploidy cells are not created. The endometriosis process mostly occurs in animal cells and is rare in angiosperms (flowering plants). The endoreduplication nucleus may replicate DNA without going through the mitotic process, resulting in 4n, 8n, 16n, and so on (Ghasemi et al. 2021). Diploidy refers to the presence of two complete set of chromosomes in somatic cells, one from each parent, whereas polyploidy refers to the presence of three or more complete sets of chromosomes in somatic cells (Park et al. 2021). Each organism has a specific number of chromosomes that are divided into different groups. Ploidy refers to the number of chromosomal groupings. In this case, the number of chromosomes may alter. Euploidy and aneuploidy are the two types of alterations that occur. Polyploidy is formed by two mechanisms: somatic doubling and the creation of unreduced gametes (2n).

According to their origin, polyploidies can be classified as auto-polyploidies and allopolyploids (Ghasemi et al. 2021). Polyploidy can arise as a result of

autopolyploidy, which describes the occurrence of polyploidy inside a species, or allopolyploidy, which is described as the development of polyploidy as a result of hybridization between two different species (Park et al. 2021). Polyploidy refers to the duplication of the same genome in autopolyploids or the duplication of separate genomes in allopolyploids (Pradhan et al. 2018). The growth of chromosomes in one kind of diploid results in two (or more) homologous chromosomal pairs, resulting in auto-polyploidies. Allopolyploids are the result of chromosomal growth following the hybridization of two distinct species. In general, the terms euploid and aneuploid are used to refer to organisms with different ploidy a status, which is discussed below. In euploidy, the total number of chromosomes is the exact multiple of the base chromosome series. Euploids can be divided into three groups: monoploids, diploids, and polyploids. But most euploids have two sets of chromosomes and are diploid. However, some euploid species have more than two chromosomal classes and are polyploidy (Ghasemi et al. 2021). According to Talebi et al. (2016) autotetraploidy polyploidization increases the number of chromosomes, gene dosage, concentration, and activity of several enzymes and amino acids. An increased number of genes elevates phenylalanine and tyrosine levels, which are further required for the biosynthesis of phenylpropanoids. It is worth mentioning that geneticists use the letter x to denote the number of base chromosomes, which are collections of single chromosomes that combine to form a full set. The number of chromosomes in each gamete or ploidy surface is also indicated by the letter n. New and modified cultivars of commercially significant species have been developed in recent decades by inducing artificial polyploidies with mutagenic agents. However, polyploidy is not as simple as genome multiplication, and it generates a wide variety of molecular and physiological modifications (Ghasemi et al. 2021). Polyploidy in angiosperms is stable, implying that this genome status has adaptive value and is positively selected (Iannicelli et al. 2020). The majority of polyploidies (natural and artificial) have distinct characteristics from their ancestor (Ghasemi et al. 2021). Polyploidy can disclose novel properties that are not found in either of the diploid progenitors, or it can outperform the progenitor's characteristics. Prior investigations suggest that polyploidy can alter the qualitative or quantitative features of plant metabolites, which has implications for plant metabolism (Park et al. 2021). In most plant species, artificial polyploidy boosted the vitality of determinate plant portions. It has the potential to improve the production of vegetative organs and plant biomass. Polyploidy can be employed to improve the quality and quantity of essential therapeutic substances in plants, as well as to boost secondary metabolites. In flowering plants, chromosome doubling may alter secondary chemistry. DNA methylation, histone changes, and antisense RNA, as well as silence and up- or down regulation of genes involved in secondary metabolite biosynthesis, are all affected by genome doubling (Talebi et al. 2016). Normally, polyploids exhibit an increase in secondary metabolites or enlargement of different plant sections, however, there have been instances of no effect or a negative effect on the number of secondary metabolites. Polyploids make use of their resources through shorter metabolic pathways. They also have a larger cell size and methylation of cytosine (Pradhan et al. 2018). Some of these features, such as bigger organ size (leaf, flower, etc.), a

wider biomass range, dryness tolerance, disease tolerance, and a variety of flowering times, might help polyploids adapt to new environmental changes (Ghasemi et al. 2021). Polyploidy alters physical and physiological alterations, which affect habitat, geographical distribution, reproductive systems, and breeding systems (Talebi et al. 2016). Furthermore, these different polyploidy features increase the likelihood of them being selected for agricultural uses (Ghasemi et al. 2021). It is mostly observed that polyploidy enhances the agronomic traits and attributes such as size of plant leaf and flowers as well as the increased tolerance owing to doubling of genome sets (Iannicelli et al. 2020). Polyploidy promotes growth in secondary metabolites in medicinal plants, as well as more and better adaptation to the environment, and increases the thickness of petals and the size of flower, among other things, in ornamental plants. In reality, artificial polyploidy allows for rapid genetic improvement of plants, making polyploidization one of the most important technologies in plant breeding (Ghasemi et al. 2021). Cell size enlargement is the most well-known and widely recognized result of polyploidization in plants. In reality, the generations of synthetic polyploids allow for rapid genetic improvement in plants, making polyploidization one of the most essential and common technologies employed in plant breeding (Iannicelli et al. 2020). Ploidy induction is an effective strategy for improving the genetics of many plants (Talebi et al. 2016). These distinctive polyploidy cells have almost double the volume of diploid predecessors and a 1.5-fold increase in cell surface area. Polyploids differ from their diploid status in their morpho-physiological parameters, due to the rise in cell size of enlarged flowers, leaf, and fruit sizes. The phenotypic differences are due to two polyploidization-related difficulties that contribute to the most constant impacts of the "polyploid phenotype": an increase in gene dosage. In addition to the reported increases in secondary metabolite production in autopolyploids, the finding in different MAP's suggests that the "polyploid effect" may influence pathways in diverse ways. It has been hypothesized that changes in ploidy cause changes in the control of metabolite production. As a result, while some metabolites are increased, others are reduced in favor of the former (mono and sesquiterpenoids in *A. annua*, morphine/thebaine and codeine in *Papaver somniferum*, and Scopolamine/hyoscyamine in *Centella asiatica)* (Iannicelli et al. 2020). For example, tetraploids exhibited higher levels of sesquiterpenoids in *Artemisia annua* and scopolamine alkaloid accumulation in *H. muticus* plants, presumably through increasing the expression of pathway genes (Talebi et al. 2016).

