Chapter 6
Germplasm Resources, Breeding Technologies and the Release of Cinnamon Cultivars 'Sri Wijaya' and 'Sri Gemunu' in Sri Lanka

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6.1 Introduction

6.1.1 Distribution of the Genus Cinnamomum

The plant family Lauraceae to which cinnamon belongs, encompasses about 50 genera and 2500–3000 species of trees and shrubs widely represented in Asian and American tropics, also with large numbers in Australia and Madagascar (van der Werff and Richter 1996). The genetic resources of a cultivated species can be discussed in terms of variability of that species alone or in a broader sense by considering the diversity of related species. It is worthwhile to discuss both these aspects in the case of cinnamon because there are many other species within the genus that are used as cheap substitutes for true cinnamon and also may well be used in future for transfer of valuable genetic traits.

The number of species described in the genus *Cinnamomum* varies from 100 to 341 depending on the taxonomic separators used. These species are spread across Asia, Australia, the Pacific islands and Fiji (Kostermans 1957; Purseglove 1974). The transfer of 68 American neotropical species of *Phoebe* Nees. in the 1960s (Kostermans 1961) substantially increased the number of species in the genus and gave it a pantropical status. Asiatic *Cinnamomum* species are characterised by a swollen pedicel and a fruit cup with remnants of tepals. The main identifying features of *Cinnamomum* species, according to Kostermans (1980), are the length of

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the basal and sub-basal ascendant veins, the covering of hairs (indumentum) and the thalamus cup under the fruit.

Cinnamomum cassia Presl. is the major producer of cassia (also called cassia cinnamon and Chinese cinnamon). Indonesian cassia, *Cinnamomum burmannii* Blume., is endemic to Java and Sumatra, and is the other major cultivated species of cinnamon. It is exported mainly to the United States (Krishnamoorthy et al. 1997). *Cinnamomum culitlawan* (L.) Kosterm. is a native of Malucca, Ambon and adjacent islands. Its bark has a clove smell and the oil is rich in eugenol. The cultivation of this species was attempted in India and Malaysia in the nineteenth century without success (Purseglove 1974; Weiss 2002). Northeastern parts of India are home to *Cinnamomum tamala* (Buch.-Ham.) Nees. & Eberm. (Krishnamoorthy et al. 1997), *Cinnamomum bejolghota* (Buch.-Ham.) and *Cinnamomum impressinervium* Meissner (Ravindran and Babu 2004). Commercial Indian cassia is mainly *C. tamala* (Buch.-Ham.) Nees. & Eberm., its leaves are widely used as a spice and its oil has many medicinal uses (Sharma and Nautiyal 2011). *Cinnamomum loureiroi* Nees., native to Vietnam was earlier considered to be exported in large quantities from Vietnam and was coined the names Vietnamese or Saigon cassia. According to Dao (2004), *C. loureiroi* Nees. is a rare species, and what is exported from Vietnam is now considered as *C. cassia*. Because of different harvesting and processing practices in Vietnam to that in China, the product is distinctly different and hence was considered to be derived from a different species (Dao 2004; Ravindran et al. 2004).

Kostermans (1983) described 13 species of *Cinnamomum* that occur in different parts of South India, and Krishnamoorthy et al. (1997) report eight of these to be endemic to the Western Ghats. These include *C. malabatrum* (Burm.f.) Bl., *C. perrottetii* Meissn., *C. riparium* Gamble., *C. keralanese* Kostem., *C. travancoricum* Gamble., *C. sulphuratum* Nees., *C. wightii* Meissn. and *C. heyneanum* Nees. *C. macrocarpum* is found only in the Nilgiris range along with some of the endemic species mentioned for the Western Ghats. Another five species exist in the Western Ghats although they are not endemic to that region (Krishnamoorthy et al. 1997). *C. glaucescens*, occurring in Nepal and *C. deschampsia* Gamble. in Malaysia have also been described as substitutes and adulterants of cinnamon (Weiss 2002).

Cinnamomum camphora has been heavily exploited as a source of camphor in Japan and Taiwan until the Second World War, then introduced to India during the 1950s. Owing to the availability of cheap synthetic camphor, the international demand for the natural form has declined in recent years. The use of *C. camphora* as a source of Ho leaf oil, on the other hand, has expanded in recent years and Chinese Ho wood oil has largely displaced the use of rosewood as a source of natural linalool (Frizzo et al. 2000).

A comprehensive review of morphological and anatomical features as well as biosystematics and nomenclature has been published in a review by Ravindran et al. (2004). Fujita (1967) looked at the classification and phylogeny of 25 *Cinnamomum* species in terms of the chemical constituents of their essential oils and proposed a cubic system of the genus. He also found that *C. tamala* Nees. has constituents of essential oils that are very similar to *C. zeylanicum.*

6.1.2 **Cinnamomum** *Species Endemic to Sri Lanka*

In addition to being the centre of origin of true cinnamon, *C. zeylanicum* Blume., Sri Lanka is home to another seven Asian cinnamons: *C. citriodorum*, *C. litseaefolium*, *C. ovalifolium*, *C. sinharajaense*, *C. capparu-coronde*, *C. rivulorum* and *C. dubium.* However, none of these are exploited commercially (Sritharan et al. 1993; Pathirana 2000).

6.1.3 Cytology

Chromosome studies undertaken by various workers have revealed that all *Cinnamomum* species contain a somatic number of 24 chromosomes, and hence, the genus has a basic chromosome number of 12 (Kostermans 1957; Sharma and Bhattacharya 1959; Okada and Tanaka 1975; Sritharan et al. 1993). Sritharan et al. (1993) found that fixing the root tips in the morning between 11 and 11.30 a.m. gave a significantly higher number of cells undergoing mitosis and suitable for cytological studies.

6.2 Distribution and Genetic Resources of *Cinnamomum zeylanicum* Blume.

Among the *Cinnamomum* species, a special place is occupied by *C. zeylanicum* Blume. for its delicate flavour and aroma. As a result, European importers have separate standards for Ceylon cinnamon (*C. zeylanicum*) and cassia (*C. cassia*) (CBI 2019).

