STUDIES ON GENETIC VARIABILITY AND IMPROVEMENT OF WHITE TURMERIC (*CURCUMA ZEDOARIA* ROSC.) OF KERALA

Thesis submitted
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by

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CERTIFICATE

Certified that this thesis titled **"Studies on genetic variability and improvement of white turmeric (*Curcuma zedoaria* Rosc.) of Kerala"** embodies the results of a piece of bona fide research work carried out as part fulfilment of requirements for the degree of Doctor of Philosophy in Botany of University of Calicut by Mrs. **Litty R.** under my guidance and supervision and that no part of the thesis has been submitted for any other degree.

I further certify that such helps or sources of information availed of in this connection have been duly acknowledged.

Calicut University (V.V. RADHAKRISHNAN)
09.02. 2023

DECLARATION

I, **Litty R.** hereby declare that this thesis entitled **"Studies on genetic variability and improvement of white turmeric (*Curcuma zedoaria* Rosc.) of Kerala"** being submitted in part fulfilment of requirements for the degree of Doctor of Philosophy in Botany of University of Calicut embodies the results of a bona fide research work done by me under the guidance of **Dr. V.V. Radhakrishnan**, Professor, Department of Botany, University of Calicut, Kerala, India and that no part of it has previously formed the basis for the award of any degree, diploma, associateship, fellowship, title or recognition.

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09.02. 2023

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PREFACE

Genetic diversity, defined as the magnitude of genetic variability within a population, is a crucial component of biodiversity. It has significant ecological consequences at the population, community, and ecosystem levels. It is widely recognized that the genetic diversity of valuable medicinal plants has been diminishing over the last few decades, and it is critical to preserve the existing genetic wealth for future generations. Anthropogenic habitat fragmentation isolates plant populations, raising the risk of extinction. Furthermore, uncontrolled collection of wild resources, excessive grazing, unsustainable harvests, diseases and pests, introduced species, and climate change have destroyed genetic wealth. Analysis of the existing genetic diversity within a species in quantitative parameters is crucial for the effectiveness of plant breeding practices.

Curcuma zedoaria Rosc., also known as white turmeric, is an important species of the genus *Curcuma*, belonging to the family Zingiberaceae. The plant is referred to as *Vella manjal* in Malayalam. Various parts of zedoary are used in Ayurveda and other folk medicinal systems to treat diseases such as cancer, flatulence, diarrhoea, etc. Because of the untapped therapeutic potential of the species, it could be considered a future drug. Various human activities such as habitat degradation, urbanization, and industrialization contribute to the decline of the population size of this potent medicinal plant. Understanding the degree of genetic variability in the crop will contribute to reasonable breeding approaches for exploiting gene pool diversity. Scientific studies on the variability and efforts to develop improved varieties are rare for the species. Thus, the current study aims to investigate the genetic variability of *Curcuma zedoaria* Rosc. in Kerala, India, in order to conserve and improve its genetic stock and also to identify superior genotypes from the germplasm.

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Chapter I

INTRODUCTION

Unavoidably, plants contribute to the growth of human cultures all over the world. The world's population relies upon conventional medicines, which mostly use plant extracts, for about 80% of their healthcare needs. Herbal remedies are believed to be safer to use than synthetic ones, which would have long-term adverse effects (Howes *et al*., 2003). Plants in a variety of forms such as vegetables, fruits, spices, snacks, bread ingredients or beverages, played a vital role in our ancestors' diets (Tardio *et al*., 2006; Ghirardini *et al*., 2007). The protection of various plant derived products that provide food, fuel, medicine and other necessities depends on genetic resource conservation.

All natural living things depend on genetic diversity to survive. Knowing more about genetic diversity will make it easier to decide what genotypes should be conserved and where. Since genetic variability quantifies the level of genetic variation in a population, it is the fundamental source of biodiversity. It builds the basis for evolution through natural selection (Fischer, 1930). For the conservation of genotypes, a good understanding of genetic diversity is necessary. For a species to obtain the ability to respond to environmental forces and to survive in nature over the long run, it must have genetic variation. Without significant diversity, the species will not be able to adapt to the changing environmental conditions (Menini, 1999). As a result, studying genetic diversity is a fundamental component of sustainable use of genetic resources (Chalmers *et al*., 1992).

Due to the continued use of specific genotypes as parents in various crop improvement programmes, unbalanced breeding practices that concentrated on the development of just a few traits and the introduction of a few superior lines

to several countries, natural crop variability has decreased over time. This has increased genetic similarity between modern crop varieties. Plant breeders use a variety of strategies to strengthen the genetic makeup of agricultural plants. They are all dependant on how much genetic variation there is among various genotypes.

Improved plant breeding techniques are very helpful in increasing the performance of several agricultural crops. Its objectives include enhancing crop production, nutritional quality and environmental adaptation. Such improvement programmes adopt appropriate breeding strategies based on the type and degree of genetic variability of the crop. The adoption of effective and efficient novel strategies will contribute in improving traditional breeding practices.

Understanding genetic diversity in quantitative parameters is crucial for the effectiveness of plant breeding practices. The statistical and biometrical tools used to analyse these characteristics aid the breeder in adopting the proper breeding techniques. These procedures involve the estimation of numerous parameters such as heritability, genotypic and phenotypic variability, genetic control, genetic variability, etc. (Singh and Sharma, 2002).

The heritability values of a trait show how that trait is passed down from parent to offspring. In breeding programmes, choosing traits with greater heritability values results in superior, novel varieties. More additive genes accumulate when characters with higher heritability and genetic advance are chosen. It can enhance the possibilities for further improvement of their performance (Pandey and Dobhal, 1993; Stephens *et al*., 2012).

Zingiberaceae, the largest monocotyledonous family in India, comprises of about 1200 species and 53 genera (Jain and Prakash, 1995; Kress, 1990; Kress *et al*., 2002). Several aromatic species from the family are exploited in

herbal medicines for a variety of ailments (Alviano and Alviano, 2009). As an important genus of the family with a long history of traditional medicinal applications, *Curcuma* is significant commercially. It has been used to cure a number of illnesses including diabetes, enlarged liver, cough, skin conditions, chest pain and rheumatism (Saikia and Borthakumar, 2010). It is being used as a spice, colourant, food preservative and flavouring agent (Xiang *et al*., 2011; Sahdeo and Bharat, 2011). The genus is a rich source of vitamins, minerals, starch, proteins, carbohydrates and lipids (Yadav *et al*., 2017). The genus is said to have anti-insect, anti-cancer, anti-microbial, anti-viral, anti-inflammatory, anti-diabetic, anti-cholerectic, anti-hypocholesteremic and anti-rheumatic properties (Sasikumar, 2005).

Curcuma zedoaria Rosc., also referred to as white turmeric or zedoary is an important member of the genus *Curcuma*. It is a long-used spice related to ginger and turmeric. From an economic perspective, the underground rhizome is the most significant part of the plant. The rhizome is fragrant when crushed and has a creamy white inner portion. In Ayurveda and other folk and tribal systems of medicine, different components of *C. zedoaria* are utilised (Saikia and Nath, 2003). There are analgesic, antiulcer, antiasthmatic, antiarthritic, diuretic and antipyretic effects reported for the plant. The species is used in the preparation of several herbal medicines that have anti-inflammatory, antihepatotoxic, neuroprotective, antimicrobial, and antimutagenic properties. It has cytotoxic effects on human ovarian cancer cells and is effective against *Salmonella*/microsomal system mutations induced by benzopyrene (Syu *et al*., 1998).

Zedoary rhizome is used to cure food sickness. Small rhizome pieces are employed as an antidote and given orally (Bantawa and Rai, 2009). Additionally, it relieves rheumatism, amenorrhea, cough, colic, tonsillitis, abdominal discomfort, menstrual pain, vomiting, abdominal cramps and

dyspepsia (Dhal *et al*., 2011). Zedoary protein shows strong anticancer properties (Muthukumar *et al*., 2012).

Many anthropogenic activities including habitat degradation, urbanization, industrialization, etc. contribute to the decline of the population size of this important medicinal plant. Scientific studies on its variability and efforts to produce improved varieties of the species are quite rare. Increased genetic diversity in the cultivated germplasm would benefit selection efforts, minimize crop vulnerability and enable the insertion of specific traits into new cultivars (Bisognin, 2011). Reasonable breeding approaches to exploit the gene pool diversity will result from understanding the degree of genetic variability in the crop. Thus, the goal of the current study is to examine genetic variability of *Curcuma zedoaria* Rosc. in Kerala State of India in order to conserve and improve its genetic stock.

Chapter II

REVIEW OF LITERATURE

2.1. Zingiberaceae

Zingiberaceae is the largest family of the plant kingdom which comprises of about 53 genera and 1300 species. In India a total of 22 genera and 178 species are reported. Members of the family are integral components of medicines, dyes, spices, food, perfumes, etc. Some members are used in folk medicine (Devi *et al*., 2014). The family is commonly called the 'ginger family'. Members of the family have tuberous or non-tuberous rhizomes with characteristic strong aromatic smell and various medicinal properties (Chen *et al*., 2008).

Zingiberaceae is mostly distributed in tropical regions of the world (Kress *et al*., 2002). According to Sirirugsa (1999) the family shows distribution mainly through tropical and subtropical regions of the world; the highest distribution patterns are observed in Indonesia, Brunei, Philippines, Malaysia, Singapore and Papua New Guinea and 52 genera and 1,500 species of Zingiberaceae are present across the world. Maximum number of genera (25) are reported from Malaysia with 650 species. The plants mostly grow in damp humid and shady areas. Some species grow at high elevation where they are fully exposed to the sun.

All the members of Zingiberaceae are aromatic herbaceous plants with underground rhizome. True aerial stem is absent but a pseudostem made up of sheathing leaf bases is prominent. Rhizome is thick with secretory cells which can secrete essential oil. Zingiberoideae and Costoideae are the two sub families under the family Zingiberaceae (Mabberley, 1997).

Humid and shady regions are best suited for their growth. Plants are rhizomatous and herbaceous with fleshy rhizome. Rhizome is branched and ends in an erect shoot. Rhizome is covered with scale leaves and has distinct segmentation. Successive branches emerge from the buds developing in the axils of the scale leaves of the shoot. Root arises from the rhizome and is thick, fleshy and slender (Tomlinson, 1956). The most studied species of the family is *Curcuma longa*, which possesses tremendous therapeutic potentials (Jain *et al*., 2007). Some epiphytic species are also present in the family (Sabu, 2006).

The inflorescence is a spike, which is terminal on a leafy shoot or lateral in position and comprises a central axis on which the primary bracts are spirally arranged. Both terminal and lateral inflorescences are reported in some members during different seasons. The spikes may be condensed or elongated and are either white or variously coloured. The spike bears comma bracts at its tip, which are sterile in nature. Fertile bracts bear either a single flower or multiflowered cincinni. Flowers are usually bisexual in nature and are showy and zygomorphic. The trimerous flowers carry 3 lobes each for the calyx and corolla. Sepals fuse together to form the calyx tube and petals fuse to form the corolla tube. Dorsal lobe of the corolla is longer and broader than lateral lobes. Out of the 6 stamens arranged in two whorls, only one at the posterior end of the inner whorl becomes functional. Labellum, the characteristic feature of the family is formed by the fusion of two lateral sterile staminoids in the inner androecium whorl. It is differentially coloured and is with or without stripes. Ovary is inferior with parietal or axile placentation. The bilipped swollen stigma bears characteristic fringed hairs on it. Capsule or berry is the characteristic fruit of the family. Capsule dehisces by loculicidal method (Sirirugsa, 1999; Holttum, 1950; Sabu, 2006; Sabu *et al*., 2011; Jayasree and Sabu, 2013). According to Hartanto and Sofiyanti (2014) each genus and species of the family has its own characteristic inflorescence with distinctive shape and colour.

According to Barbosa *et al*. (2017) Zingiberaceae members are commonly known as gingers and have wide applications in medicinal and culinary purposes since time immemorial. Rhizome is the commonly used part in traditional medicinal practices and in food flavouring industry (Ammon and Wahl, 1991; Ivanovic *et al*., 2021). Spices like turmeric, cardamom and ginger are from this family, and some of them have uses as ethnomedicines and in advanced systems of medicine (Kumar *et al*., 2013). Many members are also reported to have anti-inflammatory and antioxidant activities (Masuda *et al*., 1992; Masuda *et al*., 1993). According to Alviano and Alviano (2009) members of the family are the essential components of various industries like flavouring, fragrance, perfumery, etc.

2.2. Medicinally important genera

2.2.1. *Alpinia* Roxb.

Alpinia is the largest genus of Zingiberaceae and is mainly distributed in the Indo-pacific region. About 230 species have been reported from the genus in total (Kress *et al*., 2005; Sabu, 2006). Many members are important constituents in the treatment of obesity, cephalagia, arthritis, cough, fever, etc. (Warrier *et al*., 1995). *A. galanga* is the active component of the ayurvedic preparation 'rasnadi powder' (Prabhukumar *et al*., 2013a). In Asia, these are variously used for cooking purposes (*A. galanga*) and as medicine (*A. officinarum*) (Wu and Larsen, 2000).

2.2.2. *Amomum* Roxb.

The genus contains about 150 species and was formerly known as Scitaminae. It is the second largest genus under the family Zingiberaceae (Thomas *et al*., 2009). The plants are mainly cultivated in the Central Himalayan and North Eastern regions of India (Sharma *et al*., 2000). The genus

shows a higher distribution in Southeast Asia from the Himalayas to Northern Australia and extends into the Central Pacific (Smith, 1985).

Floral characteristics of the genus include the presence of white labellum with a central yellowish patch. In some species a characteristic red marking is seen on the labellum. The anthers carry a trilobular crest which may be fan shaped (Saensouk and Saensouik, 2021).

Amomum subulatum Roxb. is commonly known as large cardamom and is a well-known spice. It has a characteristic aromatic smell making them useful as flavouring agent in food industry (Bisht *et al*., 2011). It provides flavour to various vegetable and meat preparations. Fruit powder is also used as essential ingredient in spice masala mixtures (Sharma *et al*., 2007). Stomachic, stimulant, alexipharmic and astringent properties of the plant are reported. It is prescribed for the treatment of abdominal pains, biliousness, indigestion, vomiting and rectal diseases (Nadkarni, 1976).

2.2.3. *Kaempferia* L.

About 70 species of *Kaempferia* are recorded which are mainly distributed in Asia and Africa (Kam, 1980). Rhizome alcoholic infusion of the species *Kaempferia parviflora* is used to treat gastric problems, body pain, allergy, fungal infections, etc. The same is used as a tonic to treat impotency in males (Yenjai *et al*., 2004; Pengcharoen, 2002). Prabhukumar *et al*. (2013b) have reported the occurrence of three species of the genus in South India namely *K. galanga*, *K. rotunda* and one ornamental species *K. elegans*.

2.2.4. *Zingiber* Boehm.

The generic name *Zingiber* was derived from the Tamil word “ingiver” which means ginger rhizome (Sabu, 2006). The genus comprises of 141 species and are distributed throughout tropical Asia. The centre of diversity of

the genus is South East Asia (Theilade, 1999). Rhizomes are used to cure ulcers, sore throat, cough, common cold, rheumatism, cholesterol, etc. (Shukla and Singh, 2007). The widely used species of the family is *Zingiber officinale*, having the common name 'ginger'. It has uses as medicine and flavouring agent (Sabulal *et al*., 2006; Sivasothy *et al*., 2011).

2.2.5. *Elettaria* Maton.

Elettaria cardamomum L. is the important in terms of commercial importance. It is known as "Queen of Spices" in India. It grows wild and is cultivated mainly in India and Sri Lanka. Cardamom is an important economic crop and is a highly valued spice (Parthasarathy *et al*., 2008; Ashokkumar *et al*., 2020). The essential oils of cardamom were proved to have therapeutic benefits, antifungal properties and analgesic and anti-inflammatory properties (Nirmala 2000; Choudhary *et al*., 2000; Al-Zuhair *et al*., 1996). The major constituents of cardamom essential oil were identified as 1,8-cineol, α-terpinyl acetate, terpinene and fenchyl alcohol (Abbasipour *et al*., 2011.)

2.2.6. *Curcuma* Linn.

About 70 species of *Curcuma* are distributed all over the world among which 20 species belonging to China (Xia *et al.*, 2005). The genus is well represented in the Indian subcontinent where 31 species have been reported to occur (Velayudhan, 1996). The name *Curcuma* is derived from the Arabic word 'kurkum', meaning 'yellow' which refers to the colour of the rhizome. *Curcuma* species are mainly distributed in the tropics and subtropics of South and Southeast Asia and extensively cultivated in Bengal, China, Taiwan, Sri Lanka, Indonesia, Peru, Australia and West Indies (Ravindran *et al*., 2016). It is reported that the genus grows well in India, China, Indonesia, Bangladesh, Burma, Pakistan, Sri Lanka, etc. The production of *Curcuma* is the maximum in India (Anonymous, 2006).