12.4 Conclusion

Secondary metabolites are of immense importance for the plants and human beings. Their production in plants is under the influence of environmental and genetic constitutions. Ploidy level of plants affects the secondary metabolite accumulation under tight genetic control. In some plants, induction of polyploidy leads to improvement in metabolite accumulation. Various reports are available showing the genetic

variability in different species or cultivars of various medicinal and aromatic plants. These variations also contribute in alteration of secondary metabolites accumulation pattern. Various tools and techniques like genetic markers and QTL mapping can be utilized for identification of genetic diversity in plants and further phytochemical analysis of these plants confer the impact of genetic variance on the chemistry of associated plant up to significant level. Therefore, often plants differing at genetic level also vary in their metabolites either qualitatively or quantitatively. Such studies are very helpful for identification and characterization of improved varieties. Medicinal plants with improved metabolic content will have impact on global trading.

References

Ahmar S, Gill RA, Jung KH, Faheem A, Qasim MU, Mubeen M, Zhou W (2020) Conventional and molecular techniques from simple breeding to speed breeding in crop plants: recent advances and future outlook. Int J Mol Sci 21(7):2590

Akula R, Ravishankar GA (2011) Influence of abiotic stress signals on secondary metabolites in plants. Plant Signal Behav 6(11):1720–1731

Albani MC, Battey NH, Wilkinson MJ (2004) The development of ISSR-derived SCAR markers around the SEASONAL FLOWERING LOCUS (SFL) in Fragariavesca. Theor Appl Genet 109(3):571–579

Alseekh S, Tohge T, Wendenberg R, Scossa F, Omranian N, Li J, Kleessen S, Giavalisco P, Pleban T, Mueller-Roeber B, Zamir D (2015) Identification and mode of inheritance of quantitative trait loci for secondary metabolite abundance in tomato. Plant Cell 27(3):485–512

Amor G, Sabbah M, Caputo L, Idbella M, De Feo V, Porta R, Fechtali T, Mauriello G (2021) Basil essential oil: composition, antimicrobial properties, and microencapsulation to produce active chitosan films for food packaging. Foods 10(1):121

Banyai W, Sangthong R, Karaket N, Inthima P, Mii M, Supaibulwatana K (2010) Overproduction of artemisinin in tetraploid Artemisia annua L. Plant Biotechnol 27(5):427–433

Bisht D, Kumar D, Kumar D, Dua K, Chellappan DK (2021) Phytochemistry and pharmacological activity of the genus *Artemisia*. Arch Pharm Res:1–36

Brickell C, Zuk JD (1997) The American Horticultural Society. AZ Encyclopaedia of Garden Plants DK Publishing Inc., New York, pp 14–18

Calderón Bravo H, Vera Céspedes N, Zura-Bravo L, Muñoz LA (2021) Basil seeds as a novel food, source of nutrients and functional ingredients with beneficial properties: a review. Foods 10:1467. https://doi.org/10.3390/foods10071467

Carles M, Cheung MKL, Moganti S, Dong TT, Tsim KW, Ip NY, Sucher NJ (2005) A DNA microarray for the authentication of toxic traditional Chinese medicinal plants. Planta Med 71(06):580–584

Chaurasiya ND, Sangwan RS, Misra LN, Tuli R, Sangwan NS (2009) Metabolic clustering of a core collection of Indian ginseng *Withania somnifera* Dunal through DNA, isoenzyme, polypeptide and withanolide profile diversity. Fitoterapia 80(8):496–505

Chowdhury T, Mandal A, Roy SC, De Sarker D (2017) Diversity of the genus *Ocimum* (Lamiaceae) through morpho-molecular (RAPD) and chemical (GC–MS) analysis. J Genet Engi Biotechnol 15(1):275–286

Cockram J, Hill C, Burns C, Arroo RR, Woolley JG, Flockart I, Robinson T, Atkinson CJ, Davies MJ, Dungey N, Greenland AJ (2012) Screening a diverse collection of *Artemisia* annua germplasm accessions for the antimalarial compound, artemisinin. Plant Genet Resour 10(2): 152–154

Csepregi R, Bencsik T, Papp N (2016) Examination of secondary metabolites and antioxidant capacity of Anthyllis vulneraria, Fuchsia sp., Galium mollugo and Veronica beccabunga. Acta Biol Hung 67(4):442–446

Cuttriss AJ, Cazzonelli CI, Wurtzel ET, Pogson BJ (2011) University of Hawai 'i at Manoa, Honolulu, HI, USA {Department of Biological Sciences, Lehman College, The City University of New York, Bronx, New York, USA {ARC Centre of Excellence in Plant Energy Biology, Research School of Biology, Australian National University

Dar MJ, Ahmad M (2020) Neurodegenerative diseases and *Withania somnifera* (L.): an update. J Ethnopharmacol 256:112769. S0378874119339303 112769. https://doi.org/10.1016/j.jep.2020.112769

Dehghan E, Häkkinen ST, Oksman-Caldentey KM, Ahmadi FS (2012) Production of tropane alkaloids in diploid and tetraploid plants and in vitro hairy root cultures of Egyptian henbane (Hyoscyamus muticus L.). Plant Cell Tissue Organ Culture (PCTOC) 110(1):35–44

Delabays N, Collet G, Benakis A (1992, July) Selection and breeding for high artemisinin (Qinghaosu) yielding strains of *Artemisia* annua. In WOCMAP I-medicinal and aromatic plants conference: part 4 of 4 330 (pp. 203–208)

Delabays N, Simonnet X, Gaudin MDAD (2001) The genetics of artemisinin content in *Artemisia* annua L. and the breeding of high yielding cultivars. Curr Med Chem 8(15):1795–1801

Delabays NICOLAS, Darbellay CHARLY, Galland NICOLE (2002) Variation and heritability of artemisinin content in *Artemisia* annua L. Taylor and Francis, London, pp 197–209

Demarne FE, Viljoen AM, Van der Walt JJA (1993) A study of the variation in the essential oil and morphology of *Pelargonium* capitatum (L.) L'Hérit. (Geraniaceae). Part I. The composition of the oil. J Essent Oil Res 5(5):493–499

Dhar RS, Verma V, Suri KA, Sangwan RS, Satti NK, Kumar A, Tuli R, Qazi GN (2006) Phytochemical and genetic analysis in selected chemotypes of *Withania somnifera*. Phytochemistry 67(20):2269–2276

Dhama K, Sharun K, Gugjoo MB, Tiwari R, Alagawany M, Iqbal Yatoo M, Thakur P, Iqbal HMN, Chaicumpa W, Michalak I, Elnesr SS, Farag MR (2021) A comprehensive review on chemical profile and pharmacological activities of Ocimum basilicum. Food Rev Int. https://doi.org/10.1080/87559129.2021.1900230