6.2.1 Origin and Distribution of **C. zeylanicum**

True cinnamon or commercial Ceylon cinnamon is endemic to Sri Lanka (Kostermans 1983). South India and the Tennesirim hills of Myanmar have also been noted for natural populations of *C. zeylanicum* (De Guzman and Siemonsma 1999) and it is very likely that its introduction to these areas in the past centuries has resulted in natural populations. This is confirmed by the high degree of invasiveness of *C. zeylanicum* in these regions as shown by Fleischmann (1997) and CAB International (2019). Among 107 alien woody species in two of the main islands of the Seychelles, *C. zeylanicum* was confirmed as one of the most prominent invaders, accounting for 18.9% of the total invasions recorded. Endemic bulbul bird (*Hypsipetes crassirostris*) and introduced mynah bird (*Acridotheres tristis*) were the main dispersal agents. Moreover, some of the descriptions of *C. zeylanicum* (*C. verum*) collected from India have been attributed to taxonomic errors (Kostermans 1983). According to CAB International (2019), *C. zeylanicum* is present in 10 Asian countries, including eight states in India, 12 African countries, 9 countries in the Central American and Caribbean Region, 5 South American countries and almost all main islands of the Pacific. It has become an invasive species in several island nations (CAB International 2019).

C. zeylanicum is a tree of the wet tropics and grows naturally from sea level to 1500 m, but most of the productive plantations occur from sea level to 500 m in Sri Lanka and somewhat higher in India. It is in these areas that the natural diversity of *C. zeylanicum* can also be observed. In Sri Lanka, the areas of diversity coincide with humid, wet areas where forests have been cleared for plantation agriculture, so the natural populations of most of the *Cinnamomum* species are threatened (Pathirana 2000), although establishment of cinnamon plantations in the low and mid-country wet zone of Sri Lanka in the period 1767–1770 by the Dutch (Weiss 2002) helped to prevent further erosion of genetic resources through forest harvest.

An average rainfall of 2000–2500 mm is optimum in the major production areas: Sri Lanka, India and Seychelles (Purseglove 1974; Weiss 2002). Ranatunga et al. (2004) considered 1250–2500 mm rainfall with temperatures of 20–30 °C as optimal for production of quality cinnamon in Sri Lanka.

6.2.2 Germplasm Resources **of C. zeylanicum** *Blume.*

Despite the importance of cinnamon as a spice from ancient times, only a limited effort has been made to scientifically describe the large genetic variation that has been generated in this species through natural evolution and through selection in the past few centuries of cultivation. Compared with other tree spices such as clove and nutmeg, cinnamon is more genetically diverse (Krishnamoorthy et al. 1997). Nevertheless, with less than three centuries in cultivation since the Dutch established the first plantations in Sri Lanka and the long periods between replanting, cinnamon can be considered to be in its early stages of domestication. Considering cinnamon's value as a spice crop, it is logical that the intraspecific classification in Sri Lanka has been based more on organoleptic characteristics than on morphological characters. The vernacular names in Sinhalese identify different types as *Peni Miris Kurundu* (sweet & hot cinnamon), *Thitta Kurundu* (bitter cinnamon), *Peni Kurundu* (sweet cinnamon), *Naga Kurundu* (snake cinnamon), *Veli Kurundu* (sandy cinnamon implicating the granular nature), *Sevel kurundu* (slimy cinnamon) and *Kahata Kurundu* (acrid cinnamon) (Anonymous 1996). However, none of these types can be clearly identified in cultivated cinnamon at present (Wijesinghe and Pathirana 2000).

Research and development of cinnamon began in post-independence Sri Lanka by the Department of Minor Export Crops (the current Department of Export Agriculture—DEA), but systematic study of the germplasm was made possible only after the National Cinnamon Research and Training Center (NCRTC) (Formerly Cinnamon Research Station) opened at Thihagoda in the Matara District in southern Sri Lanka where more than 30% of the national cinnamon crop is produced. Cinnamon production in the Matara District (6035 Mt) comes a close second only to the adjacent Galle District (6683 Mt) (Kumari Fonseka et al. 2018).

A systematic evaluation of 11 elite cinnamon progenies was conducted by Perera (1984), which led to the identification of W-37/3, W-16-1 and W-17/9 as lines with high bark quality. As a result of a field survey for elite material conducted by the NCRTC in the 1980s, 700 superior accessions were identified from the growing areas. The selections were based on morphological characters and pungency of the leaf petiole (Wijesinghe and Pathirana 2000). During the second phase of the germplasm enhancement programme, 400 superior selections were established from green-wood cuttings at the NCTRC. Screening of the collection for characters based on a selection index began in 1996. The characters considered for indexing included the total number of branches, number of harvestable branches, erectness of stem, susceptibility to major pests and diseases, peeling ability, bark pungency and dry weight/fresh weight ratio (Wijesinghe and Pathirana 2000—Table 6.1).

One hundred superior genotypes selected from this study were then screened for bark oil content. Eight lines recorded more than 3% bark oil on a dry weight basis, compared with a mean of 1.68% in the 100 lines tested, with a range of 0.49–3.70% (Wijesinghe and Pathirana 2000—Table 6.2). Ten superior lines from this study were established in multi-location yield trials in the final stage of the screening process. As the outcome of long-term evaluation of these 10 lines in multi-location trials, two lines were released as superior cinnamon cultivars for the first time in 2009 in Sri Lanka, called ‘Sri Gemunu’ and ‘Sri Wijaya’. The variety ‘Sri Gemunu’ has a distinctive sweet and pungent flavour, and an aroma characteristic of Ceylon cinnamon, produces average bark yield of 1200–1300 kg/ha/year, and has high bark oil content (3.25–3.6%), leaf oil (3.6–3.9%) and cinnamaldehyde content (80–83%) of bark oil. Variety ‘Sri Wijaya’ produces high bark yield of 1600–1800 kg/ha/year with a higher content of eugenol (88–92%) in leaf oil. These varieties are clonally