According to Farrel (1990) *Curcuma* is used from ancient times as a medicine, dye and a ceremonial colour. Turmeric dye in combination with various alkalies is used as a colouring agent in textile industry. Rhizome part of the plants has a pungent smell and is bitter in taste. It is also used as a laxative in textile industry (Kritikar *et al*., 1987).

Yellow powder of *C. longa* rhizome is important as a colouring agent and condiment in food industry. The species along with other species like *C. kwangsiensis*, *C. wenyujin*, *C. phaeocaulis*, etc. have importance in removing blood stasis, relieving pain and activating vital energy. The most investigated member of the genus is *C. longa* along with *C. zedoaria* and *C. amada*. More than 700 compounds have been isolated from the genus, among which curcumin is widely studied. It is mainly extracted from *C. longa* (Ryu *et al*., 2006; Agrawal and Mishra, 2010; Maheshwari *et al*., 2006; Heger *et al*., 2014; Hamaguchi *et al*., 2010).

C. longa, *C. zanthorrhiza*, *C. amada*, *C. aromatica* and *C. zedoaria* are some medicinally important members of the genus used in Ayurveda, Naturopathy, Siddha, Unani and Homeopathy (Asolkar *et al*., 1992). *Curcuma* forms the integral constituent in traditional Chinese medicine to alleviate pain and to remove blood stasis (Xia *et al.*, 2005).

Starch is extracted from different species of *Curcuma*. The genus can be used as an alternative source of starch on a commercial scale (Policegoudra and Aradhya, 2008; Sasikumar and Sajitha, 2015).

Zhang *et al*. (2018) studied the distribution and diversity of twelve species of *Curcuma* in China. China is one of the diversity centres of the genus *Curcuma*, and is therefore one of the abundant sources of *Curcuma* germplasm over the world. Rhizome part of *Curcuma* is the most widely used traditional aroma enhancer and natural dye.

The genus *Curcuma* is well known for its use as favouring and colouring agents, spices, medicinal components and ornamentals in India, Bangladesh, Malaysia etc. Members of the genus are used to treat dysentery, leukorrhea, insect bite and pneumonia (Akarchariya *et al*., 2017; Basak *et al*., 2010). It is having wide applications in the ethnomedicinal system for centuries. The compounds which make them useful in ethnomedicinal systems count about 427. Phenolic compounds, organic acids, inorganic compounds, tannins, flavonoids, oils and anthocyanins are richly present in the genus with curcumin as the key component. Presence of curcumin enhances its property to heal jaundice, liver diseases, wounds, stomach ache, etc. (Ayati *et al*., 2019). *Curcuma* oil and its terpenoids have been shown to inhibit the growth of ovarian cancer, endometrial cancer and cervical cancer cells. It also reduces tumour formation, enhances the efficiency of chemotherapy drugs, and improves patients' quality of life (Zhang *et al*., 2023). Members of the family are widely used to treat respiratory, digestive and skin diseases caused by microbial infections (Irayanti and Putra, 2020).

Various workers have performed the phytochemical screening of the genus. The major curcuminoids isolated include curcumin, bisdemethoxycurcumin and demethoxycurcumin. *Curcuma* oil contains monoterpenoids and sesquiterpenoids as active compounds (Singh *et al*., 2010; Xiang *et al*., 2018). Chemical composition of the genus is strongly affected by factors like genotype, seasonal variations, fertilizer application, stress, harvesting time, extraction, etc. (Sandeep *et al*., 2015).

The antimicrobial, insecticidal, antiproliferative, anti-inflammatory, hypocholesterolemic and antidiarrheal properties of the genus have been studied by different workers (Jagtap, 2015; Oon *et al*., 2015; Sikha and Harini, 2015; Fouad and Da Camara, 2017 and Shafreen *et al*., 2018). Curcumin can

scavenge free radicals like ROS and RNS. It also inhibits the expression of HIV-1 LTR- directed genes (Balasubramanyam *et al*., 2004).

Kim *et al*. (2003) and Chowdhury *et al*. (2008) have studied the antifungal activities of the genus *Curcuma*. *C. longa* can inhibit the growth of *Candida albicans*, *Cryptococcus neoformans*, *Phytophthora infestans* and *Rhizoctonia solani*. The analgesic and anthelmintic actions of the genus have been also studied (Randeep *et al*., 2011; Baghel *et al*., 2013).

The above account on the family Zingiberaceae in general and the genus *Curcuma* in particular shows the potential importance of the family and the genus as great contributors to medicinal, culinary and cosmetic industry. From among the different species of the genus *Curcuma*, *Curcuma zedoaria* Rosc. (white turmeric) has been selected for the present study and hence a detailed review of the literature on the species is presented below.

2.3. *Curcuma zedoaria* Rosc.

C. zedoaria is an important species of the genus *Curcuma* commonly known as white turmeric. The plant was introduced by Arabs to Europe during the 6th century. In English, the plant is known as round zedoary. In India it is known by its several vernacular names, *krachura* (Sanskrit), *gandamatsi* (Hindi) and *sutha* (Bengali) (Nadkarni, 1999).

The plant has an upright pseudostem and an underground rhizome portion. The leafy shoot attains a maximum height of 1 m. Rhizome has a creamy white inner portion. It carries apical buds, which emerge above the ground as inflorescence. Inflorescence contains yellow flowers with attractive bracts in red or green colour (Bhutia and Sharangi, 2017).

The rhizome is used for the preparation of ‘Abir’, the dye used by Hindus during Holi festival. Rhizome powder is prepared and purified by

repeated washing. The purified powder is dried out and mixed with a decoction of sappan wood (*Cesalpinia sappan*) to get red colour. The stimulative and carminative effects of zedoary have made it useful in traditional medicinal practices. Cosmetic properties of zedoary have also been reported (Ravindran *et al*., 2007).

Curcuma zedoaria contains antioxidant and anti-inflammatory curcuminoids similar to that of *Curcuma longa* (Paramapojn and Gritsanapan, 2009). Phytochemical analysis for antioxidant, anti-inflammatory and inhibitory actions on human cell proliferation was done. The health benefits of the rhizome part of *Curcuma zedoaria* along with two other *Curcuma* species, *Curcuma caesia* and *Curcuma aeruginosa* using *in vitro* antioxidant, antiinflammatory and human tumour cell proliferation inhibitory activities have been studied. Drops of zedoary leaves are given to children to control worms. Root powder is given to infants as a substitute of some foreign foods (Nadkarni, 1982). It is cultivated as an ornamental plant in home yards and gardens (Alexander, 2007).

Various parts of the plant have uses in the ethnomedicinal system. Rhizome is dried and powdered to treat allergic diseases. Rhizome oil is applied for stomach pain, vomiting and menstrual pain. Leaf is ground to a paste and used as plasters to protect injuries. Leaf juice of zedoary acts against worms in children (Kirtikar and Basu, 1935; Nadkarni, 1999; Joy *et al*., 1998; Prajapati and Kumar, 2003; Khare, 2007).

2.3.1. Origin and distribution

C. zedoaria flourishes well in the tropical and subtropical regions of the world. It is an aromatic herbaceous plant indigenous to India, Sri Lanka, Bangladesh and Indonesia (Nadkarni, 1996; Manfield *et al*., 2005). The plant, also named as round zedoary or zerumat is mostly found in India and S.E. Asia.

It grows wild in Eastern Himalayas and in moist deciduous forests of coastal tract of Karnataka and Kerala (Thakur *et al*., 1989).

According to Tiphthara *et al*., 2007 and Islam *et al*., 2005, zedoary grows principally in the tropical regions of the world. A hot and moist climate is suitable for its growth. It is not adapted to low temperature regions. The highest distribution is seen in the East Asian countries like Bangladesh, Malaysia, China, India, Indonesia, Vietnam and Japan. Optimal soil type for growth of the species is alluvial or well drained loose loamy soil.

Paisooksantivatana *et al*., (2001) have reported the wide distribution of *C. zedoaria* in Bangladesh and according to him the species is also distributed towards its south to north eastern part on the slopes of hilly areas and is scattered throughout the gangetic floodplain and pleistocene plateau lands.

2.3.2. Morphology

C. zedoaria is a perennial plant, which is herbaceous in nature. The plant has a pseudostem that emerges above the soil and grows upright. Rhizome is the underground part and it possesses fleshy roots. Round or ovoid tuber like roots known as T- roots are also present which function as terminal storage structures. There is an ovate underground corm from which the axillary buds arise. These buds along with the apical buds emerging from the 3rd order rhizome grow as inflorescence above the ground during March to April. This forms the basal flower spike that reaches up to 30 cm in length. A vegetative shoot develops from a node nearer to the spike. New branches develop from the corms of successive aerial shoots (Maciel and Criley, 2002). The plant gets propagated only through rhizomes. Rhizome is characterized by its bitter taste and the camphoraceous smell (De Padua *et al*., 1999). Rhizome has a creamy white inner portion. According to Jyothi *et al*. (2003) yellowish white colour of the rhizome is due to the presence of curcumin.

Rhizome gives rise to apical buds, which emerge above the ground as inflorescence. Inflorescence contains yellow flowers with attractive bracts in red or green colour (Bhutia and Sharangi, 2017; Rahman *et al*., 2014).

According to Maciel and Criley (2002), the leafy shoot reaches up to 1 m and carries 5 to 8 narrowly ovate or oblong leaves on it. Sheathing leaf bases are present which form a pseudostem.

The plant usually attains a height of 3 feet. Large number of tubers which are palmately branched are seen in the root stock. Leaves are oblong and reach up to 1-2 ft in length. They are acuminate with long petioles. Oval shaped bracts form the attractive part of the inflorescence. The size reaches up to 1.5 inches in length. They are coloured green with red tinges. The yellow-coloured flowers are 3 lobed with a suborbicular lip. It measures 5 inches in width (Fischer, 1928).

2.3.3. Agriculture

Studies on agrotechniques for the cultivation of *C. zedoaria* have been carried out by Joy *et al*. (2002) at Aromatic and Medicinal Plants Research Station, Odakkali, Kerala, India during 1996-1999. The soil used for the study had a lower concentration of Nitrogen, medium concentration of Potassium and higher concentration of Phosphorus. pH of the soil was 5.5. The experiment was performed in randomized block design (RBD) with 3 replications. 20 different treatments were provided using 4 spacing patterns and 5 manurial treatments. The spacing patterns adopted for the study were 20 cm x 20 cm, 30 cm x 20 cm, 30 cm x 30 cm and 40 cm x 40 cm. Five different manorial treatments were the control, FYM 20 t/ha, N, P2O5 and K2 at 100: 50: 50 kg/ha, green manuring in-situ (sowing cowpea at 25 kg/ha, uprooting at flowering and using as mulch 30 days after sowing) and biofertilizer (*Azospirillum*) at 10 kg/ha. When 1.5 t/ha of seed rhizomes were planted at 60 cm x 40 cm spacing,

the maximum yield of rhizome (34 t/ha), oleoresin (5%) and essential oil (0.33%) were observed. Yield was the maximum by the application of 20 t/ha of FYM, 100 kg of K, 50 kg of P and 50 kg of K per ha in association with biofertilizers, green manuring and mulching.

The bacterial strain *Ralstonia solanacearum* was identified to cause wilt disease in three species of *Curcuma*, *C. zedoaria*, *C. aromatica* and *C. longa*. The affected plants show yellowish, curled and wilted leaves with their rhizome having a creamy-white bacterium in the inner portion (Ajitomi *et al*., 2015).

2.3.4. Taxonomic hierarchy of *Curcuma zedoaria* Rosc.

According to Angiosperm Phylogeny Group (2009), taxonomic hierarchy of *Curcuma zedoaria* is as follows:

Kingdom- Plantae, Clade- Angiosperm, Clade- Monocota, Clade- Commelinids, Order- Zingiberales, Family- Zingiberaceae.

2.3.5. Cytology

The chromosome numbers and ploidy levels are having immense importance in the field of genetics. The genus *Curcuma* possess smaller chromosomes to adopt to its tropical and subtropical habitats (Islam *et al*., 2007). The basic chromosome number of the genus *Curcuma* was suggested as x = 21 which originated by dibasic amphidiploidy from x = 9 and x = 21 by secondary polyploidy (Ramachandran, 1961; Ramachandran, 1969).

Curcuma zedoaria karyotype was prepared and analyzed by Lijuan *et al*. (2011) by CPD staining and FISH techniques using 45s rDNA. Metaphase chromosomes were analyzed and the karyotype was prepared on the basis of chromosome length, position of centromere and the occurrence of nuclear organizer region (NOR). Analysis of karyotype using CDP staining and fluorescence methods has shown the presence of 5 pairs of 45s rDNA signals

on different chromosomes. The investigation has revealed *C. zedoaria* as an aneuploid with the karyotype 2n = 62+1 = 40m+12 sm+1m.

2.3.6. Anatomical and histochemical studies

C. zedoaria Roscoe root segments were histologically analysed for studying the nature of callogenesis and organogenesis by Mello *et al*. (2001). Root segments collected from *in vitro* grown plants of *Curcuma zedoaria* were used as explants for callus generation. Murashige and Skoog agar medium supplemented with 2.2 mM of 6-benzylaminopurine and 13.4 mM of a-naphthalene acetic acid was used for the culturing of explants. Numerous lateral roots appeared from explant pericycle after 20 days of culture. Rupturing of epidermis and accumulation of starch in some outer cortical parenchyma cells were observed after 30 days of culture.

Different species of *Curcuma* show almost similar anatomical characters, but slight variations in the number and shape of curcumin cells, number and arrangement of primary and secondary bundles, orientation of tissues and the number and arrangement of starch grains and oil cells. The primary vascular bundles are scattered and abundant in both outer and inner zones in *C. amada* and *C. longa*, less in *C. zedoaria* and the least in *C. aromatica*. The number of curcumin cells was higher for *C. longa* and *C. zedoaria* and minimum in *C. amada* and *C. aromatica*. Starch grains were abundant and larger in *C. aromatica* and *C. zedoaria*. Oil cells and curcumin cells were higher in the apical and nodal regions of different species than in the internodal region. *C. longa* possesses a continuous ring of endodermis along with the pericycle; *C. amada* has more or less circular endodermis and *C. aromatica* and *C. zedoaria* have wavy and discontinuous endodermal layer (Sherlija *et al*., 1998; Remashree and Balachandran, 2006).

2.3.7. Pharmacognosy

Srivastava *et al*. (2011) has worked on the pharmacognostic analysis of *C. zedoaria* rhizome. Moisture content was found to be 83.22% by the analysis. Percentage of total ash was 6.64 and that of acid insoluble ash was 0.64. Percentage of sugar and starch were 12.51 and 15.70 respectively. Water soluble extractives were present in a higher percentage (18.96) than alcohol soluble extractives (15.53). Total volatile oil was determined as 2.8%.

2.3.8. Phytochemistry

The rhizome of *C. zedoaria* contains different terpenoids- curcumene, curcumenone, curdione, curcumenol, curzerenone epoxide, a volatile oil (1.0-1.5%) resembling ginger oil and starch (50%). In the traditional medicinal systems, the rhizome is used for the treatment of goitre. The antitumor, antimicrobial and the antiallergic properties of zedoary has also been elucidated (Srividya *et al.*, 2012).

The composition of the essential oil of *C. zedoaria* along with 7 other species of *Curcuma* was investigated by Angel *et al*. (2014). The highest concentration of essential oil was reported in *C. zedoaria* with a yield of 1.4 ml/100 g fw. The compounds curzerene and epicurzerenone were uniquely reported in zedoary by GC-MS analysis. Camphor, camphene, 1,8-cineole, α-pinene and β-pinene were identified in most of the studied species.

Chen *et al*. (2013) have determined various compounds in *C. zedoaria* using mass spectroscopy and gas chromatography. The compounds were identified as eucalyptol, 8,9-dehydro-9-formyl-cycloisolongifolene, 6-methyl-4,5,6,7-tetrahydro-3,6-dimethyl-5-isopropenyl-trans-benzofuran and γ- elemene.

The phytochemical effect of *C. zedoaria* on hydrogen peroxide induced oxidative stress in mouse neuroblastoma-rat glioma hybridoma cells NG108-15 was studied by Hamdi *et al*. (2015). A total of nine sesquiterpenes, germacrone, dehydrocurdione, curcumenol, isoprocurcumenol, curcumenone, procurcumenol, zerumbone epoxide, zederone and gweicurculactone along with one labdane diterpene, zerumin A were isolated from *C. zedoaria*. 100% protection of NG108-15 cells was shown by curcumenol and dehydrocurdione at the concentrations of 4 μM and 10 μM respectively.

Large number of sesquiterpenoids were isolated from the rhizome essential oil of zedoary. Different compounds isolated were curdione, dehydrocurdione, germacrone-7,8-epoxide, 13-hydroxygermacrone, zederone, furanodienone, furanodiene, furanogermanone and isofuranodienone (Hikino *et al*., 1972; Hikino *et al*., 1967; Hikino *et al*., 1970; Hikino *et al*., 1975; Shibuya *et al*., 1987). Zedoary root portion contained ethyl p-methoxy cinnamate which was proved to have antifungal property (Tang and Eisenbrand, 1992; Hong *et al*., 2001).