Dudai N, Li G, Shachter A, Belanger F, Chaimovitsh D (2018) Heredity of phenylpropenes in sweet basil (*Ocimum basilicum* L.) chemotypes and their distribution within an F2 population. Plant Breed 137(3):443–449

Enegide C, Charles CO (2021) Ocimum species: ethnomedicinal uses phytochemistry and pharmacological importance. Int J Curr Res Physiol Pharmacol (IJCRPP):1–12. https://doi.org/10.31878/ijcrpp.2021.52.01

Erb M, Kliebenstein DJ (2020) Plant secondary metabolites as defenses, regulators, and primary metabolites: the blurred functional trichotomy. Plant Physiol 184(1):39–52

Fang J, Reichelt M, Hidalgo W, Agnolet S, Schneider B (2012) Tissue-specific distribution of secondary metabolites in rapeseed (Brassica napus L.). PLoS One 7(10):e48006

Farooqi AHA, Luthra R, Fatima S, Bansal RP, Sangwan NS, Sangwan RS (2000) Physiological, biochemical and environmental aspects of essential oil biosynthesis. Cymbopogon-The Aromatic Grass. Lucknow: Central Institute of Medicinal and Aromatic Plants: 199–222

Fenwick AL, Ward SM (2001) Use of random amplified polymorphic DNA markers for cultivar identification in mint. Hort Sci 36(4):761–764

Ferreira JF, Simon JE, Janick J (1995a) Developmental studies of *Artemisia* annua: flowering and artemisinin production under greenhouse and field conditions. Planta Med 61(02):167–170

Ferreira JF, Simon JE, Janick J (1995b) Relationship of artemisinin content of tissue-cultured, greenhouse-grown, and field-grown plants of *Artemisia* annua1. Planta Med 61(04):351–355

Fierascu RC, Fierascu I, Ortan A, Georgiev MI, Sieniawska E (2020) Innovative approaches for recovery of phytoconstituents from medicinal/aromatic plants and biotechnological production. Molecules 25(2):309

Gallardo A, Picollo MI, González-Audino P, Mougabure-Cueto G (2012) Insecticidal activity of individual and mixed monoterpenoids of geranium essential oil against Pediculus humanus capitis (Phthiraptera: Pediculidae). J Med Entomol 49(2):332–335

Gang DR, Wang J, Dudareva N, Nam KH, Simon JE, Lewinsohn E, Pichersky E (2001) An investigation of the storage and biosynthesis of phenylpropenes in sweet basil. Plant Physiol 125(2):539–555

Gantait S, Mukherjee E (2021) Induced autopolyploidy—a promising approach for enhanced biosynthesis of plant secondary metabolites: an insight. J Genet Eng Biotechnol 19(1):1–13

Ghasemi M, Aelaei M, Akbari Soltankohi F, Minaei Chenar H, Bahrami Rad E (2021) Studies on polyploidy induction for improvement of quality traits in ornamental and medicinal plants. Central Asian J Plant Sci Innov 1(2):76–90

Gobert V, Moja S, Colson M, Taberlet P (2002) Hybridization in the section Mentha (Lamiaceae) inferred from AFLP markers. Am J Bot 89(12):2017–2023

Graham LA, Besser K, Susan B, Branigan CA, Czechowski T, Elias L, Guterman I, David H, Peter GI, Khan AM, Larson TR, Li Y, Pawson T, Penfield T, Rae AM, Rathbone DA, Reid S, Ross J, Smallwood MF, Segura V, Townsend T, Vyas D, Winzer T, Bowles D (2010) The genetic map of *Artemisia annua* L. identifies loci affecting yield of the antimalarial drug artemisinin. Science 327(5963):328–331. https://doi.org/10.1126/science.1182612

Greiner S, Köhl K (2014) Growing evening primroses (Oenothera). Front Plant Sci 5:38

Gupta MM, Singh DV, Tripathi AK, Pandey R, Verma RK, Singh S, Shasany AK, Khanuja SPS (2005) Simultaneous determination of Vincristine Vinblastine Catharanthine and Vindoline in Leaves of Catharanthus roseus by high-performance liquid chromatography. J Chromatogr Sci 43(9):450–453. https://doi.org/10.1093/chromsci/43.9.450

Gupta S, Pandey-Rai S, Srivastava S, Naithani SC, Prasad M, Kumar S (2007) Construction of genetic linkage map of the medicinal and ornamental plant *Catharanthus roseus*. J Genet 86(3): 259–268

Gupta P, Goel R, Pathak S, Srivastava A, Singh SP, Sangwan RS, Asif MH, Trivedi K (2013) De novo assembly, functional annotation and comparative analysis of *Withania* somnifera leaf and root transcriptomes to identify putative genes involved in the withanolides biosynthesis. PLoS One 8(5):e62714

Gupta S, Srivastava A, Shasany AK, Gupta AK (2018) Genetics, cytogenetics, and genetic diversity in the genus Ocimum. In: The Ocimum genome. Springer, Cham, pp 73–87

Gurav TP, Dholakia BB, Giri AP (2021) A glance at the chemodiversity of Ocimum species: trends, implications, and strategies for the quality and yield improvement of essential oil. Phytochem Rev:1–35

Hannan MA, Dash R, Haque M, Choi SM, Moon IS (2020) Integrated system pharmacology and in silico analysis elucidating neuropharmacological actions of *Withania somnifera* in the treatment of Alzheimer's disease. CNS Neurol Disord Drug Targets (Formerly Curr Drug Targets-CNS Neurol Disord) 19(7):541–556

Hayat MQ, Khan MA, Ashraf M, Jabeen S (2009) Ethnobotany of the genus *Artemisia* L. (Asteraceae) in Pakistan. Ethnobot Res Appl 7:147–162

Hiremath C, Philip R, Sundaresan V (2021) Molecular characterization of India Ginseng *Withania somnifera* (L) using ISSR markers. Mol Biol Rep: 1–7

Hong, L., Guo, Z., Huang, K., Wei, S., Liu, B., Meng, S. and Long, C., 2015. Ethnobotanical study on medicinal plants used by Maonan people in China. J Ethnobiol Ethnomed, 11(1):1–35

Hu Y, Zhang Q, Xin H, Qin LP, Lu BR, Rahman K, Zheng H (2007) Association between chemical and genetic variation of *Vitexrotundifolia* populations from different locations in China: its implication for quality control of medicinal plants. Biomed Chromatogr 21(9):967–975