Table 6.1 Selection indices developed by the NCTRC for selecting superior cinnamon genotypes

Character	Total score
Total stems	10
Number of harvestable stems	5
Erectness of stem	15
Major pests	10
Major diseases	10
Peeling ability	10
Pungency of the bark	10
Yield ratio: dry weight/(fresh weight × 100)	30
Total	100

Table 6.2 Bark oil percentage of 20 superior selections after screening 100 clones in the Matara District, Sri Lanka

Clone ID	Bark oil (%)	Clone ID	Bark oil (%)
VP-241	3.7	VP-110	2.6
VP-126	3.6	VP-218	2.56
VP-203	3.5	VP-219	2.43
VP-307	3.19	VP-098	2.41
GACG 1-1	3.1	VP-340	2.34
VP-179	3.09	VP-028	2.3
VP-308	3.07	VP-049	2.24
VP-334	3.05	VP-161	2.24
VP-116	2.7	VP-085	2.17
VP-05	2.65	VP-186	2.12

Mean of 100 clones tested—1.68%; range—0.49–3.7%

Fig. 6.1 Cultivars 'Sri Gemunu' (left) and 'Sri Wijaya' (right) released for cultivation in Sri Lanka by the Department of Export Agriculture as a result of clonal selection

propagated and distributed to growers. The morphological features of two released varieties in Sri Lanka are shown in Fig. 6.1.

Wijesinghe and Pathirana (2000) have shown the danger of genetic erosion of this indigenous crop because of the development and spread of vegetatively propagated elite clonal material. To ensure the conservation of diverse genetic resources, they proposed the establishment of isolated seed gardens of cinnamon in the Intermediate and Wet Zones of Sri Lanka. The seed-derived plantations established over the past few centuries in Sri Lanka, initially from elite selections by the growers themselves and subsequently by the Department of Export Agriculture, are a valuable genetic resource for the further improvement of cinnamon. Further sampling from these populations for the genetic collections at the NCTRC will add value to this resource.

The other major collection of true cinnamon has been established at the Indian Institute of Spices Research (IISR) at its experimental farm in Peruvannamuzhi in Kerala State where most of India's cinnamon is grown (Krishnamoorthy et al. 1996). This collection consists of about 300 accessions of *C. zeylanicum* and another 64 wild *Cinnamomum* species (Krishnamoorthy et al. 1988, 1997). Accessions from the commercially important *C. zeylanicum* are derived mainly from selected plants of cinnamon estates of Ancharakandi in Kannar District and Mangalamcarp Estate in Wynad, both in Kerala. Plants in these oldest cinnamon estates were established from seeds brought from Sri Lanka by the British in the nineteenth century (Ravindran et al. 2004). Another 105 Sri Lankan progenies from 13 accessions were added to this collection in 1970 and 1979 (Krishnamoorthy et al. 1997). Thus, the two major collections in India and Sri Lanka have a common ancestry. Nevertheless, selection within the original heterogeneous collections under local conditions over several generations of cultivation has resulted in genetic variation for agronomic characters, including tolerance to various environmental factors. Thus, for example, there are distinct populations of cinnamon adapted to the drier conditions of the Intermediate Zone of Sri Lanka and for the higher rainfall areas in the Wet Zone.

In India, testing of 12 Sri Lankan progenies introduced in the 1970s along with another 291 superior lines selected at IISR resulted in the release of Sri Lankan line SL 53 and Indian line IN 189 for cultivation with the names 'Navashree' and 'Nithyashree', respectively (Krishnamoorthy et al. 1996). According to Peter and Nirmal Babu (2009), five high yielding varieties of cinnamon have been recommended for release.

6.3 Genetic Studies on Agronomic and Quality Traits of *C. zeylanicum* Blume.

Most of the genetic studies in cinnamon have focused on the variability for characters that contribute to the yield or quality of the bark or oil. Even morphological and anatomical studies have been directed mainly towards the identification of structures related to oil biosynthesis. For instance, a study conducted at NCRTC to investigate different morphological features of *C. zeylanicum* in relation to yield and quality of bark and leaf revealed seven different leaf types on the basis of leaf morphology and a significant positive correlation between bark yield and leaf characters. The trees with large round leaves showed high bark yield while plants with inwardly curved leaves showed higher cinnamaldehyde percentage in bark oil. High leaf oil content was found in trees with small round leaves (Wijesinghe and Gunarathna 2001). However, the mode of inheritance of such characters has not been published, owing to limited or no crosses, the heterozygous nature of the crop and, being a tree crop, the long time required for each generation. Nevertheless, few studies of the genetic variation in agronomic characters have been conducted.

6.3.1 Genetic Variation Within C. zeylanicum

Krishnamoorthy et al. (1992) analysed nine characters for variation and correlations in a collection of 71 cinnamon accessions from India and Sri Lanka maintained at Peruvannamuzhi, India. A high level of variability was observed for the dry and fresh weight of the bark (8–305 g with a mean of 64.7 g and 30–840 g with a mean of 207 g, respectively), the oleoresin content of the bark (1.3–20.0% with a mean of 8.5%) and the oil content (0.72–4.8% with a mean of 2.0% for leaf oil and 0.5–3.85% with a mean of 1.81% for bark oil). Recovery of bark was also highly variable, ranging from 10.7% to 80% with an average of 32.1%, and the data were skewed towards lower recovery rates. Leaf length, breadth and leaf size index (derived from length and breadth) were not as variable as the yield parameters, probably because selection for yield may have indirectly selected for an optimum leaf size. Correlations were performed on these data, and as expected, the highest correlation was between dry and fresh weights of bark. There were significant positive correlations between dry weight of the bark and leaf oil percentages, and a negative correlation between bark oil and leaf oil. The authors used data from the collection of varieties, hence these correlations can be categorised as genotypic and will be useful in planning selection. However, the results are based on the measurements from a single plant of each of the accessions and need to be interpreted cautiously. Similar results were shown while evaluating ten clonally propagated cinnamon accessions with elite characteristics selected from germplasm collection at NCRTC, Sri Lanka. Bark yield, oil percentage of bark and leaf, cinnamaldehyde percentage in bark oil and eugenol percentage in leaf oil, showed highly significant differences among the selections.