Using the simplex lattice design Monton *et al*. (2021) have optimized the mass ratio of the rhizomes of three species of *Curcuma* for getting the highest curcuminoid content. *C. zedoaria, C. longa* and *C. aromatica* were the selected species for the purpose. High performance liquid chromatography was used to determine the individual curcuminoids (curcumin, demethoxycurcumin and bis-demethoxycurcumin) and total curcuminoid content in all the three species. The highest curcumin content was provided by a mixture of *C. longa* and *C. aromatica* in the mass ratio of 72:28%. Bis-demethoxycurcumin and demethoxycurcumin were reported in a higher concentration from *Curcuma longa* than in *C. zedoaria* and *C. aromatica*.

C. zedoaria essential oil was analyzed by GC-MS and the chemical composition has revealed a higher composition of epicurzerenone and curdione

at the percentage of 46.6 and 13.7 respectively. Antimicrobial property of the essential oil was evaluated against the bacterial strains *Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Salmonella typhimurium, Vibrio parahaemolyticus* and *Bacillus cereus*. Essential oil from zedoary inhibited the growth of *V. parahaemolyticus* while *E. coli* strain was resistant (Lai *et al*., 2004).

Thirty-six compounds including 6 ketones, 17 terpenes and 13 alcohols were identified from the rhizome essential oil of *C. zedoaria*. Epicurzerenone was prominent (24.1%) among all the identified compounds followed by curzerene (10.4%). 5- isopropylidene-3,8-dimethyl-1(5H)-azulenone from the fraction 4 of essential oil exhibited higher antioxidant potential (Mau *et al*., 2003).

Rahman *et al.* (2014) have worked on the antioxidant properties of leaf extract and essential oil from *C. zedoaria*. Essential oil extracted from the dried leaves by hydrodistillation had a yellowish colour. It was subjected to GC-MS analysis. 24 different compounds were identified which were all evaluated for their antioxidant property using two different methods, inhibition of the free radical 1,1-diphenyl-2-picrylhydrazyl (DPPH) and the super oxide radical scavenging activity. By DPPH method, a higher activity was shown by the ethyl acetate extract with an IC50 value of 17.56±1.6 µg/ml. Ethyl acetate extract exhibited superior activity over all the other extracts in the superoxide scavenging activity assay (IC50: 23.47±1.2 mg/ml).

Retnowati *et al*. (2014) have investigated the composition of zedoary essential oil. The essential oil was obtained by steam distillation method. A total of 20 compounds were fractioned from the oil using GC-MS method. Major compounds isolated from the essential oil were identified as camphor (49.51 %), isobornyl alcohol (12.66 %), borneol (4.23 %), furanodiene (3.61 %) and furanodienone (3.49 %).

Shiobara *et al*. (1985; 1986) have reported the extraction of 3 novel sesquiterpenoids: 13-hydroxygermacrone, a furanocardinane 'curzeone', and a furanoguaiane 'zedoarol' and two new spirolactones - curcumanolide A and curcumanolide B along with the new cyclopropanosesquiterpene curcumenone from zedoary. Germacrone-4,5-epoxide, an intermediate in the biogenesis of germacrone type sesquiterpenoids was also reported from the young shoots of *C. zedoaria*.

Garg *et al*. (2005) extracted the essential oil from zedoary leaves and analysed for its compound composition and concentrations. The essential oil contained 38% of sesquiterpene hydrocarbons, 26% of oxygenated monoterpenes, 13.5% of oxygenated sesquiterpenes and 2.3% of monoterpene hydrocarbons. Dehydrocurdione, Selina-4(15), 7(11)-dien-8-one, α- terpinyl acetate and isoborneol were the major constituents of leaf essential oil.

Rhizome ethanolic extract of the species collected from various locations in Thailand were subjected to HPLC analysis by Paramapojn and Gritsanapan (2007). The result showed that zedoary rhizome extract contained dimethoxycurcumin (3015±0.15 to 10.98±0.28 w/w), curcumin (1.46±0.45 to 5.73± 0.11w/w) and bisdemethoxycurcumin (0.49±0.02 to 2.99±0.20 w/w). Curcuminoid content of the extract showed the highest value of 16.83±0.62 w/w and the lowest value of 6.09±1.79 w/w.

Chen *et al*. (2016) isolated 2 sesquiterpenoids from the rhizomes of zedoary. The isolated compounds were identified as 1-oxocurzerenone and 13-hydroxycurzerenone. These compounds along with 8 other compounds procurcumenol, ermanin, curcumin, curcolone, germacrone, curzerenone, stigmast-4,22-dien-3,6-dione and stigmast-4-en-3,6-dione were effective against collagen induced platelet aggregation.

Using gas chromatographic mass spectroscopic analysis, the composition of oleoresin extracted in ethanol, isopropanol and ethyl acetate essential oil of zedoary rhizome was determined by Singh *et al.* (2013). Major components of the volatile oil were identified as curzerenone and germacrone with 31.6% and 10.8% concentrations. 40 components were identified from the ethyl acetate oleoresin, 26 components from the ethanol extract and 25 components from the isopropanol extract. The major components identified were curcumenol, germacrone, camphor and curzerenone.

According to Melo *et al*. (2017) the essential oil of *C. zedoria* has potential herbicidal activity. The rhizome essential oil was analysed by gas chromatographic-mass spectroscopic methods to reveal its chemical composition. Oxygenated sesquiterpenes comprised 47.54% of essential oil, 21.95% of sesquiterpene hydrocarbons, 25.88% of oxygenated monoterpenes and 4.63% of monoterpene hydrocarbons.

Ethanolic extract of the rhizome was analysed and the components were isolated by Azam *et al*. (2017). The components isolated were identified as alkaloids, terpenoids, flavonoids, carbohydrates, steroids, tannins and saponins.

Qualitative estimation of rhizome ethanolic extract of 6 species of *Curcuma* was performed by Dutta (2015). Different species used in the experiment were *C. zedoaria*, *C. angustifolia*, *C. longa*, *C. leucorrhiza*, *C. amada* and *C. caesia*. Phytochemical screening revealed the presence of alkaloids, flavonoids, phenols, terpenoids, saponins and tannins in the species. Curcumin content of the extract was analysed quantitatively. The highest curcumin content was recorded in *C. longa* (125 mg/100g) followed by *C. angustifolia* (71 mg/100g). The lowest curcumin content was reported in *C. caesia* (8 mg/100g), *C. amada* (11 mg/100g) and *C. leucorrhiza* (15 mg/100g).

Purkayastha *et al*. (2006) analysed zedoary rhizome essential oil from Northeast India and 37 components were reported. Major components isolated included curzerenone by 22.3%, 1-8-cineole by 15.9% and germacrone by 9.0%. Azulenone, an active component of *C. zedoaria* essential oil which plays an important role in antioxidant activity of the plant was also isolated (Zeeshan *et al*., 2018).

The total phenolic content of leaves and rhizomes of zedoary was tested by Samarasinghe *et al*. (2021). The leaf extract exhibited 2.164±0.001 g of GA equivalents/1 g dried powder. Phenolic content of the rhizome was slightly lower than that of leaves, i.e., 1.687±0.001 g of GA equivalents/1g dried powder.

Nine major compounds were isolated from the methanolic extract of zedoary. Compounds were identified as cucurbitacin 1 glycopyrrolate, 24-dijydroxyvitamin D3, 26,26,26,27,27,27-hexafluoro1-alphamethyl gamboginate, ibuprofen, protoporphyrinogen IX, flurandrenolide, phenylbutazone glucuronide and propofol (Jadhao and Bhuktar, 2019).

2.3.9. New compounds

Eun *et al*. (2010) discovered a new sesquiterpenoid, curcuzederone from zedoary. NMR spectroscopic analysis revealed the compound as $C_{15}H_{20}O_5$. The compound shows close relation with zederone, a known compound reported from zedoary. Along with the novel sesquiterpenoid, a known flavonoid ‘naringenin’ was also reported from the plant for the first time.

A new compound ‘zedoaroxane A’ was isolated from zedoary rhizome by Van *et al*. (2021). The compound was identified as a diphenyl heptanoid and its structure was elucidated using NMR spectroscopy. It was also proved to have inhibitory action against α-glucosidase with an IC50 value ranging from 35.2 to 89.0 μM.

Hsiao *et al*. (2020) isolated 13-hydroxy-1-oxocurzerenone, a new sesquiterpenoid derivative from zedoary. The cytotoxic activity of the isolated compound was proved with an IC50 value of 3.48μM against the DLD-1 cell line. An inhibitory action on LPS-induced NO generation (IC50- 4.63 μM) was also observed for the compound. Six other known compounds, 1-oxocurzerenone, curcolone, curcumenone, procurcumenol, curcumenol and zerumin A were also isolated from the plant.

2.3.10. Food value

Both processed and unprocessed rhizomes of zedoary are consumed as food. The processed rhizomes yield 'shoti', the starch produced from it. It is given as a food for infants. In case of extreme food scarcity, the unprocessed rhizome parts were also consumed in some areas of Bangladesh. Protein is removed from shoti during its preparation. Rhizomes are collected, washed, minced and then soaked for hours. The white residue alone is collected. It is then washed several times to remove unwanted particles, the camphoraceous smell and its bitter taste. After the process of repeated washing, the collected residue is dried and ground to the powder, "shoti" (Latif *et al*., 1979).

2.3.11. Starch content

Curcuma zedoaria was tested for starch content in order to use it as starch raw material in food industry by Leonel *et al*. (2003). Starch content was the same as that of potato tubers. Amylose content of 21% was observed in *C. zedoaria*. The final viscosity and the pasting temperature of the starch was different from that of commercially used natural starches. *C. zedoaria* exhibited the final viscosity of 427 RVU and pasting temperature of 78ºC when compared with *C. longa* (740 RUV and 81ºC respectively).

Starch from *C. zedoaria* was compared with the starch of *C. malabaricum* by Jyothi *et al*. (2003). X-ray diffraction method has shown that,

starch of both the species have 'B' type diffraction pattern. The presence of yellow pigment curcumin is the reason for starch of *C. zedoaria* to appear in slight yellow colour. The viscosity and swelling volume values were higher for *C. malabaricum* when compared to *C. zedoaria*.

According to Fujimoto *et al*. (1984), white turmeric starch is having a large amount of amylose content. Digestion rate was lower when treated with the enzyme glucoamylase. The starch granules are flat and thin. Linear fibrils arranged radially constitute the major part of starch in zedoary (Sterling, 1976).

2.3.12. Cosmetic value

According to Krishnamoorthy *et al*. (2010) the combined application of *C. zedoaria* and *Aloe vera* extracts has the capacity to decrease melanin synthesis in B16F10 murine melanoma cells. The extract does so by inhibiting the action of tyrosinase enzyme, which catalyses the oxidation of tyrosine to melanin. 50-150% reduction of melanin synthesis was observed at a concentration of 1-5 µl of the extract. Zedoary is also used as odoriferous agent in some skin cosmetic products (Nadkarni, 1998).

2.3.13. Medicinal value

Ayurveda and other folk medicinal systems make use of various parts of *C. zedoaria* for the treatment of cancer, flatulence, diarrhoea etc. (Lobo *et al.*, 2009). The rhizome juice is used to treat worms in children and the leaf juice for dropsy and leprosy. Zedoary is used to treat abdominal pain and rheumatic pain and to regulate blood circulation (Maeda *et al*., 1984; Ruby *et al*., 1995).

The leaves and rhizomes of zedoary are used to treat headache and helminthiasis. Administration of a mixture containing macerated leaves of the plant and limewater relieves helminthiasis. Macerated rhizomes are applied on forehead to improve eyesight and to relieve headache (Karim *et al*., 2011).

Rhizome made into a fine paste is applied against dermatitis and sprain (Rahmatullah *et al*., 2009).

Zedoary rhizome is used as an appetizer. Tonics containing zedoary extract is prescribed for ladies after childbirth. The decoction of cinnamon (*Cinnamomum verum*), long pepper (*Piper longum*), zedoary and honey is given for common cold. In ayurvedic medicines, zedoary has important role in treating high fever (Thakur *et al*., 1989).

The dried tubers of *C. zedoaria, C. aromatica, C. longa* and *C. kwangsiensis* are commonly called Radix Curcumae. The sedative, analgesic and choleretic properties of Radix Curcumae are proved. It is also used for the treatment of menstrual disorders, epilepsy and hepatitis (Tang and Eisenbrand, 1992).

The rhizome part of zedoary is used to treat food poisoning. Small pieces of the rhizome are used as an antidote and administered orally (Bantawa and Rai, 2009). It also relieves tonsillitis, abdominal pain, menstrual pain, vomiting, abdominal cramps, rheumatism, amenorrhea, cough, colic and dyspepsia (Dhal *et al*., 2011).

According to Rudrapal *et al.* (2012), the dried fruits of zedoary are used to cure headache, anaemia, cough and asthma. The same mixed with black pepper is also used to treat asthma. Rhizome is proved to be effective against dyspepsia, cough, cold, flatulence, fever, anaemia, liver cancer, coronary heart disease, pelvic inflammation and leucopoenia (Ghani, 1998).

A homogenous polysaccharide (ZWP) obtained from zedoary exhibited significant effect in wound healing in diabetic rats. ZWP when applied in combination with platelet rich plasma exosome (PRP-Exos) was more effective in wound healing. The extract upregulated the synthesis and deposition of collagen with an increased rate of angiogenesis in wound site. Zedoary

polysaccharide can be used for skin repair in diabetes (Xu et al., 2018). The long leaves of zedoary were traditionally used as plasters in the treatment of adenites and lymbangitis (Kirtikar and Basu, 1935).

Zedoary rhizome along with nutmeg, fennel and honey is made into a paste and given for the treatment of common cold. Chewing the raw rhizome relieves cough. Rhizome is recommended during parturition and is a part of medicines given after childbirth. The root has several health benefits and are used in tonics for brain and heart health (Nadkarni, 2005).

Aphrodisiac function of the plant was studied and was recommended for uterus health and to regularise menstruation by some scientists. It purifies blood and is used as a body stimulant (Lindley, 2011; Chopra *et al*., 1956). Rhizome oil is orally administered to treat menstrual hematometra, stomach ache, etc. Fresh root is given to treat leucorrhoea discharge. Dried rhizome is powdered and used to cure allergic disorders (Kayum *et al*., 2021).

Matsuda *et al*. (2004) studied the antiallergic properties of *C. zedoaria* cultivated in Thailand. Acetone extract of the rhizome was found to inhibit the release of β-hexosaminidase as a marker of antigen IgE mediated degranulation in RBL-2H3 cells. Four different types of curcuminoids curcumin, dihydrocurcumin, tetrahydrodemethoxycurcumin and tetrahydrobisdemethoxycurcumin along with two bisabolene type sesquiterpenes were isolated from Thai zedoary. The antiallergic effects of all the four curcuminoids were examined. Curcumin exhibited the highest activity against β-hexosaminidase release with an IC_{50} value of 5.3 Mm.

The analgesic activities of *C. zedoaria* rhizome from Brazil was investigated by Navarro *et al*. (2002). The strongest activity was exhibited by the pure compound curcumenol, when treated in mice to cure pain.

Gupta *et al*. (2003) investigated the anti-ulcer action of the rhizome powder of *C. zedoaria* on pyloric ligated albino rats. The root powder was administered at a dosage of 200 mg/kg and significant effect was observed in reducing the ulcer index, free acid and the pH of gastric juice. The anti-ulcer action was similar to the standard drug omeprazole given at a dosage of 30 mg/kg. Result has emphasised the usage of zedoary for the treatment of peptic ulcers. Garos used zedoary to treat stomach pain and sores (Mia *et al*., 2009).

The antiasthmatic effect of zedoary was investigated by Pathan *et al*. (2015). Petroleum ether, ethanol and water extracts of zedoary were applied at the dosages of 25, 50 and 100 mg/kg to screen the efficiency of the extract against milk induced eosinophilia. Results showed that ethanol extract at the dosage of 100 mg/kg had the highest potential over petroleum ether and water extracts. A decrease in the eosinophil count in turn reduced type I hypersensitivity in asthma.

Zedoary extract is used in oral hygienic products. The extract was tested for its cytotoxic activity by Fernandes *et al*. (2012). The parameters such as cell viability and cell growth were monitored in the cell culture after the administration of the extract. Trypan blue dye exclusion assay was employed to test the viability and growth rate in oral mucosal cells. Maintenance of a viable and progressively dividing cell line in the culture was observed. It has proven the safe use of zedoary extract in oral hygienic products.

Curcumin and refined oil were used as an anti-inflammatory and anticancer agent. It also has the capacity to boost immunity (Hou and Jin, 2005).