Iannicelli J, Guariniello J, Tossi VE, Regalado JJ, Di Ciaccio L, Van Baren CM, Álvarez SP, Escandon AS (2020) The "polyploid effect" in the breeding of aromatic and medicinal species. Sci Hortic 260:108854

Jadaun JS, Sangwan NS, Narnoliya LK, Tripathi S, Sangwan RS (2017a) *Withania* coagulans tryptophan decarboxylase gene cloning, heterologous expression, and catalytic characteristics of the recombinant enzyme. Protoplasma 254(1):181–192

Jadaun JS, Sangwan NS, Narnoliya LK, Singh N, Bansal S, Mishra B, Sangwan RS (2017b) Over-expression of DXS gene enhances terpenoidal secondary metabolite accumulation in rose-scented geranium and *Withania* somnifera: active involvement of plastid isoprenogenic pathway in their biosynthesis. Physiol Plant 159(4):381–400

Jadaun JS, Kushwaha AK, Sangwan NS, Narnoliya LK, Mishra S, Sangwan RS (2020) WRKY1-mediated regulation of tryptophan decarboxylase in tryptamine generation for withanamide production in *Withania* somnifera (Ashwagandha). Plant Cell Rep 39(11):1443–1465

Kennedy DO, Bonnländer B, Lang SC, Pischel I, Forster J, Khan J et al (2020) Acute and chronic effects of green oat (Avena sativa) extract on cognitive function and mood during a laboratory stressor in healthy adults: a randomised, double-blind, placebo-controlled study in healthy humans. Nutrients 12(6):1598

Khan S, Shah RA (2016) Assessment of genetic diversity among India Ginseng, *Withania somnifera* (L) Dunal using RAPD and ISSR markers. *Research*. Biotechnology 7

Kim YS, Hahn EJ, Murthy HN, Paek KY (2004) Effect of polyploidy induction on biomass and ginsenoside accumulations in adventitious roots of ginseng. J Plant Biol 47(4):356–360

Kulkarni RN, Baskaran K, Ramesh S, Kumar S (1997) Intra-clonal variation for essential oil content and composition in plants derived from leaf cuttings of rose-scented geranium (Pelargonium sp.). Ind Crop Prod 6(2):107–112

Kumar A, Mishra P, Rodrigues V, Baskaran K, Verma RS, Padalia RC, Sundaresan V (2019) Delineation of *Ocimum gratissimum* L. complex combining morphological, molecular and essential oils analysis. Ind Crop Prod 139:111536

Kumar M, Patel M, Chauhan R, Tank C, Solanki S, Patel P, Bhadauria H, Gami R, Pachchigar K, Soni N, Patel P (2020) Elucidation of genotype–environment interactions and genetic stability parameters for yield, quality and agromorphological traits in ashwagandha (*Withania somnifera* (L.) Dunal). J Genet 99(1):1–16

Kushwaha AK, Sangwan NS, Trivedi K, Negi AS, Misra L, Sangwan RS (2013a) Tropine forming tropinone reductase gene from *Withania* somnifera (Ashwagandha): biochemical characteristics of the recombinant enzyme and novel physiological overtones of tissue-wide gene expression patterns. PLoS One 8(9):e74777

Kushwaha AK, Sangwan NS, Tripathi S, Sangwan RS (2013b) Molecular cloning and catalytic characterization of a recombinant tropine biosynthetic tropinone reductase from *Withania* coagulans leaf. Gene 516(2):238–247

Labra M, Miele M, Ledda B, Grassi F, Mazzei M, Sala F (2004) Morphological characterization, essential oil composition and DNA genotyping of *Ocimum basilicum* L. cultivars. Plant Sci 167(4):725–731

Lai R (2003) High yielding eugenol rich oil producing variety of *Ocimum sanctum* CIM-Ayu. J Med Arom Plant Sci 25:746–747

Lal RK (2015) Genotype selection for agronomical trait–seed yield in kewachh {*Mucuna pruriens* (L.)}. Ind Crop Prod 65:62–70

Lal RK, Khanuja SPS, Agnihotri AK, Shasany AK, Naqvi AA, Dwivedi S, Mishra HO, Dhawan OP, Kalra A, Singh A, Bahl JR, Singh S, Patra DD, Agarwal S, Darokar MP, Gupta AK, Gupta ML, Chandra R (2004) An early, short duration, high essential oil, methyl chavicol and linalool yielding variety of Indian basil (*Ocimum basilicum*)'CIM-Saumya'. J Med Arom Plant Sci 26:77–78

Lal RK, Khanuja SPS, Rizavi H, Shasany AK, Ahmad R, Chandra R, Naqvi AA, Misra HO, Singh A, Singh N, Lohia RS, Bansal K, Darokar MP, Gupta AK, Kalara A, Dhawan OP, Bahl JR, Singh AK, Shankar H, Kumar D, Alam M (2008) Registration of a high yielding dark purple pigmented, variety 'CIM-Angana'of Shyam tulsi (*Ocimum sanctum* L.). J Med Aromat Plant Sci 30:92–94

Larson TR, Branigan C, Harvey D, Penfield T, Bowles D, Graham IA (2013) A survey of artemisinic and dihydroartemisinic acid contents in glasshouse and global field-grown populations of the artemisinin-producingplant *Artemisia* annua L. Ind Crop Prod 45:1–6. https://doi.org/10.1016/j.indcrop.2012.12.004

Lavania UC (2005) Genomic and ploidy manipulation for enhanced production of phyto-pharmaceuticals. Plant Genet Resour 3(2):170–177

Li Y, Ding WL (2010) Genetic diversity assessment of Trollius accessions in China by RAPD markers. Biochem Genet 48:34–43

Liu W, Liu Y (2016) Youyou Tu: significance of winning the 2015 Nobel Prize in Physiology or Medicine. Cardiovasc Diagn Therapy 6(1):1

Ma N, Zhang Z, Liao F, Jiang T, Tu Y (2020) The birth of artemisinin. Pharmacol Ther 107658

Madani H, Escrich A, Hosseini B, Sanchez-Muñoz R, Khojasteh A, Palazon J (2021) Effect of polyploidy induction on natural metabolite production in medicinal plants. Biomol Ther 11(6): 899

Mahajan V, Rather IA, Awasthi P, Anand R, Gairola S, Meena SR, Bedi YS, Gandhi SG (2015) Development of chemical and EST-SSR markers for Ocimum genus. Ind Crop Prod 63:65–70