Krishnamoorthy et al. (1991) studied the progeny performance of nine lines and found a significant variation in plant height, number of branches per tree, fresh and dry bark weight and bark recovery. Considering the heterozygous nature of cinnamon and its pollination behaviour, high variability can be expected among progenies even from the same plant after controlled hybridisation, and should be taken into consideration in breeding programmes.

Joy et al. (1998, as quoted by Ravindran et al. 2004) studied leaf oil yield in a collection of cinnamon accessions in Odakkali, India and found a variation of 36.5–294.7 mL/tree/year. However, oil content is highly dependent also on the weather conditions and therefore multi-year, multilocation studies would reveal the genotype × environment interaction useful for selecting high oil genotypes. In a collection of 239 cinnamon accessions Krishnamoorthy et al. (1988) found 55.6% plants to have green flush colour and the others to be purple. A significantly higher bark oil percentage (1.84%) was recorded in varieties with purple flush colour than in varieties with green flush (1.43%). There were no significant differences in bark oleoresin content or leaf oil percentage between the two groups.

Analysis of morphological characters from 47 cinnamon accessions collected within Matara district, the second largest cinnamon cultivation district in Sri Lanka, resulted in nine groups indicating variation for leaf and bark characters (Azad et al.

2016). Although the authors used 14 characters in their study, 11 of those were related to leaf morphology. The same authors have studied the morphology of a segregating population from two plants selected in a plantation and reported a high variability in leaf characters (Azad et al. 2015), as expected in a highly heterozygous species.

6.4 Interspecific Variation in *Cinnamomum*

Variation has been extensively studied in relation to the taxonomy of different *Cinnamomum* species. Balasubramanian et al. (1993) used petiole anatomy to characterise four species. They found differences in shape of the petiole and the vascular strands in *C. camphora*, *C. zeylanicum*, *C. macrocarpum* and *C. tamala.* Using 20 morphological characters in a cluster analysis involving eight *Cinnamomum* species, Ravindran et al. (1991) showed that *C. malabatrum* is closely related to *C. zeylanicum.* The species *C. camphora*, *C. perrottetii*, *C. riparium* and *C. cassia* were very distinct types and formed independent clusters.

More recently, Ariyarathne et al. (2018) conducted a study on morphological and chemical characteristics of two selected true cinnamon accessions ('Sri Wijaya' and 'Sri Gemunu') and six wild species of the genus *Cinnamomum* (Schaeff.) endemic to Sri Lanka, that is, *C. citriodorum*, *C. capparu-coronde*, *C. litseaefolium*, *C. revulorum*, *C. dubium* and *C. sinharajaense*. They examined morphological characters of flower, leaf, seed and bark and bark yield and leaf oil content, as well as the chemical characters of leaf and bark oil. A dendrogram (Fig. 6.2) was constructed and a diagnostic key (Fig. 6.3) was developed based on oil yield, and morphological and chemical characters. The analysis revealed that the studied species could be

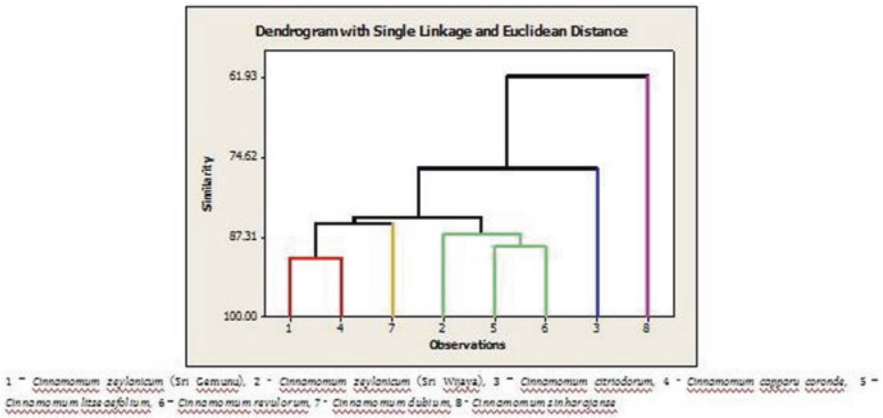

Fig. 6.2 Dendrogram developed by Ariyarathne et al. (2018) illustrating the relationship between six *Cinnamomum* species endemic to Sri Lanka and two *C. zeylanicum* cultivars released by the Department of Export Agriculture, Sri Lanka

1. Pinnate venation	*C. citriodorum*
1. Three veined venation with prominent midrib and two lateral veins	
2. Eugenol present in leaf oil	
3. Cinnamaldehyde absent in leaf oil	*C. sinharajaense*
3. Cinnamaldehyde present in leaf oil	
4. Leaf lamina shape - oval	
5. Leaf apex acute	*C. liseaefolium*
5. Leaf apex ovate to elliptic	*C. zeylanicum* ('Sri Gemunu')
4. Leaf lamina shape - lanceolate	
5. Leaf apex acute	*C. zeylanicum* ('Sri Wijaya')
5. Leaf apex acuminate	*C. capparu-coronde*
2. Eugenol absent in leaf oil	
3. Contains benzyl benzoate in bark oil	*C. revulorum*
3. Benzyl benzoate absent in bark oil	*C. dubium*

Fig. 6.3 Key to *Cinnamomum* species endemic to Sri Lanka including the two cultivars of *C. zeylanicum* released by the Department of Export Agriculture. (Modified from Ariyarathne et al. 2018)

grouped into five clusters. *C. sinharajanse* was entirely different from the rest of the tested species and formed a separate cluster. 'Sri Gemunu' and 'Sri Wijaya' being improved accessions of *C. zeylanicum* exhibited less variability in leaf size, colour, fragrance etc. but showed high variations in shape and the apex of the leaf (Ariyarathne et al. 2018).