2.3.14. Anticancer activity

Curcuma zedoaria has been used as a traditional agent against malignant diseases (Hadisaputri *et al.*, 2015). Chemical substances in white turmeric

show a lot of activities such as anticancer, antifungal, antiamebic, larvicidic, antimicrobial, antioxidant, antiplasmodial, hypoallergenic, and analgesic. Based on the data, it was concluded that white turmeric contains chemical subtances like curcuminoid, RIP (Ribosome Inactivating Protein), isocurcumenol, demethoxycurcumin, bisdemethxycurcumin, epicurzerenone, curdione, and ethyl p-methoxycinnamate which serve to disable the development of cancer cells and inhibit their growth (Putri, 2014)

A partially purified polysaccharide extracted from *C. zedoaria* rhizome was tested for its antitumor activity by Kim *et al*. (2000). The polysaccharide fractions CZ-1 and CZ-1-III inhibited tumour growth in mice transplanted with sarcoma 180 cells. Cytotoxic action of the polysaccharide was increased with increase in its concentration.

The capacity of ethyl acetate extract of *C. zedoaria* in preventing breast cancer was studied by Lourembam *et al*. (2019). MDA-MB231 breast cancer cell line was treated with the extract and its cytotoxiciy was studied using MTT assay. The mechanistic pathway was established using western blotting, confocal microscopy and wound healing migration assay. The ethyl acetate extract exhibited cytotoxicity on MDA-MB231 cell line in a dose dependent manner ($p<0.05$). The activation of cleaved caspase 9 in the treated cells has been revealed by confocal microscopy. The regulation of cytochrome C and suppression of Bcl-2 in the cell line was confirmed by Western blotting analysis. Thus, it was proved that *C. zedoaria* inhibited cancer cells through the cascase-dependent pathway.

The anticancer action of rhizome solvent extracts of *C. zedoaria* was compared with *C. amada* by Muthukumar *et al*. (2012). Crude protein from both the species were tested for the purpose. Protein from *C. zedoaria* exhibited higher anticancer activity than the protein from *C. amada*. Also, the isopropyl

extract of *C. zedoaria* exhibited high anticancer activity compared to acetone extract of *C. amada*.

Curcumin from zedoary rhizome was studied to prove its inhibitory action on cell proliferation and promotion of apoptosis in human laryngeal cancer cells by Mou *et al*. (2017). A decrease in the expression of Bcl-2 phosphoinositide 3-kinase (PI3K) and protein kinase B (Akt) along with an increased expression of microRNA-15a (miR-15a) was observed in the curcumin treated cell line.

C. zedoaria crude protein extract showed higher anticancer activity when compared with *C. amada* extract. Anticancer activity of the methanolic extract of zedoary against gastric cancer was proved by Jung *et al*. (2018).

Jung *et al*. (2018) carried out repeated column chromatography and semi-preparative HPLC purification together to separate the bioactive components of MeOH extract of *C. zedoaria*. Cytotoxic activity of the extract and its active compounds were measured in human gastric cancer AGS cells. Western blotting analysis was employed to evaluate the expression of apoptotic proteins. The extract exhibited cytotoxic activity against the tested cell line with an IC50 value of 96.60±4.87 μg/ml. Detailed chemical evaluation of the extract revealed the presence of five sesquiterpenes including isoprocurcumenol, germacrone, curzerenone, curcumenol and curcuzedoalide. Curcuzedoalide showed the highest activity in suppressing gastric cancer in a dose-dependent manner with an IC_{50} value of 125.11±2.77 μM. Curcuzedoalide proved to inhibit AGS human gastric cancer cell viability by activating caspase-8, caspase-9, caspase-3 and PARP which lead to apoptotic cell death of gastric cancer cells.

The apoptotic effect of α-curcumene extracted from *C. zedoaria* was studied by Shin and Lee (2013). The antitumor effect was tested in human

ovarian cancer cells, the SiHa cells. α-curcumene inhibited the viability of SiHa cells by 78% after 48 hours of incubation. A noticeable increase in the number of sub diploid cells, a key feature of apoptosis was observed. Treatment with α-curcumene caused the activation of capsase in SiHa cells which ultimately leads to apoptosis.

The antiproliferative action of the rhizome fraction in different solvents (methanol, ethyl acetate, hexane and dichloromethane) was tested by Hamdi *et al*. (2015). Cytotoxic action was tested in 2 human cancer cell lines (Ca Ski and MCF-7) and the non-cancer cell line HUVEC. About 19 compounds were isolated from hexane and dichloromethane fractions of zedoary rhizome. Among these, the strongest antiproliferative activity was exhibited by curcumenone and curcumenol.

Saetung *et al*. (2005) studied the cytotoxic activity of the ethanolic extract of *C. zedoaria*. Two cancer cell lines, the prostate cancer cell line (PC3) and large cell lung carcinoma cell line (CORL-23) and a normal human cell line, the fibroblast cells (10FS) were used for the study. The plant extract exhibited significant cytotoxic activity (IC50 < 30 µg/ml) against both prostate and lung cancer cell lines with IC50 values of 6.05 (CORL-23) and 17.84 (PC3) µg/ml respectively. Negligible action was observed towards the normal human cell line 10FS (IC50 55.50 µg/ml).

Lee *et al*. (2019) worked on the cytotoxicity of methanolic extract (MeOH) of *C. zedoaria* rhizome against gastric cancer AGS cell line. Components of the extract were separated using column chromatography and semi-preparative HPLC purification method. 2 new sesquiterpenes were extracted from the MeOH extract, curcuzedoalide B and curcumenol-9,10-epoxide along with 12 other sesquiterpenes. MTT cell viability assay was employed to test the cytotoxicity of separated compounds. The strongest activity was observed for curcumenol, 4,8-dioxo-6β-methoxy-7α,11-

epoxycarabrane and zedoarofuran with their IC50 values ranging from 212-392µM.

Five sesquiterpenes germacrone, curzerenone, curcuminol, isoprocurcumenol and curcuzedoalide were identified from the MeOH extract of *C. zedoaria* rhizome and were tested for their cytotoxic activity against human AGC cell line. The strongest action was observed for the curcuzedoalide fraction in suppressing the proliferation of gastric cancer cell line. The IC50 value of the fraction was observed as 125.11±2.77µM (Jung *et al*., 2018).

Syamsir *et al*. (2017) extracted the essential oil from *C. zedoaria* rhizomes collected from Malaysia and Indonesia. GC and GC-MS analysis revealed the oil from both rhizome types as sesquiterpenic in nature with an abundance of camphor (17.56% and 19.69%). Both of them were tested for their cytotoxic activity against human breast (MCF-7 and MDA-MB 231), cervical (SiHa and HeLa S3) and lung (A549 and SK-LU-1) cancer cell lines. Higher activity was reported for the rhizome oil from Malaysian zedoary against HeLa S3, SiHa, MCF-7 and SK-LU-1 cell lines.

Zedoary root hexane fraction was tested for its cytotoxicity on the proliferation of Hep G2, SNU-1 and SiHa cell lines by Kim *et al*. (2003). Different solvent fractions of the extract were used for the analysis. The fraction H2-3 showed action against all the three cell lines with almost equal effect. The H2-3-3 and H2-3-1 fractions showed the lowest IC50 values on both SiHa and Hep G2 cell lines. The fractions were tested for their apoptotic potential using the ^{3}H thymidine incorporation assay. The H2-3-1, H2-3-3 and H2-3-5 fractions exhibited significant cell death induction at the dosages of 50 mg/ml and 100 microgram/ml. By C- NMR, H- NMR and mass spectroscopic analysis the major compounds in both the fractions (H2-3-1 and H2-3-3) were identified as (-)-α- curcumene and β- tumerone.

The cytotoxic effect of *C. zedoaria* on human and murine cancer cells was studied by Lakshmi *et al*. (2011). Spectroscopic method was employed to characterize the compound Isocurcumenol, active compound found to inhibit the proliferation of cancer cells with no toxicity to normal cells.

The effect of *C. zedoaria* crude extract on tumour progression was studied in C57Bl/6J mice, injected with B16F10 murine melanoma cells. Intraperitoneal administration of the crude extract showed significant increase in RBC count of the test organism after 15 and 60 days of the treatment (Carvalho *et al*., 2010).

The antitumor activity of a polysaccharide collected from zedoary was tested against sarcoma 180 cells by Faradilla and Iwo, 2014. The study was conducted to measure the immunomodulatory effects of the polysaccharide. Swiss Webster mice was the experimental organism which was tested for the phagocytic index from reticuloendothelial system, delayed type hypersensitivity reaction (DTH), splenocyte proliferation assay and total antibody titers after incorporating the polysaccharide. The result showed that the polysaccharide fraction stimulated splenocyte proliferation at the concentration of 10-6 to 1 mg/mL ($p < 0.01$). Carbon clearance test revealed that the polysaccharide showed a phagocytic index of 1.34 at the dosage of 300 mg/kg bw. Humoral immune response was also higher at the same dosage. Thus, the polysaccharide fraction of zedoary was proved as a potent immunomodulator at the dosage of 300 mg/kg bw.

Curcuma zedoaria water extract (WE-CZ) at the dosage of 250 and 500 mg/kg for 6 weeks reduced the metastasis of B16 melanoma cells of lungs (Seo *et al*., 2005).

The antimetastatic effect of zedoary on pulmonary metastatic B16 cells was examined by Hwang *et al*. (2005). Mice injected with B16 melanoma cells

were provided with the plant extract at different dosages after 14 days of injection. The extract at the dosage of 250mg/kg showed a significant antimetastatic effect in the test organism.

Zedoary rhizome extract potentially reduced mammary tumour volume in female albino wistar rats at the dosage of 5mg/kg. The extract was given to the rats in which 7-12 Dimethylbenz[a]anthracene (DMBA) was administered to develop tumour. Extract was continuously given for 30 days and a significant reduction in tumour volume was observed (Shaikh *et al*., 2015).

Antiproliferative action of petroleum ether extract of zedoary against human breast cancer cell line MDA-MB-231 was studied by Gao *et al*. (2014). The extract significantly reduced SDF-1, CCR7 proteins and CXCR4 mRNA at the concentration of 300 µg/mL in MDA-MB-231 cell line. This resulted in an increased expression of E-cadherin mRNA in the cell line. The extract was proved to have capacity to arrest G0/G1 phase of cell cycle, which was comparable to the action of the standard drug 'epirubicin'.

2.3.15. Antidiabetic activity

Activity of zedoary against hyperglycemia was tested by Ahad *et al*. (2009). Dried rhizomes of zedoary and dried fruits of *Eucalyptus globulus* were powdered together and tested for their combined action in 1:10 ratio. Aqueous extract, alcoholic extract and petroleum ether extracts of the powder mixture were prepared and tested. The highest hypoglycemic action was observed for the aqueous extract at 600mg/kg dosage in albino rats. The activity was comparable to the action of standard drug Glibenclamide to lower blood sugar at a dosage of 180 µg/kg.

2.3.16. Antidiarrhoeal activity

` Antidiarrhoeal action of the extract of zedoary was tested in mice treated with castor oil to induce diarrhoea by Azam *et al*. (2017). A reduction in frequency and sensitivity of diarrhoea was observed after the administration of extract.

2.3.17. Antinociceptive activity

The active terpenoids dihydrocurdione and curcumenol from mother rhizome, rugous rhizome and roots of zedoary were tested for their seasonal variations. Mother rhizome contains three times more terpenoids in autumn season than in all other seasons. The extracted terpenoids were proved to possess antinociceptive action. It was tested in mice using acetic acid induced abdominal constriction model. Terpenoids extracted during winter and autumn seasons from the mother rhizome were used for the assay. The autumn extract when applied at a dosage of 10mg/kg body weight of mice, 91.1% of abdominal constriction was noted whereas the winter extract showed 93.4% of inhibition at the same dosage (Pamplona *et al*., 2006).

2.3.18. Antilarval activity

Human lymphatic filariasis and St. Louis encephalitis are caused by the bancroftian filarial parasite. The major vector of the parasite is *Culex quinquefasciatus*. Silver nanoparticles from *C. zedoaria* essential oil were tested for their antilarval activity against *Culex quinquefasciatus*. The experiment was performed with both the insecticide susceptible and insecticide resistant strains of *Culex quinquefasciatus*. Zedoary silver nanoparticle showed toxicity towards both the strains of *Culex* after 24 hours of exposure (Sutthanont *et al*., 2019).

Curcuma zedoaria along with the herbal plant *Boesenbergia rotunda* (L.) was tested for their larvicidal, pupicidal and oviposition-deterrent potential against *Aedes aegypti* L. mosquito larvae. The larval mortality rate was observed at the time intervals of 1, 5, 10, 15, 30 and 60 minutes and 24 hours. Zedoary essential oil showed the highest toxicity towards *Aedes aegypti* larvae. Lethal concentration of the essential oil was 0.05% for the larvae and 1.22% for the pupae (Phukerd and Soonwera, 2013).

2.3.19. Cholesterol control

C. zedoaria was proved to reduce meat cholesterol in broilers. A group of 100 broilers were used for the experiment, all of which were 1 day old. Four different treatments were given to the test organisms with every treatment repeated 5 times. A single replicate was included with 5 chicks. Different treatments given were represented by R0, R1, R2 and R3. R0 was the base ration. R1, R2 and R3 represented the base ration provided with 3.5%, 4.5% and 5.5% of *C. zedoaria* meal. Statistical analysis revealed the effect of zedoary in reducing the cholesterol level of chick meat at the concentration of 4.5% (Widjastuti and Andriani, 2010).

2.3.20. Hemagglutination activity

Sangvanich *et al*. (2007) reported the hemagglutination activity of zedoary extract. The activity was measured as 1.75×10^{-3} mg/ml. The mannose binding lectin isolated from *C. zedoaria* rhizome showed hemagglutinating capacity against rabbit erythrocytes. The lectin was purified and run with SDS-PAGE. A single band with molecular mass of 13 kDa was obtained. Lectin from zedoary showed good similarity with the peptide sequence of mannose binding lectin from Orchidaceae (Tiphthara *et al*., 2007; Islam *et al*., 2005).

2.3.21. Antioxidant activity

Cho and Kim (2012) have worked on the antioxidant properties of the methanolic extract of *C. zedoaria*. Three parameters, the nitric oxide level (NO), radical scavenging activity and reducing power of the extract were measured in the experiment. The expression of cyclooxygenase-2 (cox2) and inducible nitric oxide synthase (iNOS) in lipopolysaccharide (LPS) stimulated RAW 264.7 cells were measured. The LPS induced iNOS production was strongly inhibited by the MeOH extract. Antioxidant activity of the extract was mainly due to its active components, flavonoids (16.33 mg/g), anthocyanin (30.57 mg/g), and phenolics (1.062 mg/g).

The free radical scavenging activity of dried and fresh rhizomes of *C. zedoaria* was compared by Sumathi *et al*. (2013). Methanolic extracts of both the rhizome types were prepared and tested for their radical scavenging activity. Both the types of rhizomes showed almost similar radical scavenging activity.

Presence of alkaloids in *C. zedoaria* rhizome was reported by Srividya *et al*. (2012). The total flavanol was measured as 45.57±2.38 mg/g and the total phenol was measured 34.46±1.96 mg/g of the extract. *In vitro* antioxidant activity was detected by two methods. The activity was > 1000 µg/ml in the nitric oxide method and was 930±16.35 µg/ml in the DPPH method.

Protein isolated from the rhizomes of six *Curcuma* species were tested for their antioxidant and anti-inflammatory activities (Angel *et al*., 2013). The antioxidant property of purified protein was tested using ferric reducing power and DPPH scavenging activity. Four species were observed to possess higher reducing power and DPPH scavenging action with their IC50 values of 0.70 (*C. brog*), 0.73 (*C. amada*), 0.80 (*C. caesia*) and 0.84 (*C. zedoaria*). Anti-inflammatory activity was tested in rats by injecting carrageenan in the rat paw to induce edema and the purified protein extract was given. Percentage of

inhibition was measured and the result showed that all the species exhibited significant anti-inflammatory activity with percentage of inhibition ranging from 67-75. The highest inhibition was recorded for *C. zedoaria* (77%).

The chloroform soluble fraction of *C. zedoaria* exhibited 67.97±1.06% of DPPH inhibition at the concentration of 250 µg/ml. The IC50 value was 117.08±0.71for the same fraction. A moderate antioxidant activity was expressed by the ethyl acetate soluble fraction of zedoary (Riaz *et al*., 2011).

Zedoary has its use as an emmenagogue agent as reported by Monton *et al*. (2021). Antioxidant activity of the plant helps to protect skin. Rhizome extract contains curcuminoids in abundance. Different curcuminoids include curcumin, demethoxycurcumin and bis-demethoxycurcumin. Curcumin is the most abundant curcuminoid with significant antioxidant activity.