Makmur K, Chikmawati T, Sobir S (2020) Genetic variability of lemon basil (Ocimum× africanumLour.) from Indonesian archipelago based on morphological and inter-simple sequence repeats markers. Biodivers J Biol Divers 21(12)

Mannan A, Ahmad I, Ashrad W et al (2010) Survey of artemisinin production by diverse *Artemisia* species in northern Pakistan. Malar J 9:310

Manokari M, Badhepuri MK, Cokulraj M, Sandhya D, Dey A, Kumar V, Faisal M, Alatar AA, Singh RK, & Shekhawat MS (2022) Validation of metaTopolin in organogenesis, improved morpho-physio-chemical responses, and clonal fidelity analysis in *Dioscoreapentaphylla* L.–an underutilized yam species. S Afr J Bot

Marieschi M, Torelli A, Poli F, Sacchetti G, Bruni R (2009) RAPD-based method for the quality control of Mediterranean oregano and its contribution to pharmacognostic techniques. J Agric Food Chem 57(5):1835–1840

Mauro R, Portis E, Acquadro A, Lombardo S, Mauromicale G, Lanteri S (2009) Genetic diversity of globe artichoke landraces from Sicilian small-holdings: implications for evolution and domestication of the species. Conserv Genet 10(2):431–440

Maurya S, Sangwan NS (2020) Profiling of essential oil constituents in Ocimum species. Proc Natl Acad Sci India Sect B Biol Sci 90(3):577–583

Maurya S, Chandra M, Yadav RK, Narnoliya LK, Sangwan RS, Bansal S, Sandhu P, Singh U, Kumar D, Sangwan NS (2019) Interspecies comparative features of trichomes in Ocimum reveal insights for biosynthesis of specialized essential oil metabolites. Protoplasma 256(4): 893–907

Mengist MF, Grace MH, Xiong J, Kay CD, Bassil N, Hummer K et al (2020) Diversity in metabolites and fruit quality traits in blueberry enables ploidy and species differentiation and establishes a strategy for future genetic studies. Front Plant Sci 11:370

Mirjalili MH, Fakhr-Tabatabaei SM, Alizadeh H, Ghassempour A, Mirzajani F (2009) Genetic and withaferin A analysis of Iranian natural populations of *Withania* somnifera and W. coagulans by RAPD and HPTLC. Nat Prod Commun 4(3):1934578X0900400307

Mishra S, Sangwan RS, Bansal S, Sangwan NS (2013) Efficient genetic transformation of *Withania* coagulans (Stocks) Dunal mediated by Agrobacterium tumefaciens from leaf explants of in vitro multiple shoot culture. Protoplasma 250(2):451–458

Mishra S, Bansal S, Sangwan RS, Sangwan NS (2016) Genotype independent and efficient Agrobacterium-mediated genetic transformation of the medicinal plant *Withania* somnifera Dunal. J Plant Biochem Biotechnol 25(2):191–198

Mishra B, Bose SK, Sangwan NS (2020) Comparative investigation of therapeutic plant *Withania* somnifera for yield, productivity, withanolide content, and expression of pathway genes during contrasting seasons. Ind Crop Prod 154:112508

Moghbel N, Borujeni MK, Bernard F (2015) Colchicine effect on the DNA content and stomata size of Glycyrrhiza glabra var. glandulifera and Carthamus tinctorius L. cultured in vitro. J Genet Eng Biotechnol 13(1):1–6

Momeni S, Shiran B, Razmjoo K (2006) Genetic variation in Iranian mints on the bases of RAPD analysis. Pak J Biol Sci 9(10):1898–1904

Mukherjee PK, Banerjee S, Biswas S, Das B, Kar A, Katiyar CK (2021) *Withania somnifera* (L.) Dunal-Modern perspectives of an ancient Rasayana from Ayurveda. J Ethnopharmacol 264:113157

Narnoliya LK, Kaushal G, Singh SP, Sangwan RS (2017) De novo transcriptome analysis of rose-scented geranium provides insights into the metabolic specificity of terpene and tartaric acid biosynthesis. BMC Genomics 18(1):1–14

Narnoliya LK, Sangwan RS, Singh SP (2018) Transcriptome mining and in silico structural and functional analysis of ascorbic acid and tartaric acid biosynthesis pathway enzymes in rose-scanted geranium. Mol Biol Rep 45(3):315–326

Narnoliya LK, Jadaun JS, Singh SP (2019) The phytochemical composition, biological effects and biotechnological approaches to the production of high-value essential oil from geranium. In: Essential oil research. Springer, Cham, pp 327–352

Narnoliya LK, Sangwan N, Jadaun JS, Bansal S, Sangwan RS (2021) Defining the role of a caffeic acid 3-O-methyltransferase from Azadirachta indica fruits in the biosynthesis of ferulic acid through heterologous over-expression in Ocimum species and *Withania* somnifera. Planta 253(1):1–13

Nayak S, Srivastava G, Debata BK, Sangwan NS (2003) Evaluation of agronomically useful somaclonal variants in Jamrosa (a hybrid Cymbopogon) and detection of genetic changes through RAPD. Plant Sci 164:1029–1035

Nayak SA, Kumar S, Satapathy K, Moharana A, Behera B, Barik DP, Acharya L, Mohapatra PK, Jena PK, Naik SK (2013) In vitro plant regeneration from cotyledonary nodes of *Withania somnifera* (L.) Dunal and assessment of clonal fidelity using RAPD and ISSR markers. Actaphysiologiaeplantarum 35(1):195–203

Negi MS, Singh A, Lakshmikumaran M (2000) Genetic variation and relationship among and within *Withania* species as revealed by AFLP markers. Genome 43(6):975–980

Nezhadi F, Safari H, Karami E, Rahimi AR (2021) A cytogenetic, morphological, and ecological comparison of seven different species of Achillea. ssp. Accessions in Kurdistan Province, Iran.