A comparison of leaf morphology of the two released cinnamon cultivars and the cinnamon species endemic to Sri Lanka is presented in the Fig. 6.4.

6.5 Reproductive Biology of Cinnamon in Relation to Breeding

6.5.1 Morphology of the Floral Organs

Cinnamon flowers are produced on long green peduncles in lax terminal or axillary panicles. They are insect pollinated, mostly by honey bees (*Apis cerana*, *A. corea*, *A. dorsata*) in India (Mohankumar et al. 1985). Flowering occurs during November to March in most cinnamon-growing countries and about 12% of open-pollinated flowers produce fruits when there are sufficient insect pollinators.

In the cinnamon flower, six tepals are organised in two whorls with three tepals per whorl. It has a trimerous androecium, essentially with four whorls, although the outer whorl is reduced and represented by staminodes. Fertile stamens in cinnamon show valvular dehiscence (Ravindran et al. 2004). The ovary is superior, unilocular with a single pendulous anatropous ovule and a long style. The stigma is dry and papillate (Heslop-Harrison and Shivanna 1977).

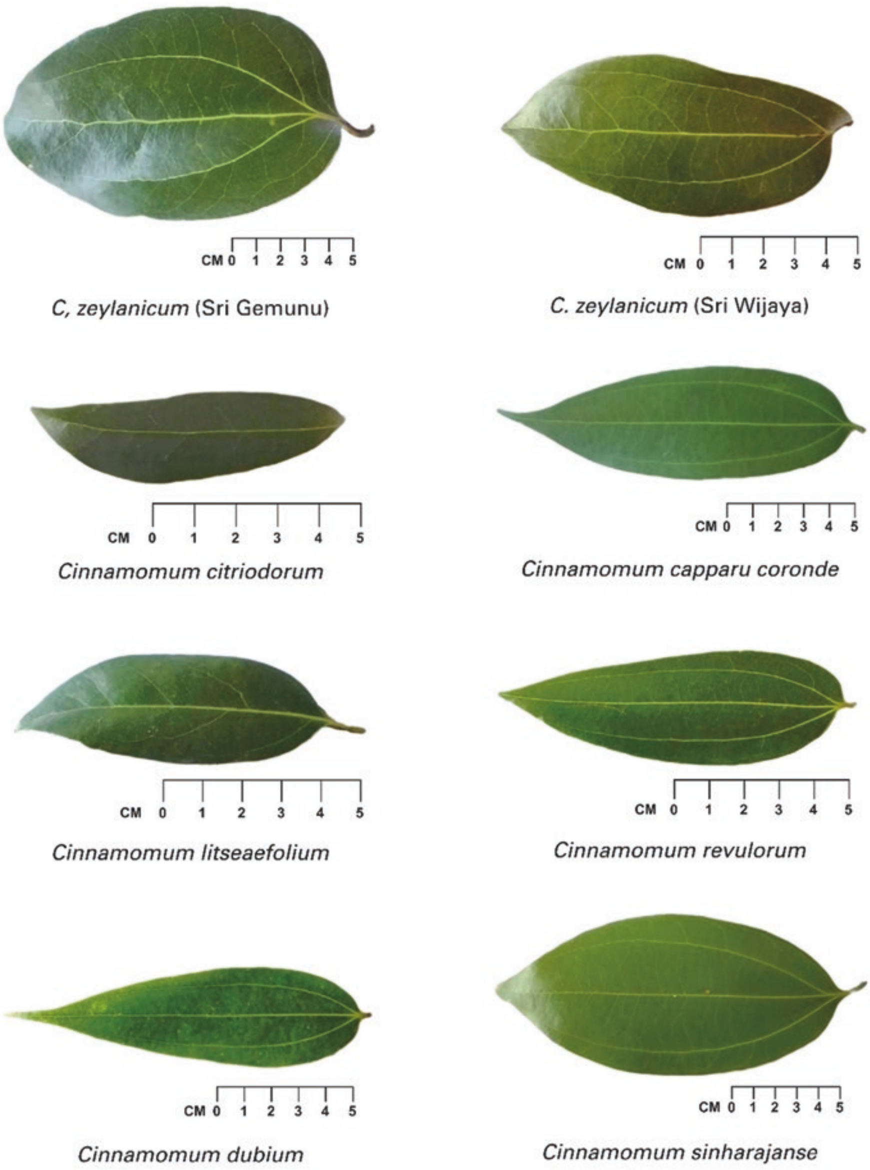

Fig. 6.4 Leaf morphology of cinnamon species native to Sri Lanka in comparison with two released cultivars of *C. zeylanicum*

6.5.2 Synchronised (Protogynous) Dichogamy as a Mechanism Ensuring Cross-Pollination

Many species of the Lauraceae show dichogamy, both in old and new world taxa (Kubitzki and Kurz 1984). A conspicuous trait of the floral biology of Lauraceae is a very pronounced protogyny, which Kubitzki and Kurz (1984) described as a prerequisite to the phenomenon of synchronous dichogamy, in which each flower opens twice, either in the morning or in the afternoon of the first day, and the next day either in the afternoon or in the morning, respectively. This phenomenon has also been confirmed in *C. zeylanicum* in Sri Lanka (Wijesinghe and Pathirana 2000). In Sri Lanka, flowering season starts in November and lasts till early March. In cinnamon, it is now known that the maturing of female phase precedes the male phase by about 24 h ensuring outcrossing, thus this phenomenon is identified as protogynous dichogamy.

Synchronous dichogamy also occurs in avocado (*Persea americana*), the other economically important species of Lauraceae (Sedgley and Griffin 1989). Synchronous dichogamy in *P. americana* requires the presence of two morphs, whose flowering has to be complimentarily synchronised. Pollen transfer is possible only between individuals belonging to the two morphs, providing that rain or other disturbing factors do not lead to an asynchrony of flower opening. Thus, synchronised dichogamy represents a kind of temporal dioecism. Periods of light and dark have been implicated in the control of synchronous dichogamy of avocado (Sedgley 1985).