2.3.22. Antiinflammatory activity

The active constituents of *Curcuma* spp. such as alkaloids, flavonoids, and terpenoids can act on various targets in cell signalling pathways, restrain pro-inflammatory enzymes, lower the production of inflammatory cytokines and chemokines and reduce oxidative stress which will subsequently suppress inflammatory processes. Antiinflammatory effect of various extracts of *C. zedoaria* was studied in albino rats injected with carrageenan and histamine to induce paw edema. The highest activity was observed for 400mg/kg of chloroform extract, 200 mg/kg of petroleum ether extract and the ethanolic rhizome extract. Two sesquiterpenes- furanodiene and furanodienone reduced the TPA- induced inflammation in rat ears at the dosage of 1.0 µmol. The percentages of reduction were 75 and 53 respectively. Sesquiterpenes from zedoary essential oil were proved to have anti-inflammatory activities (Rahaman *et al.*, 2021; Makabe *et al*., 2006).

Three active compounds1,7-bis (4-hydroxyphenyl)-1,4,6-heptatrien-3-one, procurmenol and epiprocurmenol were isolated from *C. zedoaria* methanolic extract and proved to have antagonistic activity against the Tumor Necrosis Factor- α (TNF- α). Lipopolysaccharide activated macrophages were activated with the compounds which inhibit TNF- α production. The IC50 value of 1,7-bis (4-hydroxyphenyl)-1,4,6- heptatriene-3-one was 12.3 micro mole and that of procurcumenol was 310.5 micro mole (Jang *et al*., 2001).

Ullah *et al.* (2014) have tested the antinociceptive and anti-inflammatory activity of *C. zedoaria* ethanolic rhizome extract. Phytochemical screening has revealed the presence of various secondary metabolites like tannins, saponins, flavonoids, steroids, alkaloids, reducing sugars and terpenoids in the extract. Dehydrocurdione, the major component of *Curcuma zedoaria* has anti-inflammatory potency related to its antioxidant effect (Yoshioka *et al*., 1998).

Quantitative analysis study of free radical scavenging activity of the curcuminoids was done with the ethanolic extracts of *C. zedoaria* collected from 10 different locations of Thailand by Paramapojn *and* Gritsanapan (2009). HPLC technique was employed for the analysis. All the extracts were screened for their free radical scavenging activity using 1,1-diphenyl-2-picrylhydrazyl scavenging assay. The curcumin content of the extracts ranged from 1.46±0.45 to 5.73±0.11 % w/w (average 2.73±1.24 % w/w). Dimethoxy curcumin was in the range of 3.15±0.15 to 10.98±0.28 %w/w (average 7.37±2.71 %w/w). Range of bisdemethoxycurcumin was 0.49±0.02–2.99± 0.20 %w/w (average 1.40±0.82 %w/w). The separated compounds were observed to have free radical scavenging activity in the descending order of curcumin > demethoxycurcumin > bisdemethoxycurcumin.

2.3.23. Antibacterial activity

The antimicrobial and antifungal activity of *C. zedoaria* and *C. malabarica* rhizome extracts were tested by Wilson *et al*. (2005). Six bacterial and two fungal strains were used to test the inhibitory action of the extracts by agar gel diffusion and broth dilution methods. The chloroform, hexane, acetone, petroleum ether, ethanol and acetone extracts of zedoary tuber exhibited both antibacterial and antifungal activities.

Antibacterial effects of *C. zedoaria* against both gram positive (*Bacillus subtilis*, *Bacillus cereus, Bacillus magaterium, Staphylococcus aureus, Sarcinlutea)* and gram negative (*Pseudomonas aeruiginosa*, *Salmonella paratyphi, Salmonella typhi, Vibrio parahaemolyticus, Vibrio minicus, E coli*, and *Shigella)* strains were reported by Philip *et al.* (2009) and Silalahi (2020).

Two terpenoids with antibiotic activity were isolated from zedoary rhizome by Jeong and Shim (2015). The NMR analysis of both the compounds revealed their identity as ketolactone and orobanone. The antibiotic potential of both the compounds was tested against 5 bacterial strains - *Listeria monocytogenes, Salmonella typhimurium, Bacillus cereus, Escherichia coli* and *Staphylococcus pseudointermedius* by disc diffusion assay. Ketolactone exhibited strong antibacterial action against *Listeria monocytogenes* and *Staphylococcus pseudointermedius* at the concentration of 100 µg/dics. Orobanone in the same concentration inhibited *B. cereus*. The inhibition zones around *L. monocytogenes* and *S. pseudointermedius* measured 24 mm and 16 mm each, and that around B. *cereus* measured 18 mm.

Puspita *et al*. (2019) proved the effective use of *C. zedoaria* as an alternative for root canal irrigation system by studying the inhibitory action against *Streptococcus viridans*. Four different concentrations of zedoary

extracts (100%, 50%, 25% and 12.5%) were used as different test groups. Chlorhexidine and distilled water were used as positive and negative controls. 5 mL solutions from each test group and control group were applied to *S. viridins* culture in solid BHI medium and inhibition zone diameter was analysed. The strongest inhibition zone was measured at the concentration of 100% and was proved to have the highest antibacterial activity. The extract at 50% concentration and positive control group (2% chlorhexidine) showed significant inhibitory effect. Moderate antibacterial effect was recorded for 25% and 12.5% concentrations of the extract.

The antimicrobial activity of *C. zedoaria* extract was tested against the bacterial strains *Streptococcus mutans*, *Enterococcus faecalis*, *Staphylococcus aureus* and *Candida albicans* using the linear regression method by Bugno *et al*. (2007). The experiment proved the antimicrobial efficacy of zedoary as similar to the commercially using mouth rinse products. The result says that zedoary can be used as an alternative to improve the antimicrobial efficacy of oral hygiene products.

Chemical constituents from the essential oil of two *Curcuma* species, *C. angustifolia* and *C. zedoaria* were screened and tested for their antibacterial and antifungal activity by Jena *et al*. (2020). Two-dimensional gas chromatographic method along with time-of-flight mass spectroscopy were employed for the assay. A total of 139 compounds were detected from *C. angustifolia* essential oil and 147 compounds from the *C. zedoaria* fraction. Curzerenone (17.72 %), γ-eudesmol acetate (15.85 %), and germacrone (6.50 %) were the major constituents of zedoary extract. Both the species were recorded with significant activity towards five bacterial and three fungal strains. *C. zedoaria* essential oil showed the strongest inhibition against *Staphylococcus aureus* and *Candida albicans*.

Methanolic extract of zedoary at the dosage of 500 µg/disc showed significant antibacterial activity against *Shigella dysenteriae*, *Escherichia coli*, *Shigella sonnei* and *Salmonella typhi*. The inhibition zone diameters of tested bacteria were 19 mm, 18 mm, 17 mm and 16 mm respectively. The ethyl acetate extract had a moderate inhibitory action with the inhibition zone diameter of 17 mm against *S. sonei* and *Sh. dysenteriae* and 16 mm for *Sh. boydii* and *E. coli* (Shahriar, 2010).

Rhizome extract of zedoary was tested for its antibacterial activity by both *in vitro* and *in vivo* methods by Banisalam *et al*. (2011). Four bacterial strains of both gram positive and gram negative category were used for the study. *Pseudomonas aeruginosa*, *Escherichia coli*, *Staphylococcus aureus* and *Bacillus cereus* were used for the analysis. The *in vitro* system was supplemented with indole-3-butyric acid (IBA) and 6-benzyl aminopurine (BAP) as growth hormones. The growth of *E. coli* and *P. aeruginosa* was inhibited in the *in vitro* system only after the administration of the extract. The highest inhibition rate in the *in vitro* system was observed for *S. aureus*. *B. cereus* showed the highest inhibition rate in *in vivo* system compared to *in vitro* system.

Antibacterial and antifungal activity of the ethanolic extract of zedoary rhizome was reported by Chachad *et al*. (2016). Ethanolic extract at the concentration of 40 mg/ml was treated against different pathogenic bacteria and fungi. No activity was observed against *Klebsiella pneumoniae* and *Salmonella paratyphii*. The highest activity was recorded against *Staphylococcus aureus* and *Trychophyton mentagrophytes*.

Antibacterial activity of *C. zedoaria* was tested against the fish pathogen *Vibrio alginolyticus* by Maharajan and Sajin (2011). The pathogen caused heavy mortality rate of cultured fishes whose consumption would influence human health. *V. alginolyticus* was isolated from the infected fish *Lates*

calcarifer. The effect of zedoary extract was tested in 3 different organic solvents, dichloromethane, ethyl acetate and ethyl alcohol. Pathogen was controlled by all the 3 solvent extracts with the maximum inhibition rate recorded for ethyl alcohol extract. Inhibition zone formed by ethyl alcohol extract was measured as 30.16±0.20 mm at the concentration of 500 µg/disc. Nan *et al*. (2015) investigated the non-specific immune response by the combined action of *C. zedoaria* and *Z. zerumbet* extracts in *Epinephelus coioides*. Parameters like respiratory burst activity, lysozyme activity, SOD activity, phagocytic activity and reactive oxygen species were analysed in the study. The best immune response was recorded for the diet with 0.5g/kg of *C. zedoaria* combined with 1g/kg and 2.5 g/kg of *Z. zerumbet* extract.

Different concentrations of zedoary extract (0.05, 1.0 and 2.5 g/kg) were administered to *Epinephelus fuscoguttatus* to test the non-specific immune response of the extract. *E. fuscoguttatus* was provided the extract twice a day for 14 days. Observations were made on 0, 1, 2, 4, 7 and 14 days after treatment application to test the parameters such as phagocytic activity, respiratory burst activity, SOD (Super Oxide Dismutase) activity, chemiluminescent response and lysozyme activity. Significant effect on immune response was noted at the concentration of 0.5 g/kg of the extract on days 2 to 4. Fishes treated with 0.58/kg of the extract showed higher production of superoxide anion on 4^{th} and 7^{th} days after the application. Superoxide dismutase activity was higher on the 14^{th} day. Experimental result recommends the use *C. zedoaria* extract at the concentration of 0.5 g/kg in aquaculture to boost immune response of cultured fishes (Samad *et al*., 2022).

2.3.24. Endophytes

The endophytic bacterial diversity in *C. zedoaria* was studied by Sulistiyani *et al*. (2014). 16S DNA technology was employed for the identification of isolates. Among the 207 bacterial colonies isolated from

different parts of the plant, 73 colonies were identified as endophytic in nature. Endophytes were prevalent in the rhizome (38%) than in the stem (32%) and leaves (30%). Sequence analysis has revealed that the isolates belong to 23 different genera including *Stenothropomonas, Pseudomonas, Enterobacter, Providencia, Klebsiella, Dickeya, Pantoea, Bacillus, Acinetobacter, Citrobacter*, *Mycobacterium, Cellulomonas, Microbacterium, Methylobacterium, Penylobacterium, Roseomonas, Agrobacterium, Bosea, Xanthobacter, Rhizobium, Burkholderia, Ralstonia* and *Alcaligenes.*

Endophytic bacteria were identified from three different *Curcuma* species, *C. xanthorriza*, *C. aeruginosa* and *C. zedoaria* by Indrawati *et al*. (2018). The isolates were identified as *Bacillus cereus*, *B. amyloliquefacians*, *Lysinibacillus sphaericus* and two other *Bacillus* species. The isolates were tested for their antibacterial action against methicillin resistant *Staphylococcus aureus* (MRSA), *Citrobacter freundii* and *Klebsiella pneumoniae*. Maximum antibacterial activity was observed for *B. amyloliquefaciens* which produced 16 mm inhibition zone against the MRSA and 8 mm inhibition zone against *K. pneumoniae*. Two compounds, pyranon and di-(2-ethyl-hexyl) phthalate were extracted from the endophytic fungal genus *Penicillium* from *C. zedoaria* leaves (Muharni *et al*., 2014).

Sulistiyani *et al*. (2019) investigated the antimicrobial and toxic activities of secondary metabolites produced from the endophytic bacteria associated with *C. zedoaria*. The activity was tested against 6 microbial pathogens, *Pseudomonas aeruginosa*, *Escherichia coli*, *Candida albicans*, *Bacillus subtilis*, *Staphylococcus aureus* and *Saccharomyces cerevisiae*. A total of 73 endophytes were isolated from zedoary and tested for their antimicrobial potential. 16 bacterial isolates showed their capacity towards the tested microbes. Maximum activity was recorded for *Bacillus subtilis*, *Pseudomonas otitidis*, *Citrobacter freundii* and *Burkholdesia cenocepacia*. Toxicity of

endophytes was measured and the highest value was recorded for *Burkholderia cenocepacia* with an LC50 value 12.6 ppm.

2.3.25. Antifungal activity

Three antibiotic compounds were isolated from the ethanolic rhizome extract of zedoary which were resistant to the fungal strains, *Aspergillus niger*, *Trychophyton rubrum* and *Saccharomyces cerevisiae*. The most abundant compound was discovered as ethyl p-methoxycinnamate (EPMC) by MS, UV, IR and PMR analysis. EPMC showed antifungal activity against an array of fungal strains at varying concentrations. At the concentrations lesser than 10 mug/ml, the compound inhibited *Aspergillus niger, Epidermophyton floccosum, Saccharomyces cerevisiae and Trychophyton rubrum.* The growth of *Aspergillus fumigatus, Penicillium purpurogenum, Sclerotium rolifsii, Microsporum gypseum, Fusarium oxysporum, Trignoposis variabilis* and *Helminthosporium oryzae* was inhibited at the concentration lesser than 25 mug/ml. EPMC inhibited the growth of *Trichophyton mentagrophytes* and *Candida krusei* at a concentration lesser than 50 mug/ml (Gupta *et al*., 1976).

Shinobu *et al*. (2011) have reported the antifungal activity of *C. zedoaria* hydroalcoholic extract against yeast of the genus *Candida*. The genus causes oropharyngeal candidiasis in the patients infected with HIV. The growth of *C. albicans*, *C. tropicalis* and *C. glabrata* was inhibited at lower concentrations of zedoary extract.

The use of *C. zedoaria* extract as an antifungal agent was proved by Ficker *et al*. (2003). The test was performed using 11 different species of Zingiberaceae. Disc diffusion method was employed for the purpose. Significant antifungal activity was observed for the rhizome extracts of *C. zedoaria*, *C. purpureum* and *Alpinia galanga.* Prominent antifungal activity was recorded for *C. zedoaria* extract against an array of human pathogenic

fungi. The property was best proved against the fungal strains which were resistant to the commonly used antifungal compounds like Amphothericin B and ketoconazole.

Curcumenol from *C. zedoaria* was tested for its broad-spectrum antifungal activity against an array of fungal phytopathogens. Significant activity was observed against the fungal strains *Fusarium graminearum*, *Phoma wasabiae*, *Plasmodiophora brassicae* and *Magnaporthe grisea* with the minimal inhibition concentrations of 0.5, 1.0, 1.0 and 1.0 mg/mL respectively. The variation in pH (2-10) and temperature (30-80 Â°C) did not affect the activity of extract. The studied fungi were examined under transmission electron microscope and scanning electron microscope. It revealed that curcumenol destroyed the fungal cell wall and caused damage to the nutrient circulation in them (Chen *et al*., 2013).

2.3.26. Antiviral activity

The antiviral effect of *C. zedoaria* volatile oil on H_5N_1 avian influenza virus was studied by Huang *et al*. (2009). The extract was proved to have the properties of virus killing, virus inhibition and the prevention of virus spread in the MDCK cell line.

2.3.27. Antipyretic activity

According to Azam *et al*. (2014) the ethanolic extract of zedoary rhizome possesses antipyretic effect tested via yeast induced pyresis method. The study was performed on young Long Evan rats that weighed from 178 g to 210 g, fasted for 18 hours before the experiment and only provided with water. A 20% aqueous suspension of brewer's yeast at the concentration of 20 mL/kg was injected to the rats for inducing fever. Rectal temperature was measured 18 hours before and 18 hours after the administration of brewer's yeast. The test group was provided with aqueous suspension of zedoary ethanolic extract

and the rectal temperature was measured after 1, 2 and 3 hours of its application. The drug when provided at a dosage of 500 mg/kg showed significant antipyretic activity after 2 hours of its application. The dosage at 750 mg/kg had a moderate activity after 2 hours and significant activity after 3 hours of drug administration.

Zedoary derived dihydrocurdione was tested for its analgesic and antipyretic activity. The analgesic activity of the extract was tested by measuring the number of writhes induced by acetic acid in mice. Significant activity was observed at the dosage of 40 mg/kg and above. Measuring the rectal temperature in rats treated with baker's yeast was employed to prove the antipyretic effect of the compound. Rectal temperature in rats was reduced significantly by the administration of dihydrocurdione. When the rats were treated with dihydrocurdione at the dosage of 80 or 200 mg/kg a significant reduction in fever index was observed (Matsuda *et al*., 1998).