Nganthoi M, Sanatombi K (2019) Artemisinin content and DNA profiling of *Artemisia* species of Manipur. S Afr J Bot 125:9–15

Nigam M, Atanassova M, Mishra AP, Pezzani R, Devkota HP, Plygun S, Salehi B, Setzer WN, Sharifi-Rad J (2019) Bioactive compounds and health benefits of *Artemisia* species. Nat Prod Commun 14(7):1934578X19850354

Omidbaigi R, Mirzaei M, Moghadam MS (2010) Difference of growth traits, essential oil content and compositions between diploid and iinduced tetraploid plants of basil (*Ocimum basilicum* L.). J Essent Oil Bearing Plants 13(5):579–587

Padalia RC, Verma RS, Chauhan A, Goswami P, Chanotiya CS (2016) Essential oil composition of *Artemisia* stelleriana Besser from India. J Essent Oil Res 28(5):400–405

Padilla-González GF, Frey M, Gómez-Zeledón J, Da Costa FB, Spring O (2019) Metabolomic and gene expression approaches reveal the developmental and environmental regulation of the secondary metabolism of yacón (Smallanthus sonchifolius, Asteraceae). Sci Rep 9(1):1–15

Pandey N, Meena RP, Rai SK, Pandey-Rai S (2016) In vitro generation of high artemisinin yielding salt tolerant somaclonal variant and development of SCAR marker in *Artemisia* annua L. Plant Cell Tissue Organ Culture (PCTOC) 127(2):301–314

Parita B, Kumar SN, Darshan D, Karen P (2018) Elucidation of genetic diversity among ashwagandha [*Withaniasomnifera* (L.) Dunal] genotypes using EST-SSR markers. *Research*. J Biotechnol 13(10):52–59

Park SH, Scheffler J, Scheffler B, Cantrell CL, Pauli CS (2019) Chemical defense responses of upland cotton, Gossypium hirsutum L. to physical wounding. Plant Direct 3(5):e00141

Park CH, Park YE, Yeo HJ, Yoon JS, Park SY, Kim JK, Park SU (2021) Comparative analysis of secondary metabolites and metabolic profiling between diploid and tetraploid Morus alba L. J Agric Food Chem 69(4):1300–1307

Paton AJ, Springate D, Suddee S, Otieno D, Grayer RJ, Harley MM, Willis F, Simmonds MS, Powell MP, Savolainen V (2004) Phylogeny and evolution of basils and allies (Ocimeae, Labiatae) based on three plastid DNA regions. Mol Phylogenet Evol 31(1):277–299

Peters MD, Xiang QYJ, Thomas DT, Stucky J, Whiteman NK (2009) Genetic analyses of the federally endangered *Echinacea laevigata* using amplified fragment length polymorphisms (AFLP)—Inferences in population genetic structure and mating system. Conserv Genet 10(1): 1–14

Pradhan SK, Gupta RC, Goel RK (2018) Differential content of secondary metabolites in diploid and tetraploid cytotypes of *Siegesbeckia orientalis* L. Nat Prod Res 32(20):2476–2482

Prakash P, Gupta N (2005) Therapeutic uses of *Ocimum sanctum* Linn (Tulsi) with a note on eugenol and its pharmacological actions: a short review. Indian J Physiol Pharmacol 49(2):125–131

Qari SH, Tarbiyyah I (2021) The genetic regulation of secondary metabolic pathways in response to salinity and drought as abiotic stresses. Appl Sci 11(15):6668

Rana VS, Juyal JP, Blazquez MA (2002) Chemical constituents of essential oil of *Pelargonium* graveolens leaves. Int J Aromather 12(4):216–218

Rashmi TR, Francis MS, Murali S (2014) Determination of artemisinin in selected *Artemisia* L. species by HPLC. Indo Am J Pharm Res 4:2637–2644

Rauf S, Ortiz R, Malinowski DP, Clarindo WR, Kainat W, Shehzad M, Waheed U, Hassan SW (2021) Scope of whole genome duplication in forages: a plant breeder perspective

Ravindra NS, Kulkarni RN, Gayathri MC, Ramesh S (2004) Somaclonal variation for some morphological traits, herb yield, essential oil content and essential oil composition in an Indian cultivar of rose-scented geranium. Plant Breed 123(1):84–86

Ro DK, Paradise EM, Ouellet M, Fisher KJ, Newman KL, Ndungu JM, Ho KA, Eachus RA, Ham TS, Kirby J, Chang MC (2006) Production of the antimalarial drug precursor artemisinic acid in engineered yeast. Nature 440(7086):940–943

Rodrigues L, Póvoa O, van den Berg C, Figueiredo AC, Moldão M, Monteiro A (2013) Genetic diversity in *Mentha cervina* based on morphological traits, essential oils profile and ISSRs markers. Biochem Syst Ecol 51:50–59

Rohini B, Padma Ishwarya S, Rajasekharan R, Kumar A, Kumar V (2020) *Ocimum basilicum* seed mucilage reinforced with montmorillonite for preparation of bionanocomposite film for food packaging applications. Polym Test 87:106465. S0142941820301045 106465. https://doi.org/10.1016/j.polymertesting.2020.106465

Sabir F, Sangwan Neelam S, Chaurasiya ND, Misra LN, Tuli R, Sangwan RS (2007) Micropropagation of *Withania somnifera* L. accessions from axillary meristem for rapid propagation and consistent withanolide productivity. Int J Geogr Inf Syst 13:118–128

Sangwan RS, Sangwan NS (2000) Metabolic analysis of oilchemotypic diversity in Cymbopogons. Cymbopogons: the aromatic grass monograph. CIMAP, Lucknow, pp 223–247

Sangwan NS, Sangwan RS (2014) Secondary metabolites of traditional medical plants: a case study of Ashwagandha (*Withania* somnifera). In: Applied plant cell biology. Springer, Berlin, Heidelberg, pp 325–367

Sangwan RS, Sangwan NS, Jain DC, Kumar S, Ranade SA (1999) RAPD profile based genetic characterization of chemotypic variants of *Artemisia* annua L. IUBMB Life 47(6):935–944

Sangwan RS, Yadav U, Sangwan NS (2000) Isolation of genomic DNA from defatted oil seed residue of opium poppy (Papaver sominiferum). Plant Mol Biol Report 18(3):265–270. https://doi.org/10.1007/BF02823997

Sangwan NS, Yadav U, Sangwan RS (2003) Genetic diversity among elite varieties of the aromatic grasses, Cymbopogon martinii. Euphytica 130:117–130

Sangwan N, Yadav U, Sangwan R (2001) Molecular analysis of genetic diversity in elite Indian cultivars of essential oil trade types of aromatic grasses (Cymbopogon species). Plant Cell Rep 20(5):437–444. https://doi.org/10.1007/s002990100324

Sangwan NS, Farooqi AHA, Shabih F, Sangwan RS (2001a) Regulation of essential oil production in plants. Plant Growth Regul 34(1):3–21

Sangwan N, Yadav U, Sangwan R (2001b) Molecular analysis of genetic diversity in elite Indian cultivars of essential oil trade types of aromatic grasses (Cymbopogon species). Plant Cell Rep 20(5):437–444