Unlike the hermaphroditic Amazonian taxa of Lauraceae, Asiatic *Cinnamomum* species produce ample nectar from the glands flanking the inner stamens and the staminodes corresponding to the fourth staminal whorl. In the female phase of flower opening, the outer stamens lie pressed against the petals, and the stigma appears brilliantly white and receptive. In the intermediate phase, the petals close and the stigma starts wilting. First, dark spots appear on the stigma until it becomes discoloured and is no longer receptive. In the second phase of flower opening, stamens stand up prominently in the centre of the flower and shed their pollen. Pollen remains attached to the inner surface of the valves, which are by now bent upwards. More nectar is produced during the second opening. All flowers of a given tree maintain the same diurnal rhythm of development, in that flower opening occurs either in the evening and the following morning, or in the morning and the evening of the following day. This excludes any possibility of pollination between flowers of the same tree. The phenomenon can be considered as the forerunner of dioecy present in some Amazonian taxa of Lauraceae described by Kubitzky and Kurz (1984). Both morphs appear to occur randomly in any given population of cinnamon, even within the same seed-propagated variety.

6.5.3 Controlled Pollination

Although the synchronised dichogamy described above ensures cross-pollination, it is customary among breeders to emasculate flowers the day before controlled pollinations are made. For success in controlled pollination, it is essential to select plants from two morphs in which androgenesis coincides with gynogenesis. Although pollinations can be made with preserved pollen or by artificially creating conditions for asynchrony of flowering so that pollen from flowers of the same morph can be used, it is possible that other incompatibility systems may prevent fertilisation success. Furthermore, during high humidity, plants that shed pollen in the afternoon may still be shedding viable pollen the following morning. During rainy days, asynchrony may even lead to the simultaneous availability of pollen and receptive stigmas on one plant. Attempts by Kubitzky and Kurz (1984) and Wijesinghe and Pathirana (2000) to achieve such geitonogamous pollinations failed, thus demonstrating the existence of other fertility barriers between morphs. Weerasuriya et al. (2016) were successful in hybridising ‘Sri Gemunu’ (Type A variety) with ‘Sri Wijaya’ (Type B Variety). After artificially pollinating 250 flowers taking into consideration the active period of the male and female stages, they produced 46 F_1 seedlings. The evaluation of this hybrid population is ongoing (Weerasuriya et al. 2016).

For emasculation, the tepals can be opened up with a pair of forceps while holding the base of the flower with the other hand; the stamens are then carefully detached. After emasculation of selected flowers from an inflorescence, the other flowers should be removed and the inflorescence covered by a bag to prevent any uncontrolled pollination. On the next day, pollen from the male parent can be directly transferred onto the stigma of the female parent, either by using detached male flowers or a soft brush to collect the pollen. Cinnamon pollen clump on the opened valve of the pollen sac, so it is relatively easy to pollinate selected plants using detached flowers of the male parent. Alternatively, anthers can be separated from the rest of floral parts before dehiscence and pollen can be extracted from freshly opened flowers into containers.

Pollen viability should be assessed by staining tests or by in vitro germination, especially if stored pollen is being used for pollination. Popular stains for this purpose include acetocarmine or iodine. For in vitro germination tests an agar solidified 28% sucrose solution containing 20 ppm boric acid and about 2.5 mL yeast extract may be used. Selective crossings may also be achieved by growing the plants of separate morphs of two selected parent varieties in isolation, but there is no scientific information on how to select these before flowering. Therefore, this may be achieved by growing a number of plants from the two varieties selected for crossing in an isolated plot, observe the flowering behaviour when they reach the reproductive stage and remove all plants of one morph from the first variety and all plants of the other morph of the second variety to ensure coincidence of gynogenesis of one variety with the androgenesis of the second.

6.6 Handling Hybrid Seeds and Populations

Once the fruit matures, the pulp should be removed from freshly collected berries and the seeds extracted, washed and air-dried. Uneven maturity has to be considered when harvesting berries. As with many tropical tree species, cinnamon is recalcitrant (Pathirana 2000) and therefore seeds have to be sown for germination without delay. Samaraweera (2000) reports that without special treatment, germination of seeds can drop to 40% within a period of 3 weeks after harvest, and a complete loss of viability can occur by the seventh week. However, in our recent experiments, we found that air-dried seeds stored in airtight bags, treated with 0.05% Folicure® solution can maintain a viability of above 80% for 4–6 months (Weerasuriya et al. unpublished). Seed size also seems to affect seed germination and seedling growth, with larger seeds recording higher germination percentages and improved seedling vigour (Samaraweera 2000).

A moist seedbed in a screen house results in better germination than in the open field. Seedlings 8–10 months old can be transplanted into unprotected fields during the rainy season. Field establishment is 80–90% when seedlings are raised in polythene bags compared with 60–70% establishment for seeds germinating in a seedbed prior to transplanting in the field (Abeykoon 2000). Therefore, seed from cross-pollinations should be raised in bags before transplanting into the field.

Seed progeny of each cross should be planted in separate plots and evaluated for germination and vigour of growth. The crosses can be handled in different ways in tree species such as cinnamon, depending on the breeding plan. They can be individually assessed for the characters of interest, and promising plants can serve as parents for establishing clonal progenies for further evaluation for release as varieties. This approach allows quicker release of varieties. Maintaining records of yield and other agronomic characters in parents and progeny of different combinations of crosses allows the identification of varieties with good combining ability for use in the future. On the other hand, if the objective of the breeding programme is to combine superior characters of two selected varieties, then it is more efficient if the progenies are grown in isolation, the inferior plants culled, the superior plants allowed to flower and seeds collected for the second and subsequent generations. In each generation, only the plants with superior characteristics should be allowed to produce seed.