2.3.28. Antimutagenic activity

The antimutagenic effects of zedoary rhizome extract was examined along with 35 other crude drugs used in Chinese herbal medicinal systems by Lee and Lin (1988). The investigation was conducted using the salmonella/ microsomal system to test the activity against banzo(a)pyrene or picrolonic acid. The boiling water extract of zedoary was proved to have a moderate antimutagenic activity against benzo(a)pyrene.

Antimutagenic activity of *C. zedoaria* was studied by Peng *et al*. (2010). Both the methanolic and aqueous extracts of zedoary rhizome were tested against 2 mutagenic compounds, 2-amino 3-methyl imidazol (4,5-f) quinoline (IQ) and 4-nitroquinoline-N-oxide (4-NQO) on *Salmonella typhimurium*. Methanolic extract showed higher antimutagenicity compared to the aqueous

extract. Curcuminoid content of the methanolic extract was measured to be as 44.3 mg/g while that of aqueous extract was only 0.09 mg/g.

2.3.29. Bioinsecticidal activity

Bioinsecticidal action of essential oil (EO) from the rhizomes of zedoary was studied by Oliveira *et al*. (2019). EO was extracted using hydro distillation method from the rhizomes of both dormant and budding stages. Experiment was performed on *Aedes aegypti* larvae and pupae using the immersion test method. EO from the dormant rhizome was recorded with the LC99.9 value of 0.01 mg/mL for larvae and 1.38 mg/mL for pupae. EO from budding rhizome was recorded with the LC99.9 value of 0.08 mg/mL and 2.63 mg/mL for larvae and pupae respectively. Autobiographical method was employed to determine the action potential of EO from both the rhizome types. EO from dormant rhizome showed higher inhibition on acetylcholinesterase enzyme (0.039 mg/mL) than the budding rhizome (1.056 mg/mL).

2.3.30. Drug substitution

Use of *C. zedoaria* as a substitute for *Aristolochia rotunda* was studied by Ansari *et al*. (2021). The Albino Wistar rats were provided with CCl_4 at the concentration of 1 ml/kg to induce hepatotoxicity. The treated rats were categorized into 5 different groups. First two groups were served as plain control and negative control while the 3rd group was provided with the drug sylimarin. *A. rotunda* and *C. zedoaria* drugs were administered to the 4th and 5th groups respectively. Analysis has shown a decline in the total serum bilirubin, direct bilirubin, ALT and AST levels in test groups. Liver samples were collected and analysed. Rats provided with *A. rotunda* and *C. zedoaria* exhibited significant recovery rate from hepatic toxicity. Thus, the use of *C. zedoaria* as a substitute of *A. rotunda* was suggested.

2.3.31. Ca^{2+} channel blockage

Dihydrocurdione from zedoary rhizome blocks Ca^{2+} channel in the intestinal and vascular smooth muscles of mice. At 1 mM concentration dihydrocurdione reduced K^+ ion stimulated increase of cytosolic Ca^{2+} level in rats. The blockage of extracellular Ca^{2+} is the reason for inhibitory effects of dihydrocurdione on the Ca^{2+} channels of vascular smooth muscles and intestine (Irie *et al*., 2000).

2.3.32. Prevention of drunkenness

Effect of the isolated compound curcumenone from zedoary in preventing drunkenness was studied by Kimura *et al*. (2013). The activity was measured for ethanol extract, n-hexane soluble fraction and an isolated compound of zedoary. Ethanolic extract at the dosage of 1000 mg/kg has prevented alcohol induced drunkenness at 60 and 120 minutes after the administration of 40% alcohol. The isolated compound prevented drunkenness at 30, 60 and 120 minutes after alcoholic intake at varying concentrations (3, 10 and 30 mg/kg). The same was effective to increase liver ADH activity after 30 and 60 minutes of administration of alcohol (40%). NMR spectral analysis identified the isolated compound as curcumenone.

2.3.33. Antivenom activity

The inhibitory action of *C. zedoaria* extract against cobra venom has been proved by ELISA test. Western immunoblotting analysis revealed that the major components of the venom are being targeted by the extract. The neurotoxin and the protein degradative enzymes which were the major components of the venom were the targets of the extract. It attenuated the action of the venom toxin by extending the contraction time of diaphragm muscle after the process of envenomation and also protected cellular proteins

from venom degradative enzymes (Daduang *et al*., 2005). Alam (2014) has also reported the use of *C. zedoaria* as antivenom in the treatment of cobra bite.

2.3.34. Other properties

The antifertility effect of *C. zedoaria* ethanolic extract in rat testis was studied by Ongko *et al*. (2019). 24 white male Wistar rats (*Rattus noverticus*) were used for the study, which weighed about 160 - 200 g and came under the age group of 6-8 weeks. They were randomly grouped into 4, each consisting of 6 rats. The ethanolic rhizome extract was given for each group and then observed. Data were recorded and tested using normality test and ANOVA. The result showed the effect of ethanolic rhizome extract in causing significant difference in number of spermatogenic cell layers and mitotic count.

Herbal tea derived from *C. zedoaria* (ZHT) was tested for its anti-lipidemic and anti-hypercholestrolemic activities. 3 samples of ZHT- T1, T2 and T3 were prepared using 50 mg, 1 g and 1.5 g of dried powder of zedoary in 200 ml of boiling water for 5 minutes. The samples were studied for their DPPH inhibition, total phenolics, colour tonality, pH, total soluble solids, sensory acceptance, etc. 3 groups of male human volunteers (G1, G2, G3) each with 10 mild hypercholestrolemic males were provided with the ZHT samples for two months. Blood drawn from the volunteers after 12 hours of fasting was collected on day 0, day 30 and day 60. The blood serum parameters like total cholesterol, HDL-cholesterol, LDL-cholesterol and triglyceride concentration were tested for all the samples. The result showed that test group T3 exhibited the highest TPC, DPPH inhibition and total flavonoid values (Tariq *et al*., 2016).

It is reported that administration of gambir leaf waste and white turmeric increased the milk production and antioxidant content in dairy cows. It reduced the mastitis rate in cow. 5 different treatments were given to the test group. A-

containing 0% gambir leaf waste provided with 0.02% white turmeric, B- with 20% gambir leaf waste with 0.02% white turmeric. C, D and E contained gambir leaf waste at the concentrations of 30%, 40% and 50% supplemented with 0.02% white turmeric. A significant effect was observed for the treatment with 30% gambir leaf waste and 0.02% white turmeric. The same treatment increased milk production by 18.15% and antioxidant content of the milk by 26.50% in dairy cows (Nurdin and Susanty, 2015).

2.3.35. Genetic variability

Genetic variability of any species indicates its adaptability to different environments and the diversity of its genetic base. In the case of food crops, higher variability and broad genetic base ensure food security and provide the raw material for improvement. Analysis of the existing genetic diversity within a species is very important for designing breeding programmes (Cooper *et al*., 2001). The following part of the review of literature provides available information on previous efforts to study the genetic variability of the species under study. However, only very few efforts have been made in this direction especially using conventional tools.

Syahid and Heryanto (2017) collected 12 accessions of C. zedoaria from different regions of Indonasia. The morphological, growth and yield parameters of all the 12 accessions were determined in the experiment conducted. Each accession was planted and raised in a 2.5 m x 3.5 m plot at 50 cm x 50 cm of plant spacing in Randomized Block Design (RBD). Stem type, leaf base, leaf shape, leaf tip, blade colour and stem colour were the morphological characters observed. The growth characters included plant height, number of tillers, number of leaves per stem, leaf length, leaf width and the thickness of leaves. The yield characters studied were weight of rhizome, rhizome length, rhizome width and the thickness of rhizome. All the studied characters showed significant variation among the 12 accessions collected.

2.3.36. Molecular characterization

Molecular characterization has been attempted in some species of *Curcuma* to study its genetic variability during the first and second decades of the present century. Angel *et al*. (2008) attempted to study the genetic variability of starchy *Curcuma* species using RAPD (Random Amplified Polymorphic DNA) method. The RAPD pattern generated by 20 primers during the study revealed high degree of polymorphism. 264 bands out of the 274 bands generated were polymorphic in nature. Using UPGMA characterization technique, all the studied species were separated into 3 clusters. *C. leucorrhiza, C. aromatica* and *C. brog* together formed a cluster within which *C. zedoaria* and *C. longa* formed a subgroup. *C. harita* was genetically distinct from all the other *Curcuma* species. RAPD technique has great application in identifying starchy *Curcuma* species because it is difficult to distinguish the species by leaf morphology.

4 genes having potential role in the curcuminoid metabolic pathway were isolated from *C. zedoaria* by Lan *et al*. (2018). Primers used for the experiment were isolated from the untranslated regions of 4 different genes (CURS1, CURS2, CURS3 and DCS) from *C. longa*. These genes play key roles in curcumin biosynthesis. The experiment proved the expression of all the 4 genes in *C. zedoaria*. Curcumin biosynthesis was also detected in the callus and rhizome of *C. zedoaria* by HPLC analysis. CZCURS1, CZCURS2 and CZCURS3 were measured to be 1240, 1288 and 1265 nu respectively in length. CZDCS gene had the maximum length of 1382 nu.

Yeast extract and salicylic acid were tested for their effect on the expression of two genes involved in curcuminoid biosynthesis in *C. zedoaria*. CzDCS and CURS1-3 were the genes considered for the study. Cells were treated with both yeast extract and salicylic acid and observed for the level of gene expression. The treated cells have shown 1.14 to 3.64 fold expression of

curcuminoid genes than in the control group. Concentration of curcumin in the treated cells was 1.61 to 2.53 folds more than that in the control group (Lan *et al.,* 2019).

2.3.37. Micropropagation

Miachir *et al*. (2004) has established the protocol for *in vitro* micropropagation and callogenesis of *C. zedoaria*. Shoot apices raised from the rhizome and from *in vitro* plants were grown in the Murashige & Skoog culture medium. The medium was supplemented with 2.0 mg/L of benzyl amino purine and 30 g/L of sucrose. New plantlets raised in the medium were transferred to greenhouse conditions and kept for at least 10 days for the success of acclimatization. Acclimatization and the development of plantlets were made easier by treating the plantlets with endomycorrhizal fungi.

Bharalee *et al*. (2005) have performed the *in vitro* clonal propagation of *C. zedoaria* and *C. caesia* using rhizome buds as explant. The shoot multiplication response of *C. caesia* was the best when MS basal medium was provided with 4 mg/L BAP and 1.5mg/L NAA. The concentrations 0.5 mg/L of NAA and 1 mg/L of BAP were optimal for the shoot multiplication response in *C. zedoaria*.

The micropropagation protocol for *C. zedoaria* and *C. zerumbet* was proposed by Stanly and Keng in 2007. Shoot explants cultured in liquid medium produced almost twice the number of new shoots than in the solid medium with the same composition. *C. zedoaria* shoot explants when cultured in solid MS medium provided with 0.5 mg/L IBA and 0.5 mg/L BA produced 2.3 shoots per explant. Liquid MS medium with the same composition of medium components produced 6.1 shoots per explant.

The quantification of micropropagation of *C. zedoaria* was done by Mello *et al*. (2000). Shoot meristems were cultured in solid MS agar medium

supplemented with 30g/L of sucrose and 2.0 mg/L of BAP and then incubated at 25±2ºC for 16/8 hours of photoperiod (light/dark). The cultures were transferred at an interval of 40 days and the number of plantlets produced per transfer was determined.

Rhizome sprout culture of zedoary was done by Loc *et al*. in 2005. Basal MS medium supplemented with coconut water (CW) and benzylaminopurine (BA) at the concentrations ranging from 0.5 mg/L to 5.0 mg/L were used for the culture. Emerged shoots were then sub cultured in the MS medium provided with varying concentrations of BA and kinetin and 20 (V/V) CW along with Indole Butyric Acid (IBA). After 30 days of culturing, 5.6 shoots were obtained from the MS medium provided with 0.5 mg/L IBA and 3 mg/L BA along with 20 (V/V) CW.

3 culture systems, the agar gelled medium system, temporary immersion system (TIS) and shake flask system were employed for the micropropagation of *C. zedoaria* and *Z. zerumbet in vitro* by Stanly *et al*. (2010). Shoot multiplication rate in both the species was not influenced by immersion period in TIS. TIS eliminated the problem of hyper hydrocity in the shake flask system. Plants of both the species developed by the remaining systems were also grown well and no morphological changes were observed after the process of acclimatization. The result says that the best culture system for *in vitro* production of both the species is TIS.

Micropropagation using rhizome buds for the propagation of *C. zedoaria* was attempted by Shahinozzaman *et al*. (2013). Explants were inoculated and raised in the Murashige and Skoog medium. Different combinations of NAA and BA were applied to the culture medium to screen the best combination at which maximum shoot multiplication occurred. 1.0 µM NAA coupled with 8.0 µM of BA exhibited the maximum capacity for shoot multiplication.

Shoot formation using *in vitro* culture system was practiced in *C. zedoaria* by Yulizar et al. (2014). Different concentrations of BAP and sucrose were added to the MS medium to find out the optimal concentration for shoot induction. 1.5, 3 and 4.5 mg/L concentrations of BAP were used in the medium along with 3% and 5% sucrose. Shoot induction was best induced by the application of 1.5 mg/L of BAP with 5% sucrose.

The above review provides an account of the available information on the status of research works carried out on *Curcuma zedoaria.* Based on this, the following experiments have been designed and executed so as to explore the level of genetic variability in the germplasm of the plant collected presently and the interrelationship of the growth and yield characters. An effort has also been made to identify the superior genotypes.

Chapter III

MATERIALS AND METHODS

Curcuma zedoaria Rosc. is one of the valuable plants belonging to the family Zingiberaceae with considerable pharmacognostic and medicinal importance. The plant is still underexplored in terms of genetic diversity, variability and plant improvement despite its wide range of possible uses. Hence, using the germplasm collected from various locations across Kerala State, India, a study was carried out to evaluate its genetic divergence, genetic variability, genetic control of characters, character association and variation of agronomic morphometric features. The study also concentrated on selecting superior genotypes from the collected accessions depending on their overall performance. In addition, plant growth performance was evaluated based on the type of planting material used, such as mother rhizome, primary finger and secondary finger. The experiments were planned and conducted between 2018 and 2021 in the experimental plot of the Department of Botany, University of Calicut, Kerala, India.

3.1. The experimental material

The experimental material under study was *Curcuma zedoaria* Rosc., also referred to as white turmeric, zedoary or *gajutsu* (Fig. 3.1). It is a perennial aromatic herbaceous plant of the Zingiberaceae family. The plant is utilised in a number of ayurvedic and indigenous medicinal systems (Saikia and Nath, 2003). The plant grows to a height of 100–120 cm, and its underground rhizome is the most valuable component commercially. Rhizomes have a creamy white inner portion and a distinctive camphoraceous odour. The apical buds of the rhizome give rise to a spike type inflorescence that rises above the soil. At the base of the inflorescence, there are bracts with green or red tints that cover the flowers. The upper bracts are pink, white or red in colour and do not produce flowers but instead serve to attract pollinators. The three lobed, sub orbicular-lipped flowers are yellow in colour. It has a width of 5 inches (Fischer, 1928).

The flower has six stamens in total, but only one of them is viable. A coloured structure resembling a petal is created when the five sterile stamens fuse together. Fruit is a capsule.

3.2. The experimental plot

The study was carried out in the experimental field of the Genetics and Plant Breeding Division of University of Calicut, Kerala, India (Fig. 3.2). The experimental plot is located in the Malappuram District of Kerala at 11º13' N latitude and 75º88' E longitude. Tropical monsoon climate prevails in the area with south-west monsoon rains from June to September and north-east monsoon rains from October to November. The dry spell period is from December to May, with a few summer showers in the months of March, April and May (Tables 3.1 to 3.4) (Anonymous, 2018; Anonymous, 2019; Anonymous, 2020; Anonymous, 2021). The annual rainfall was 2468 mm in 2018, 2442 mm in 2019, 2289 mm in 2020 and 2790 mm in 2021 and the average temperature ranged from 22.94ºC to 33.07ºC.

Table 3.1. Weather data of the experimental plot for 2018.

Months	Rainfall (mm)	Temperature (ºC)		Relative humidity (%)
		Minimum	Maximum	
January	0	22.16	29.83	66.81
February	0	22.66	31.66	63.12
March	26.37	24.50	32.83	70.19
April	116.02	25.66	32	78
May	332.23	26	31.16	81.81
June	474.61	24.66	28	85.12
July	527.34	24.16	27	89.19
August	522.07	23.66	26.83	91.12
September	100.2	23.83	29	84.88
October	263.67	24	29.33	79.56
November	89.65	23.66	30.16	76.31
December	15.82	23.50	30.16	71.56
Yearly value	2467.97	24.04	29.83	78.14

Fig. 3.1. *Curcuma zedoaria* - single plant, inflorescence, flower and rhizome

a. Single plant; b. Inflorescence; c. Flower; d. Rhizome; e. Rhizome CS.

Fig. 3.2. *Curcuma zedoaria* in the experimental layout.

Table 3.2. Weather data of the experimental plot for 2019.