Sangwan NS, Sharma PK, Sangwan RS (2007) Geranyl acetate esterase is commonly present but linalyl acetate esterase occurrence is highly limited in plants. Flavour Fragr J 22:169–173

Sangwan RS, Chaurasiya ND, Misra LN, Lal P, Uniyal GC, Sharma R, Sangwan NS, Suri KA, Qazi GN, Tuli R (2004) Phytochemical variability in commercial herbal products and preparations of *Withania somnifera* (Ashwagandha). Curr Sci:461–465

Sangwan NS, Kumar R, Srivastava S, Kumar A, Gupta A, Sangwan RS (2010) Recent developments on secondary metabolite biosynthesis in *Artemisia* annua L. J Plant Biol 37:1–24

Sangwan NS, Sabir F, Mishra S, Bansal S, Sangwan RS (2014) Withanolides from *Withania somnifera* Dunal: Development of cellular technology and their production. Recent Pat Biotechnol 8(1):25–35

Sangwan NS, Tripathi S, Srivastava Y, Mishra B, Pandey N (2017) Phytochemical genomics of Ashwagandha. In: Science of Ashwagandha: preventive and therapeutic potentials. Springer, Cham, pp 3–36

Sangwan NS, Jadaun JS, Tripathi S, Mishra B, Narnoliya LK, Sangwan RS (2018) Plant metabolic engineering. In: Omics technologies and bio-engineering. Academic Press, pp 143–175

Sarwat M, Das S, Srivastava S (2008) Analysis of genetic diversity through AFLP, SAMPL, ISSR and RAPD markers in *Tribulusterrestris*, a medicinal herb. Plant Cell Rep 27(3):519–528

Sarwat M, Nabi G, Das S, Srivastava PS (2012) Molecular markers in medicinal plant biotechnology: past and present. Crit Rev Biotechnol 32(1):74–92

Saxena G, Verma PC, Banerjee S, Kumar S (2008) Field performance of somaclones of rose scented geranium (*Pelargonium* graveolens L'Her Ex Ait.) for evaluation of their essential oil yield and composition. Ind Crop Prod 27(1):86–90

Shasany AK, Darokar MP, Dhawan S, Gupta AK, Gupta S, Shukla AK, Patra NK, Khanuja SP (2005) Use of RAPD and AFLP markers to identify inter- and intraspecific hybrids of Mentha. J Hered 96(5):542–549

Shinwari ZK, Sultan S, Mehmood T (2011) Molecular and morphological characterization of selected Mentha species. Pak J Bot 43(3):1433–1436

Shukla N, Sangwan NS, Misra HO, Sangwan RS (2003) Genetic diversity analysis in Boerhavia diffusa L. of different geographic locations in India using RAPD markers. Genet Resour Crop Evol 50(6):587–601

Shukla N, Sangwan NS, Misra HO et al (2005) Characterization of Boerhavia diffusa L mutant lines by RAPD and isozyme selected for agronomically valuable traits. J Genet Breed 58:37–46

Singh A, Sarin R (2010) *Artemisia* scoparia- a new source of artemisinin. Bangl J Pharmacol 5:17–20

Singh J, Sangwan RS, Gupta S, Saxena S, Sangwan NS (2015) Profiling of triterpenoid saponin content variation in different chemotypic accessions of Centella asiatica L. Plant Genet Resour 13(2):176–179

Singh M, Shah P, Punetha H, Agrawal S (2018) Varietal comparison of withanolide contents in different tissues of *Withania* somnifera (L.) Dunal (Ashwagandha). Int J Life Sci Sci Res 4(3): 1752–1758

Siriwardane AS, Dharmadasa RM, Samarasinghe K (2013) Distribution of Withaferin A, an anticancer potential agent, in different parts of two varieties of *Withania somnifera* (L.) Dunal. grown in Sri Lanka. Pak J Biol Sci 16:141–144

Solouki M, Mehdikhani H, Zeinali H, Emamjomeh AA (2008) Study of genetic diversity in Chamomile (*Matricaria chamomilla*) based on morphological traits and molecular markers. Sci Hortic 117(3):281–287

Soltis PS, Marchant DB, Van de Peer Y, Soltis DE (2015) Polyploidy and genome evolution in plants. Curr Opin Genet Dev 35:119–125

Song Z, Li X, Wang H, Wang J (2010) Genetic diversity and population structure of *Salvia miltiorrhiza*Bge in China revealed by ISSR and SRAP. Genetica 138(2):241–249

Srivastava Y, Sangwan NS (2020) Improving medicinal crops through phytochemical perspective: *Withania* somnifera (Ashwagandha). In: Advancement in crop improvement techniques. Woodhead Publishing, pp 275–295

Srivastava S, Sangwan RS, Tripathi S, Mishra B, Narnoliya LK, Misra LN, Sangwan NS (2015) Light and auxin responsive cytochrome P450s from *Withania* somnifera Dunal: cloning, expression and molecular modelling of two pairs of homologue genes with differential regulation. Protoplasma 252(6):1421–1437

Srivastava A, Gupta AK, Shanker K, Gupta MM, Mishra R, Lal RK (2018) Genetic variability, associations, and path analysis of chemical and morphological traits in Indian ginseng [*Withania* somnifera (L.) Dunal] for selection of higher yielding genotypes. J Ginseng Res 42(2):158–164

Street RA, Sidana J, Prinsloo G (2013) Cichorium intybus: Traditional uses, phytochemistry, pharmacology, and toxicology. Evidence-Based Complementary and Alternative Medicine, 2013

Talebi SF, Saharkhiz MJ, Kermani MJ, Sharafi Y, Fard FR (2016) Effect of ploidy level on the nuclear genome content and essential oil composition of Anise Hyssop (*Agastache foeniculum* [Pursh.] Kuntze). Anal Chem Lett 6(5):678–687

Talebi SF, Saharkhiz MJ, Kermani MJ, Sharafi Y, Raouf Fard F (2017) Effect of different antimitotic agents on polyploid induction of anise hyssop (A*gastache foeniculum* L.). Caryologia 70(2):184–193

Tate JA, Soltis DE, Soltis PS (2005) Polyploidy in plants. In: The evolution of the genome. Academic Press, pp 371–426

Tavan M, Azizi A, Sarikhani H, Mirjalili MH, Rigano MM (2022) Induced polyploidy and broad variation in phytochemical traits and altered gene expression in Salvia multicaulis. Sci Hortic 291:110592