6.7 Implications of the Juvenile Period and Vegetative Propagation in Cinnamon Breeding

6.7.1 Juvenile Period in Relation to Breeding of Cinnamon

Most long-term tree breeding programmes are based upon several generations derived from controlled crosses. The selected superior plants can serve as candidates for variety release while serving at the same time as parents for producing the next seedling generation. The genetic progress of a cinnamon-breeding programme would depend on the length of the juvenile period of each generation, and approaches to shorten the juvenile period would considerably increase the genetic gain.

Zimmerman (1972) showed a correlation between plant height and flowering in fruit trees, which may be related to a minimum number of nodes being required to overcome the juvenile period. Hence, as in fruit crops, heavy fertilisation enhancing vigorous growth may help in achieving early flowering in cinnamon too. Another approach is to train the tree to a single stem in dense plantations as is used for chestnut (Zimmerman 1971). Agronomic and physiological approaches to shorten the juvenile period, specifically focused on cinnamon, would certainly help in long-term breeding.

6.7.2 Vegetative Propagation in Relation to Breeding of Cinnamon

Almost all existing cinnamon plantations have been established from seedlings, mainly from selected parent plants but not from seedling progenies of controlled pollinations. In this regard, work by Haldankar et al. (1994) is unique. The researchers initially used 300 selected seedlings from Sri Lankan cinnamon collection at IISR in Calicut for screening under coconut and then vegetatively propagated the best four plants to establish a yield trial in Maharashtra State, India. Breeding line B-IV gave the highest bark yields for both fresh weight (230 g) and dry weight (84.5 g). Their approach was to screen the seedlings on the basis of organoleptic properties and use selected seedlings for further screening for yield and chemical composition of the oil. As a result, line B-IV isolated has a high percentage of cinnamaldehyde (70.2%) and eugenol (6.9%) in the bark oil. This line was later released as Konkan in the Konkan region of Maharashtra State (Ravindran et al. 2004).

Seedling progenies of the elite lines already developed in Sri Lanka (Wijesinghe and Pathirana 2000) and India (Krishnamoorthy et al. 1996) may serve as useful resources for further screening for elite material in their progenies. Parental lines for the development of elite populations can then be identified using data on yield and chemical composition of bark and leaf oil of individual plants from heterogeneous populations.

The recent successes in vegetative propagation of cinnamon both in Sri Lanka (Dayatilake 2000; Samaraweera 2000) and described in Chap. 7 of this book) and in India (Benerjee et al. 1982; Hegde et al. 1989; Ranawere et al. 1994, 1995; Thirunavoukkarasu 1996) have resulted in the possible production of clonal progeny from selected elite plants. Unfortunately, there are no reports so far published on the field performance of vegetatively propagated material in cinnamon, except for the study by Haldankar et al. (1994) on four selected clones. Unlike other plantation crops such as tea, cinnamon is grown by smallholders with minimal inputs

(Kularatne 2000). The plants have a long productive period in the field and because of a prolific root system are able to survive and yield under adverse weather with minimum inputs. Moreover, the populations have good tolerance to biotic stresses as no outbreaks of diseases or pests have been reported. In other crops where production is based on clonal material, for example, coffee, significant disease outbreaks have already occurred in Sri Lanka. Therefore, it is essential that the diversity of cinnamon be maintained, even under clonal propagation, by the release of several clones of diverse origin. Disease and pest build-up need to be monitored carefully and the performance of clonal material should be compared with improved seedling populations before converting already established seedling plantations into clonal ones.

6.8 Breeding Strategy and Objectives

Cinnamon growers in the past have consciously or unconsciously attempted to develop favourable populations in their fields by selecting superior phenotypes from natural populations. This grower-driven mass selection has resulted in the current diversely adapted populations of cinnamon in different parts of Sri Lanka and other cinnamon growing countries today. Faster gains in desirable characteristics can be expected if more sophisticated and targeted breeding approaches are adopted. Experiences from the more advanced perennial crop breeding programmes can be used in developing the strategies for cinnamon breeding.

6.8.1 The Need for Long-Term Planning, Funding and Collaboration

Perennial crop breeding is particularly costly in terms of land, labour, time and infrastructure facilities. It is therefore essential that short- and long-term breeding plans are well thought-out and executed, and supported by assured long-term funding. In this regard, partnership arrangement between central government, provincial agencies and private grower associations should be an essential feature. Obviously, the nature and scope of the breeding programme should depend on the needs and demands of the cinnamon industry. Encouraging the growers to contribute financially to the breeding effort will give them ownership of the programme and at the same time the breeders will have a source of long-term funding. However, private funding should not be allowed to hijack the long-term breeding objectives by overemphasising frequent variety releases as experienced in private breeding programmes.

The relatively high cost per plant due to larger size and longer generation time of cinnamon will be compensated to a great extent by resorting to clonal propagation for the mass production of elite material in the future. Nevertheless, a faulty strategy can nullify all breeding efforts and therefore proper scientific planning of the breeding programme is of utmost importance. Control of pollination is crucial and should be planned from the very outset; without such controls an entire 5–8-year breeding cycle can be lost. The information generated by a programme will be of no use for future breeders unless the characteristics of elite parental plant types can be described and documented. With more sophisticated molecular tools becoming available for breeders, an attempt must be made to enter into long-term collaboration with a laboratory that has capabilities in molecular markers.

6.8.2 Cultivated and Wild Gene Pools

Another important aspect of any breeding plan is the maintenance and use of a diverse gene pool that is known to contain desirable phenotypes. In this respect, Sri Lankan breeders have the luxury of the cultivated and wild gene pools of *C. zeylanicum*. The cultivated crop has a higher gene frequency of desirable traits compared with natural populations and therefore a very strong justification will be needed if extensive use of the wild gene pool is contemplated. If traits such as disease and insect pest resistance are to be emphasised, then the parents carrying these traits should first be screened for the overridingly important characteristics, such as bark and oil yield and quality to justify their inclusion in the breeding programme.