Months	Rainfall (mm)	Temperature (°C)		Relative humidity (%)
		Minimum	Maximum	
January	0	23.83	30.66	62.44
February	0	25.16	32.16	66
March	0	25.66	32.66	70.12
April	79.1	27.66	33.16	74.81
May	63.28	27.16	32.5	78.44
June	237.3	26.5	30	82.38
July	416.6	25	28.16	85.56
August	711.91	23.66	28	87.69
September	332.23	24.83	28.83	86.12
October	427.15	24.66	30	82.38
November	110.74	24.83	32.16	79.56
December	63.28	17.83	24.5	74.88
Yearly value	2441.6	24.73	30.23	77.53

Table 3.3. Weather data of the experimental plot for 2020.

Months	Rainfall (mm)	Temperature (°C)		Relative humidity (%)
		Minimum	Maximum	
January	0	23.35	35.61	69.56
February	0	22.61	38.06	65.5
March	0	24.09	37.64	70.62
April	42.19	25.53	37.06	73.38
May	179.3	25.70	34.19	80.38
June	348.05	24.43	29.73	83.19
July	479.88	23.90	29.06	87.5
August	432.42	23.54	29.41	87.38
September	437.7	23.03	29.06	87.12
October	168.75	22.16	30.38	83.38
November	142.38	22.43	33.36	76.31
December	58.01	21.87	33.22	71.44
Yearly Value	2288.68	23.55	33.07	78.00

Table 3.4. Weather data of the experimental plot for 2021.

Months	Rainfall (mm)	Temperature (°C)		Relative humidity (%)
		Minimum	Maximum	
January	100.2	22.38	33.32	76.62
February	26.37	22.32	37.02	66.51
March	58.01	24.03	38.03	70.56
April	84.16	24.36	34.63	76.69
May	378.95	24.48	31.35	81.75
June	312.47	24.13	29.93	83.81
July	438.64	23.12	27.74	88.38
August	328.43	23.56	30.14	87.69
September	252.65	23.56	30.26	87.19
October	454.53	22.09	29.48	85.44
November	334.61	21.13	29.53	83.69
December	21.32	20.09	34.25	74.06
Yearly value	2790	22.94	32.14	79.70

3.3. The experimental programme

Fifty seven accessions of *C. zedoaria* were collected from different locations across Kerala. The experimental programme was designed and performed from 2018 to 2021 with the goals of assessing the genetic diversity of the experimental plant and identifying superior genotypes from the germplasm gathered. A screening plot was planted with all collected accessions during the 2018 crop season, and sufficient planting materials were collected. The details of the accessions collected and the places of collection are given in Table 3.5 and Fig. 3.3.

Table 3.5. Accessions of *Curcuma zedoaria* studied.

Accession Number	Source	District	Latitude & Longitude
CUW 1	Kolothupadi	Thrissur	10.335602141683593, 76.2191498261729
CUW 2	Nadavarambu	Thrissur	10.335940359413831, 76.21554531534387
CUW 3	Vellangallur	Thrissur	10.303075233334388, 76.2092637362794
CUW 4	Konathukunnu	Thrissur	10.282248496370716, 76.21381049019121
CUW 5	Narayanamangalam	Thrissur	10.249291584322593, 76.20629283761569
CUW 6	Moothakunnam	Ernakulam	10.190160508723627, 76.20054062805416
CUW 7	Vadakkekara	Ernakulam	10.158934383875916, 76.21019171437734
CUW 8	Kalluchira	Ernakulam	9.908425875396533, 76.26928935139335
CUW 9	Maliankara	Ernakulam	10.181710694112724, 76.19411918766284
CUW 10	Kottuvallikkad	Ernakulam	10.182979098607921, 76.18999638364576
CUW 11	Kodungallur	Thrissur	10.22832772707788, 76.19895618212048
CUW12	Santhipuram	Thrissur	10.262267048893603, 76.17974397942744
CUW 13	Mathilakam	Thrissur	10.290482508508678, 76.16753694946495
CUW 14	Edamuttam	Thrissur	10.374657325345307, 76.12382770026888
CUW 15	Triprayar	Thrissur	10.420048216981982, 76.10552977899152
CUW 16	Ramavarmapuram	Thrissur	10.566127776149647, 76.23463423848378
CUW 17	Vellanikkara	Thrissur	10.545867982256324, 76.27132698574621

Accession Number	Source	District	Latitude & Longitude
CUW 18	Velloor	Kottayam	9.833935835204613, 76.45750978579447
CUW 19	Koyilandy	Kozhikode	11.446183855381527, 75.6990600042729
CUW 20	Mukkam	Kozhikode	11.322057198969382, 75.99551117251912
CUW 21	Chevayoor	Kozhikode	11.269555810264633, 75.82578787301661
CUW 22	Thenhipalam	Malappuram	11.134518833674331, 75.89194809757733
CUW 23	Olipramkadavu	Malappuram	11.140855085166034, 75.88329063947522
CUW 24	Arakkapparambu	Ernakulam	9.95102233923468, 76.25074540268969
CUW 25	Pithrukunnam	Kottayam	9.778926433210776, 76.3954804894629
CUW 26	Muravanthuruthu	Ernakulam	10.167971858117237, 76.213332046871
CUW 27	Murinjapuzha	Kottayam	9.819733854385928, 76.3921991161809
CUW 28	Kattikkunnu	Kottayam	9.82746844279854, 76.39310869017547
CUW 29	Kundannoor	Thrissur	10.709078432770442, 76.21186446245603
CUW 30	Arukkutti	Alappuzha	9.863488007577441, 76.32688910272907
CUW 31	Kumbalam	Ernakulam	9.902018152787115, 76.30994434438259
CUW 32	Kalathezhath	Alappuzha	11.230054450419452, 76.20958309173109
CUW 33	Pallippuram	Alappuzha	9.754567863464615, 76.35809127178395
CUW 34	Vazhathara	Alappuzha	9.691298404708155, 76.32563755850808

Accession Number	Source	District	Latitude & Longitude
CUW 35	Thirunalloor	Alappuzha	9.718024580448589, 76.36212575189369
CUW 36	Vadassery	Alappuzha	9.492107858449957, 76.34520749157855
CUW 37	Arthunkal	Alappuzha	9.66391887675194, 76.29911707728387
CUW 38	Pottikavala	Alappuzha	9.65566428378806, 76.3530594500352
CUW 39	Muttathiparambu	Alappuzha	9.658990315263207, 76.349607298989
CUW 40	Karikkad	Alappuzha	9.663633653968821, 76.37662063705949
CUW 41	Kattachira	Kottayam	9.677775694300127, 76.59802545951771
CUW 42	Kudavechoor	Kottayam	9.673500593582844, 76.41410684875363
CUW 43	Arippanthara	Kottayam	9.66606165312009, 76.42611733337438
CUW 44	Vechoor	Kottayam	9.658077933613573, 76.45385662345906
CUW 45	Kadanad	Kottayam	9.77783076793663, 76.70117846268974
CUW 46	Pizhaku	Kottayam	9.796318470769581, 76.68862581215653
CUW 47	Chazhoor	Thrissur	10.434784658917216, 76.14126205914762
CUW 48	Ammadam	Thrissur	10.455513534200051, 76.19294930357994
CUW 49	Peringottukara	Thrissur	10.428622753061799, 76.12392421228088
CUW 50	Chemmappilly	Thrissur	10.425634168239906, 76.1150040045451
CUW 51	Vadakkumuri	Thrissur	10.434157961514554, 76.11482967086872

Accession Number	Source	District	Latitude & Longitude
CUW 52	Padiyam	Thrissur	10.452308783890768, 76.10325222265985
CUW 53	Kattoor	Thrissur	10.372711648673336, 76.16610940184731
CUW 54	Manavalassery	Thrissur	10.35910908973788, 76.17581141846047
CUW 55	Konathukunnu	Thrissur	10.282185157154933, 76.2138963208783
CUW 56	Kovilakathkunnu	Thrissur	10.271494673324268, 76.21841348969154
CUW 57	Karooppadanna	Thrissur	10.269379713202072, 76.20563505450116

3.4. Planting and aftercare

In the first week of May 2019, just before the onset of the south-west monsoon, the evaluation trials and other experiments were started. Healthy rhizome portions weighing 25 g –30 g and measuring 4 cm –7 cm in length were chosen as the planting material. The rhizomes were planted in 38 cm × 35 cm polybags filled with garden soil, sand and enriched compost mixed in 3:1:1 ratio. Irrigation was carried out once a day on all non-rainy days and weeding was carried out as needed. From the 30th day of planting through the fifth month of growth, 2 g of NPK (18:18:18) was added per polybag at monthly intervals. After 8 months of growth, the plants were harvested simultaneously. Recommended package of practices and plant protection measures were followed to raise a healthy crop (KAU, 2011).

3.5. Data recording

Three plants each selected statistically from each replication in the case of each accession were used to record the data, which were then averaged. Data

Fig. 3.3. Different locations of collection of *Curcuma zedoaria*

regarding the growth characters were collected six months after planting and the data on yield characters were recorded after the harvest using destructive sampling method (Table 3.6). Leaf area was determined using the following formula from leaf length and leaf breadth:

Leaf area = Leaf length × Leaf breadth × Conversion factor.

The conversion factor for leaf area was determined as shown in Table 3.7. The conversion factor was calculated by measuring the mean area of five randomly selected leaves.

Table 3.6. Characters of *Curcuma zedoaria* observed in the present study.

Sl. No.	Character
Growth characters	
1	Plant height (cm)
2	Number of tillers
3	Number of leaves per tiller
4	Leaf length (cm)
5	Leaf breadth (cm)
6	Leaf area (cm^2)
Yield characters	
7	Number of primary fingers
8	Length of primary finger (cm)
9	Circumference of primary finger (cm)
10	Number of secondary fingers
11	Length of secondary finger (cm)
12	Circumference of secondary finger (cm)
13	Length of mother rhizome (cm)
14	Circumference of mother rhizome (cm)
15	Yield per plant (g)

Table 3.7. Calculation of conversion factor for leaf area.

Species	Conversion factor for leaf area					
Curcuma zedoaria	**R1**	**R2**	**R3**	**R4**	**R5**	**Mean**
	0.54	0.74	0.56	0.59	0.61	0.61

3.6. Study of genetic variability

Genetic variability analysis refers to the measurement of variation present among individual genotypes of a study population. The knowledge of genetic variability is crucial for the better understanding of the genetic diversity of a species. It is important both in the perspectives of conservation and improvement when it comes to crop science. In contrast to populations with reduced variability, those with higher variability are more adapted to thrive in a variety of climatic and environmental situations. It is quite useful for selecting superior breeding materials of the germplasm for breeding purposes. Hence, the genetic variability of the *C. zedoaria* population under study in terms of their morphometric agronomic characters was examined using appropriate statistical methods utilizing uniform planting materials collected from the fifty seven accessions of the germplasm grown in the experimental plot in the previous year.

Study of genetic variability of the germplasm in terms of the genetic control of agronomic characters, phenotypic and genotypic variability, heritability of characters and genetic advance of characters under selection in the case of the crop species under study was carried out during the first crop season of 2019 adopting standard cropping procedure. The experiment was laid out in randomized block design (RBD) with three replications and 18 plants per plot in polybags under open conditions.

3.6.1. Genetic control of agronomic characters

Frequency distribution analysis was carried out after pooling the data collected as mentioned above from the plants grown for the purpose of studying genetic variability in order to find out the nature of genetic control of characters and to investigate the distribution pattern of dominant and recessive contributing factors for each agronomic trait in the population.

3.6.2. Phenotypic and genotypic variability

In plant breeding programmes, phenotypic and genotypic diversity, as well as their nature and extent, play a significant role in the selection of promising genotypes. Using agronomic characters from the population that was raised in the experimental field in 2019, the phenotypic and genotypic variability of *C. zedoaria* was investigated. The experimental population was raised in Randomised Block Design with 57 accessions of the species, three replications per accession, and 18 plants per plot. The experiment was repeated in 2020 and 2021 for confirmation.

Variation within population and between populations was evaluated and analysis of variance (ANOVA) carried out to reveal the significance of the differences that existed between the accessions in terms of the fifteen morphometric characters studied. F value was calculated for the purpose and its significance was tested with reference to standard F table (Fischer and Yates,1963). Critical Difference (CD) was found out with the formula:

$$CD = t_{0.05} \times \sqrt{\frac{2VE}{r}}$$

Where $t_{0.05}$= t at error degree of freedom at 5% level, VE= Error mean square, r = Number of replications.

Genotypic and phenotypic variance of the different characters studied was calculated as per Singh and Chaudhary (1985) using the formula:

Genotypic variance ($\sigma^2 g$) = $\frac{\text{MSS for treatment–MSS for error}}{\text{Number of replications}}$

Phenotypic variance ($\sigma^2 p$) = $\sigma^2 g + \sigma^2 e$

Where $\sigma^2 e$ = Error variance

Phenotypic and genotypic coefficients of variation of the studied agronomic traits were calculated by the formula of Burton and Devane (1953). Coefficients of variation are useful in the comparison of relative variability of different traits because they are examined in different units (Stanfield, 1991).

Phenotypic coefficient of variation (PCV) = $\frac{\sigma p}{\bar{x}} \times 100$

Where σp = Phenotypic standard deviation and $\bar{x}$ is the grand mean of the character.

Genotypic coefficient of variation (GCV) = $\frac{\sigma g}{\bar{x}} \times 100$

Where σg = Genotypic standard deviation.

3.6.3. Heritability of agronomic characters

Heritability estimates of agronomic characters show the effectiveness of selection and indicate the degree at which a specific character is being transmitted through generations (Shukla *et al.,* 2006; Asfaw *et al*., 2017). The parameter of heritability (broad sense) involves all types of gene action and thus forms a broad estimate of heritability (Chahal and Gosal, 2002). It is the fraction of the total variance that is heritable and is estimated as the percentage of genotypic variance over phenotypic variance (Jain, 1982). Knowledge on heritability of the characters of a species helps the breeder to predict their responses to selection and the behaviour of succeeding generations (Ul- Haq *et al*., 2008).

Heritability (broad sense) $H^2 = \frac{\sigma^2 g}{\sigma^2 p} \times 100$

Where $\sigma^2 g$ = Genotypic variance and $\sigma^2 p$ = Phenotypic variance

3.6.4. Genetic advance under selection

Genetic advance of a character indicates the level of gain attained by the character under a specific selection pressure. The best conditions for selection are indicated by a higher value of genetic advance and better heritability (Ramanujam and Thirumalachar, 1967; Bello *et al*., 2012). Therefore, using high heritability (broad sense) coupled with higher genetic advance is more preferred. Genetic advance is the degree to which the progeny can be genetically improved by selection over the original population (Chahal and Gosal, 2002). Genetic advance was investigated as per Singh and Chaudhary (1985).

Genetic advance (GA) = $\frac{KH^2\sigma p}{\bar{x}}$

Where H^2 = Heritability (broad sense), K = Selection differential which is 2.06 at 5% intensity of selection in large samples and σp = Phenotypic standard deviation (Allard, 1960).

3.7. Study of genetic divergence

Genetic divergence of *C. zedoaria* was studied between the 57 accessions by principal component analysis using the statistical software STATISTICA following Unweighted Pair Group Method with Arithmetic mean (UPGMA) as per Sneath and Sokal (1973).

3.8. Study of correlation of agronomic characters

The mutual relationship between different agronomic characters of the species can be understood by correlation studies. It also helps in determining

the component character for selection process for the genetic improvement of a character. The interrelationships among the fifteen agronomic characters were studied presently using correlation coefficient as per the method proposed by Rangaswamy (1995). Correlation coefficients indicate the direction and intensity of interrelationship of characters under study.

3.9. Study of character association

The study was performed using the statistical software STATISTICA by factor analysis. Factor analysis by means of principal component analysis was performed based on the fifteen agronomic characters. Study of character association aids in data reduction, grouping of variables and in finding out the lead characters which can be selected for further breeding programmes.

3.10. Performance analysis of the accessions collected

The comparative performance of different accessions of *Curcuma zedoaria* has been evaluated based on six growth and nine yield characters with the help of performance indices calculated for each agronomic characters according to Amaravenmathy and Srinivasan (2003).

$$\text{Performance index} = \frac{\text{Accession mean of the character}}{\text{Grand mean of the character}}$$

3.11. Study of the performance of *Curcuma zedoaria* Rosc. populations based on the status of the planting materials used

A study was conducted to investigate the variation in yield of the species based on different planting materials, such as mother rhizomes, primary fingers, and secondary fingers. For each accession, three different types of planting materials were collected from the germplasm maintained at University of Calicut. New populations raised from these different seed materials included 57 plants for each type of the seed material. Healthy rhizome pieces measuring about 4 cm -7 cm in length and 25 g-30 g in weight were used for planting

during the 2021 cropping season. The plants reached maturity after six months of growth, and then the growth characters were recorded from the field. The plants were harvested and yield characters were recorded at 8 months of growth. For the comparison of the performance of plants developed using different types of planting materials, the collected data were compiled and analysed using analysis of variance (ANOVA).