Townsend T, Segura V, Chigeza G, Penfield T, Rae A, Harvey D, Bowles D, Graham IA (2013) The use of combining ability analysis to identify elite parents for *Artemisia annua* F1 hybrid production. PLoS One 8(4):e61989

Travadi T, Sharma S, Pandit R, Nakrani M, Joshi C, Joshi M (2021) A duplex PCR assay for authentication of *Ocimum basilicum* L and *Ocimum tenuiflorum* L in Tulsichurna. Food Control:108790

Tripathi N, Saini N, Mehto V, Kumar S, Tiwari S (2012) Assessment of genetic diversity among *Withania* somnifera collected from central India using RAPD and ISSR analysis. Med Aromatic Plant Sci Biotechnol 6(1):33–39

Tripathi S, Sangwan RS, Narnoliya LK, Srivastava Y, Mishra B, Sangwan NS (2017) Transcription factor repertoire in Ashwagandha (*Withania* somnifera) through analytics of transcriptomic resources: insights into regulation of development and withanolide metabolism. Sci Rep 7(1): 1–17

Trojak-Goluch A, Kawka-Lipińska M, Wielgusz K, Praczyk M (2021) Polyploidy in industrial crops: applications and perspectives in plant breeding. Agronomy 11(12):2574

Tu YY, Ni MY, Zhong YR, Li LN, Cui SL, Zhang MQ, Xiuzhen W, Xiaotian L (1981) Studies on the constituents of *Artemisia* annua L. Acta Pharm Sin 16:366–370

Tuli R, Sangwan RS (2009) Ashwagandha (*Withania somnifera*): a model Indian medicinal plant, 1st edn. CSIR, New Delhi

Tyagi BR, Tripathi A, Singh H, Sangwan RS, Sangwan Neelam S, Naqvi AA (2003) Narrowly lobed leaf of rose-scented geranium (Pelargonium species), a chemically induced mutation for leaf shape. J Genet Breed 57:115–124

Valles J, Arthur ED (2001) Artemisia systematics and phylogeny: cytogenetic and molecular insights. In: McArthur ED, Fairbanks DJ. comps. Shrubland ecosystem genetics and biodiversity: proceedings; 2000 June 13–15; Provo, UT. Proc. RMRS-P-21. Ogden, UT: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. pp 67–74

Vallès J, Torrell M, Garnatje T, Garcia-Jacas N, Vilatersana R, Susanna A (2003) The genus *Artemisia* and its allies: phylogeny of the subtribe Artemisiinae (Asteraceae, Anthemideae) based on nucleotide sequences of nuclear ribosomal DNA internal transcribed spacers (ITS). Plant Biol 5(03):274–284

Vallès J, Garcia S, Hidalgo O, Martín J, Pellicer J, Sanz M, Garnatje T (2011) Biology, genome evolution, biotechnological issues, and research including applied perspectives in Artemisia (Asteraceae). Adv Bot Res 60:349–419. https://doi.org/10.1016/B978-0-12-385851-1.00015-9

Van de Peer Y, Ashman TL, Soltis PS, Soltis DE (2021) Polyploidy: an evolutionary and ecological force in stressful times. Plant Cell 33(1):11–26

Vieira RF, Grayer RJ, Paton A, Simon JE (2001) Genetic diversity of *Ocimum gratissimum* L. based on volatile oil constituents, flavonoids and RAPD markers. Biochem Syst Ecol 29(3):287–304

Vieira RF, Goldsbrough P, Simon JE (2003) Genetic diversity of basil (*Ocimum* spp.) based on RAPD markers. J Am Soc Hortic Sci 128(1):94–99

Virginia BI, Geabot I, Stoie A et al (2021) Use of RAPD markers for identification of different varieties in Mentha sp. Hop Med Plants XXIX:1–2

Volkov RA, Panchuk II, Borisjuk NV, Hosiawa-Baranska M, Maluszynska J, Hemleben V (2017) Evolutional dynamics of 45S and 5S ribosomal DNA in ancient allohexaploid Atropa belladonna. BMC Plant Biol 17(1):1–15

Watthanachaiyingcharoen R, Komatsu K, Zhu S, Vajragupta O, Leelamanit W (2010) Authentication of *Coscinium fenestratum* among the other menispermaceae plants prescribed in Thai folk medicines. Biol Pharm Bull 33(1):91–94

Wright WC (2002) Artemisia. Taylor and Francis, London, pp 1–344

Xie H, Huo KK, Chao Z, Pan SL (2009) Identification of crude drugs from Chinese medicinal plants of the genus *Bupleurum* using ribosomal DNA ITS sequences. Plantamedica 75(01):89–93

Xue XF, Song ZW, Hong XY (2007) Five new species of the genus *Tegolophus Keifer* (Acari: Eriophyidae: Phyllocoptinae: Anthocoptini) from China. Zootaxa 1525(1):19–30

Yadav RK, Sangwan RS, Sabir F, Srivastava AK, Sangwan NS (2014) Effect of prolonged water stress on specialized secondary metabolites, peltate glandular trichomes, and pathway gene expression in *Artemisia annua* L. Plant Physiol Biochem 74:70–83

Yadav RK, Sangwan RS, Srivastava AK, Maurya S, Sangwan NS (2015) Comparative profiling and dynamics of artemisinin related metabolites using efficient protocol and expression of biosynthetic pathway genes during developmental span of two elite varieties of *Artemisia* annua L. J Plant Biochem Biotechnol 24(2):167–175

Yi Q-y, Yi S-w, Qin T-f, Li C-y (2018) Flavour Fragr J 33(1):75–81. https://doi.org/10.1002/ffj.3427

Yin J, Khayatnezhad M, Shakoor A (2021) Evaluation of genetic diversity in geranium (Geraniaceae) using RAPD marker. Genetika 53(1):363–378

Youssef NM, Taha LS, Abd El-Kha SN (2021) Antioxidant activity active compounds and oil characterization of in vitro *Sequoia sempervirens* cultures. Asian J Plant Sci 20(4):673–683. https://doi.org/10.3923/ajps.2021.673.683

Yu X, Liang C, Fang H, Qi X, Li W, Shang Q (2018) Variation of trichome morphology and essential oil composition of seven Mentha species. Biochem Syst Ecol 79:30–36

Yunus A, Hikam MP, Widyastuti Y (2018, March) Polyploid response of *Artemisia* annua L. to colchicine treatment. In IOP Conference Series: Earth and Environmental Science (Vol. 142, No. 1: 012020). IOP Publishing