Moreover, clonal propagation represents monoculture in its most extreme form, with a single genotype spread over a wide area. With the two major cinnamon producing countries, Sri Lanka and India, both at the brink of making extensive use of clonal propagation for plantation establishment, the specific resistance of the crop will be negated by the natural selection of the pathogen or the pest unless the resistance was selected for durability. When deciding on conversion of seedling plantations to clonal, advantage of monocropping for the product quality will have to be evaluated against maximising the vulnerability of the crop to biotic and abiotic stress factors. The experience of the tea and rubber industries will be useful in this regard. Unlike rubber and tea, cinnamon is indigenous to Sri Lanka and plantations exist side by side with natural populations. Hence, there is a constant influx of pathogens and pests specific to cinnamon from the natural populations. It is therefore important to embark on a multiclonal strategy from the very outset, even within the same plantation.

With oil quality being an important aspect of breeding, there needs to be good analytical facilities with trained staff to support the breeding programme. In this regard, identifying the genotypes with a high frequency of genes for particular chemical traits and using them with high bark and oil yielding parents will help in combining genes for quality and yield.

6.9 Unconventional Breeding Methods

6.9.1 Interspecific Hybridisation and Embryo Culture

There are no reports on attempts to produce interspecific hybrids of *Cinnamomum* species. *C. zeylanicum* has the best quality among the different species and apparently does not need the infusion of genes from other related species for improvement. A similar argument applies to disease and pest resistance. Close relationships that seem to exist between *C. zeylanicum*, *C. tamala* and also some species indigenous to Sri Lanka (see Chap. 4 of this book for details), should allow successful interspecific hybridisation but this would be more useful in understanding taxonomic relationships than in developing new cultivars.

6.9.2 Induced Mutations

Induced mutations have been used widely to improve perennial fruit species, and the availability of vegetative propagation techniques should allow fixing of any useful mutants in a mutation breeding programme. There have been no serious attempts to improve cinnamon through mutation induction techniques and there are no reports on the effects of radiation on seeds or vegetative organs. Therefore, establishing growth reduction curves is a prerequisite (Pathirana 2011) before initiating mutation breeding programmes in cinnamon.

With the development of techniques for clonal propagation, induced mutations will become increasingly more valuable for improvement of cinnamon, because this technique allows the improvement of a single trait without affecting the other characters, as no hybridisation steps are involved (Pathirana 2011). Therefore, as a first step, the development of protocols including dose response for different mutagens will help future breeding using this technique.

6.9.3 In Vitro Techniques for Genetic Improvement

There are several reports on successes in tissue culture regeneration in cinnamon (Rai and Jagdishchandra 1987; Babu et al. 1998; Dayatilake 2000). One possible use of this technology would be crop improvement through in vitro mutagenesis as well as somaclonal variation. Tissue and cell culture techniques may find other applications in cinnamon breeding, such as in distant hybridisation through embryo rescue, induction of polyploidy and in-plant genetic transformation.

6.9.4 Molecular Genetic Approaches

Molecular genetic approaches have the potential to increase the efficiency of cinnamon breeding, understanding evolution of the genus and the genetic relationships among species. First, mapping populations involving different variety groups need to be developed. Thereafter, the construction of high density (HD) maps by developing microsatellite (SSR) and/or single nucleotide polymorphic (SNP) markers, building a multi-parent consensus map of cinnamon by comparative quantitative trait loci (QTL) analyses, and developing user-friendly molecular marker sets for future practical applications, can follow. These individual populations and the HD maps can be used to perform QTL analyses on agronomic characters, including growth, flowering, yield, oil content and quality. If several identical characters are evaluated in different genetic backgrounds, it will be possible to compare QTL locations in several genetic backgrounds. However, traditional methods for developing molecular markers are costly, time consuming and laborious. Considering the increasing affordability of the use of next generation sequencing (NGS) platforms, marker development based on high throughput sequencing for crops such as cinnamon where molecular data are limited, will be more efficient. The advent of NGS has allowed the development of SSR markers in species such as cranberry for which molecular genetics data were lacking (Zalapa et al. 2012). NGS technologies have also facilitated the development of novel SNP platforms for genotyping by sequencing (Bhat et al. 2016). For the implementation of such highly desirable DNA marker-based breeding strategies for cinnamon, it is important for the two cinnamon improvement groups in India and Sri Lanka to collaborate and to find partners in laboratories with facilities for molecular biology research. Continuous funding of such research is essential for any practical outcome to be anticipated.

6.10 Conclusions

Despite being the oldest known spice, cinnamon has been in cultivation for less than three centuries since the Dutch established the first plantations in Ceylon (Sri Lanka) in 1765. The crop is still in the early stages of domestication. There is appreciable genetic variation in agronomic and quality traits that could be used in breeding programmes. Plant breeding programmes with limited genetic collections have been established in Southern Sri Lanka and Kerala State, India. However, the crop genetic diversity still exists largely in the plantations established using seedlings, in growers' lands, and also in closely related indigenous species in the wild. Therefore, further collection and establishment of *ex situ* germplasm collections is important, especially with the release of new cultivars which are being clonally propagated and established, replacing seed-derived plantations. Establishment of isolated seed gardens from selected elite materials could also help to conserve the genetic diversity of cinnamon. Limited success in controlled pollination of elite material has been reported and the progenies are under evaluation at the National Cinnamon Research and Training Center, Sri Lanka. Synchronous (protogynous) dichogamy exhibited by the genus makes controlled pollinations difficult and therefore most of the elite material developed to date is based on progenies derived from open pollination of selected plants. This method commenced in the 1980s at the National Cinnamon Research and Training Center, Sri Lanka, resulting in the release of two cultivars, 'Sri Gemunu' and 'Sri Wijaya'. Agronomic and physiological studies leading to shortening of the juvenile phase and the availability of quick and reliable analytical facilities for oil quality are important cornerstones in improving the cinnamon crop. Advanced genetic techniques, such as molecular marker-based selection, need to be introduced into the breeding programmes to identify useful progeny and reject the undesirable ones early. High throughput next generation sequencing platforms should be considered as molecular data are limited. Considering the long-term nature of breeding programmes with this perennial crop, careful planning, availability of funds in the long term and collaboration among scientists of relevant disciplines and grower associations are important.

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