Chapter IV

RESULTS AND DISCUSSION

The genetic diversity in crop plants provides plant breeders with the opportunity to develop improved varieties with desirable characteristics. All systematic plant breeding programmes rely on intra and inter populational variability of crop plants. *Curcuma zedoaria* Rosc., popularly known as zedoary is a well known ethnomedicinal plant from the Zingiberaceae family that is used in ayurvedic medicines. In addition, it is a well known component of Indian traditional folk medicines. Although the plant has a wide range of economic applications, it continues to remain underutilized. The current study was conducted with the aim of analysing important genetic parameters of the plant including genetic variability, genetic divergence, character association and performance of different genotypes of *C. zedoaria* collected from various districts across Kerala State, India. Data on the agronomic traits of the genotypes collected and analysed for this purpose are discussed under appropriate headings in the following sections.

4.1. Genetic variability of *Curcuma zedoaria* Rosc.

The magnitude and nature of a crop's genetic diversity aid breeders in choosing suitable selection criteria and breeding techniques to develop novel varieties of the species. Results of the experiments carried out to analyse the genetic variability of the species under study in the study area are given below.

4.1.1. Genetic control of growth and yield characters

To identify the mechanism of genetic control involved in the agronomic traits of crop plants, frequency distribution analysis can be used. An analysis of the distribution of dominant and recessive alleles in the gene pools of these characters is performed here. In the present experiment, six growth characters

and nine yield characters of zedoary namely plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and the circumference of mother rhizome were analyzed based on a population of 513 plants grown for the purpose.

Quantitative characters with polygenic control show normal frequency distribution when the allelic combinations are distributed in the gene pool of the population as per the principles of probability and when the dominant and recessive alleles are in equal frequencies. The bell-shaped normal distribution curve shows skewness when there is variation in the frequency of the dominant and recessive alleles in the distribution (Chahal and Gosal, 2002). All of the morphometric traits in the current experiment displayed continuous frequency distribution, as shown by their frequency curves, which are presented in Tables 4.1 to 4.15 and Figures 4.1 to 4.15. Continuous frequency distribution that includes all possible intermediates implies that these traits are under polygenic control.

Among the growth characters studied, plant height and leaf breadth showed continuous distribution with accumulation of dominant contributing alleles slightly skewing the distribution towards the right side of the curve. This distribution indicates that the gene pool of these characters shows accumulation of higher number of dominant alleles even when maintaining fairly good genetic base ranging from comparatively lower to higher values. The frequencies of classes with higher values of the characters, however, are relatively low. Hence, for the development of promising varieties, it is necessary to select genotypes that accumulate the maximum amount of dominant contributing factors while maintaining a broad genetic base.

The growth characters like number of tillers, leaves per tiller, leaf length and leaf area have shown continuous distribution with accumulation of higher number of recessive contributing factors, indicating that the gene pools of these characters exhibit skewness towards the proximal side of the distribution curve. This indicates that there is a greater accumulation of recessive contributing alleles in the population under study. For these characters, the accumulation of the dominant contributing factors is very low. Therefore, it is necessary to employ scientific selection processes to develop varieties with higher accumulation of dominant alleles of these characters. However, the frequency distributions for these characteristics also show rather broad genetic base, demonstrating the existence of genotypes with different levels of allelic combinations, ensuring the genetic diversity of the plant population under study.

Among the yield characters studied, all the nine characters namely yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and the circumference of mother rhizome showed skewness towards the proximal side of the distribution. This indicates that a higher number of recessive contributing alleles are accumulated in the population studied. This also suggests that choosing better genotypes and phenotypes with more dominant contributing alleles is necessary to develop superior varieties.

However, the broad spread that can be seen in the frequency curves of these characters indicates that such distributions have an advantage from conservation point of view.

Table 4.1. Frequency distribution of plant height in the *Curcuma zedoaria* population.

Plant height (cm)	Number of plants
10-25	9
25-40	83
40-55	93
55-70	62
70-85	91
85-100	143
100-115	26

Fig. 4.1. Frequency curve of plant height.

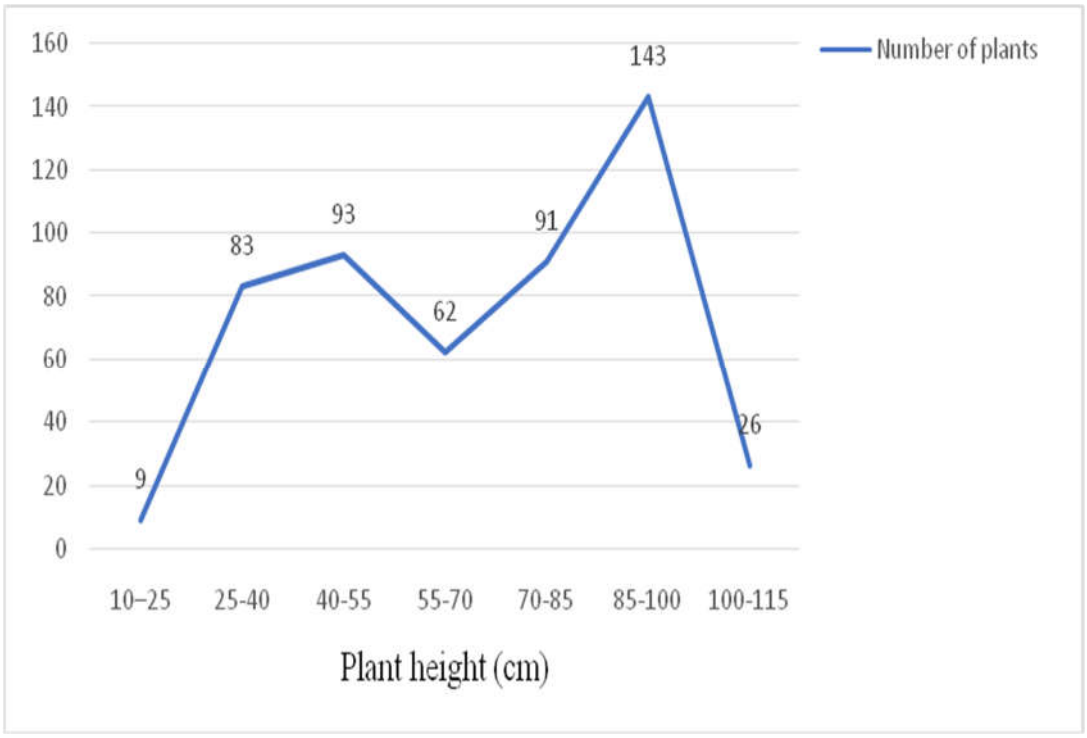


Table 4.2. Frequency distribution of number of tillers.

Number of tillers	Number of plants
1 – 2	408
2 – 3	96
3 – 4	4

Fig. 4.2. Frequency curve of number of tillers.

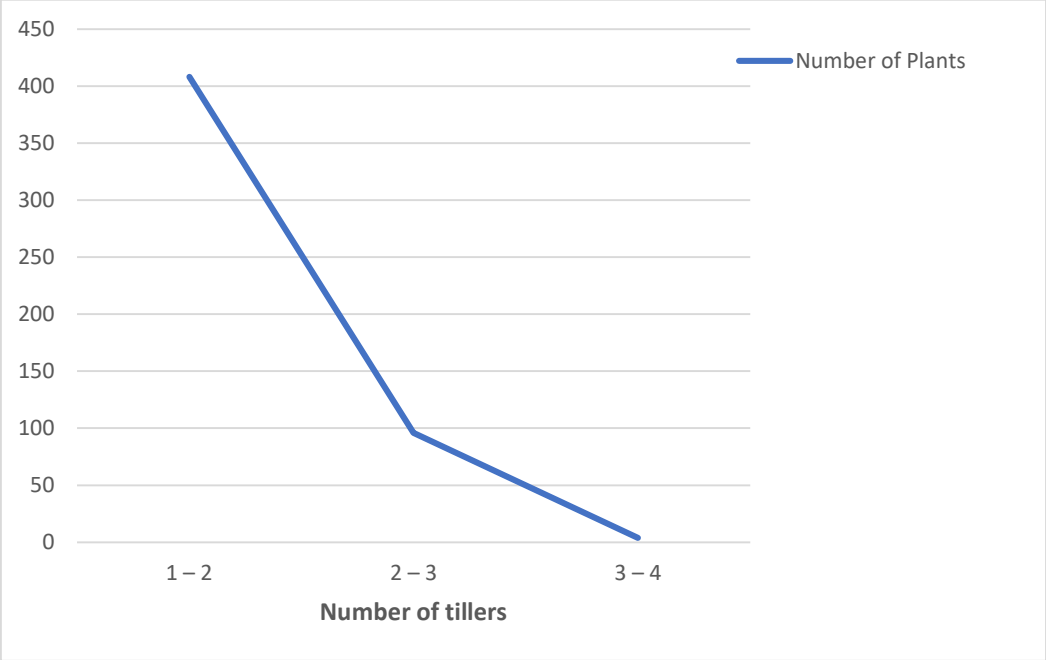


Table 4.3. Frequency distribution of number of leaves per tiller.

Number of leaves per tiller	Number of plants
1-5	40
5-10	455
10-15	13

Fig. 4.3. Frequency curve of number of leaves per tiller.

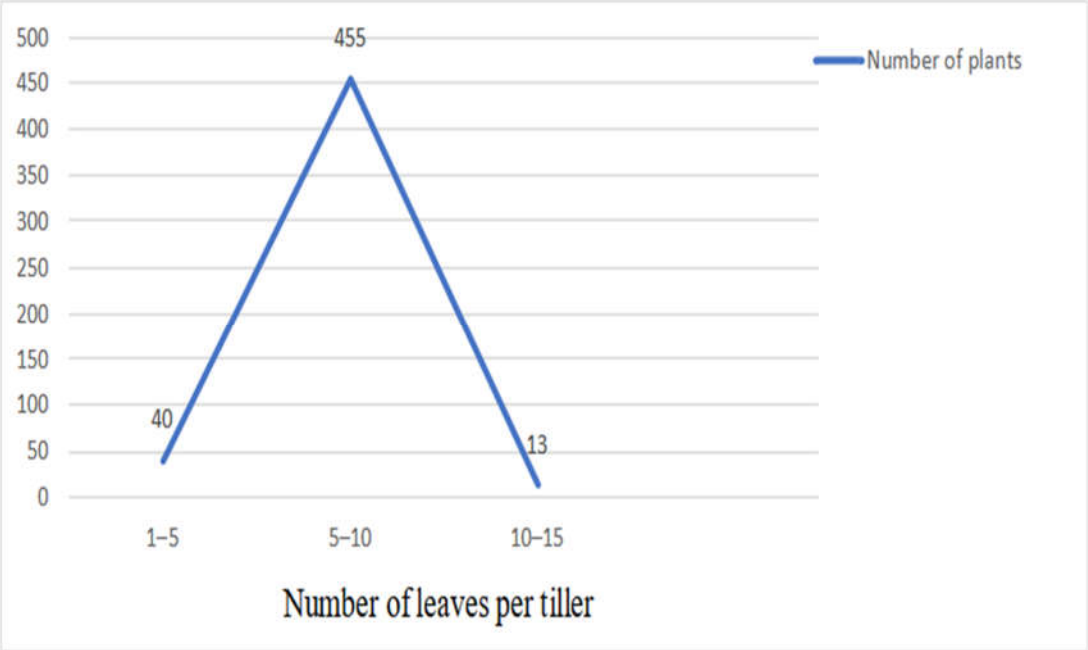


Table 4.4. Frequency distribution of leaf length.

Leaf length (cm)	Number of plants
10-20	38
20-30	67
30-40	89
40-50	82
50-60	67
60-70	63
70-80	2

Fig.4.4. Frequency curve of leaf length.

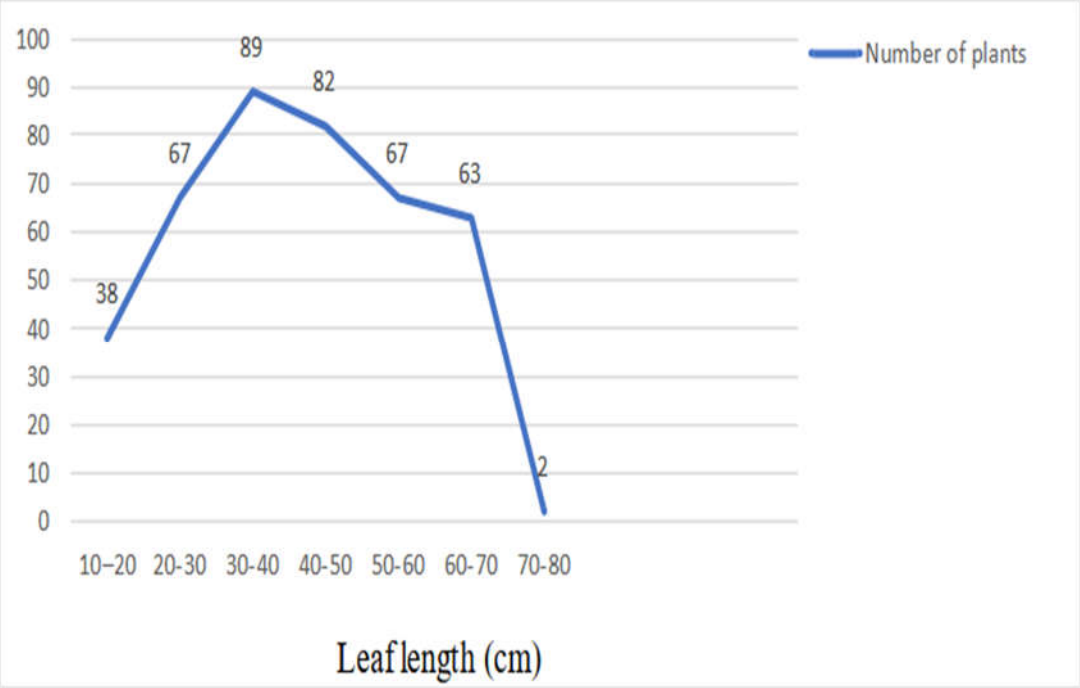


Table 4.5. Frequency distribution of leaf breadth.

Leaf breadth (cm)	Number of plants
2-3	24
3-4	90
4-5	51
5-6	33
6-7	55
7-8	88
8-9	86
9-10	63
10-11	16
11-12	2

Fig. 4.5. Frequency curve of leaf breadth.

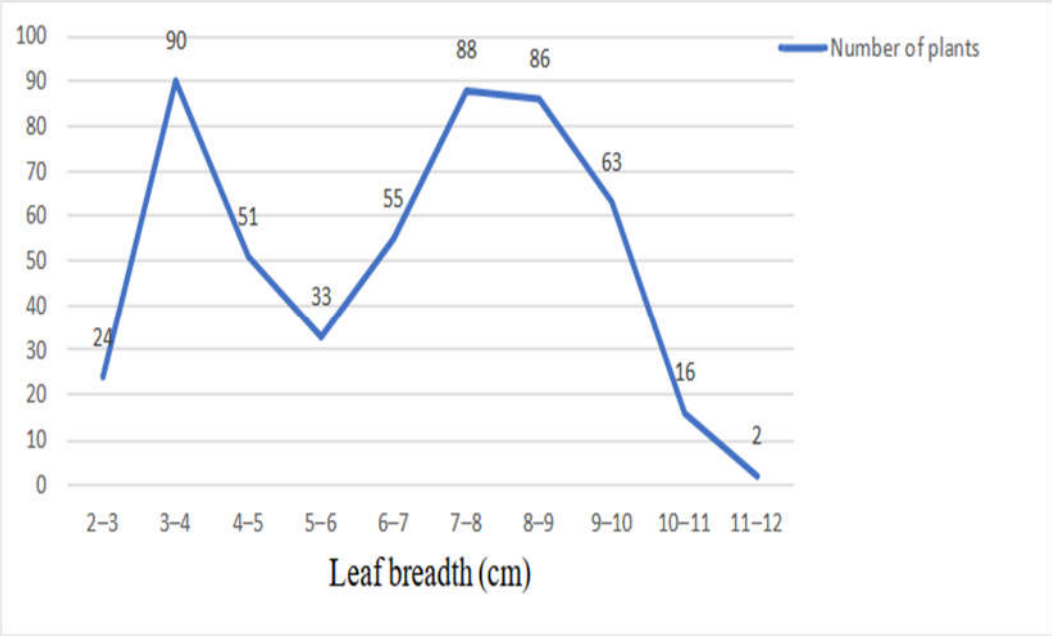


Table 4.6. Frequency distribution of leaf area.

Leaf area (cm^2)	Number of plants
15-75	117
75-135	72
135-195	51
195-255	91
255-315	108
315-375	58
375-435	11

Fig. 4.6. Frequency curve of leaf area.

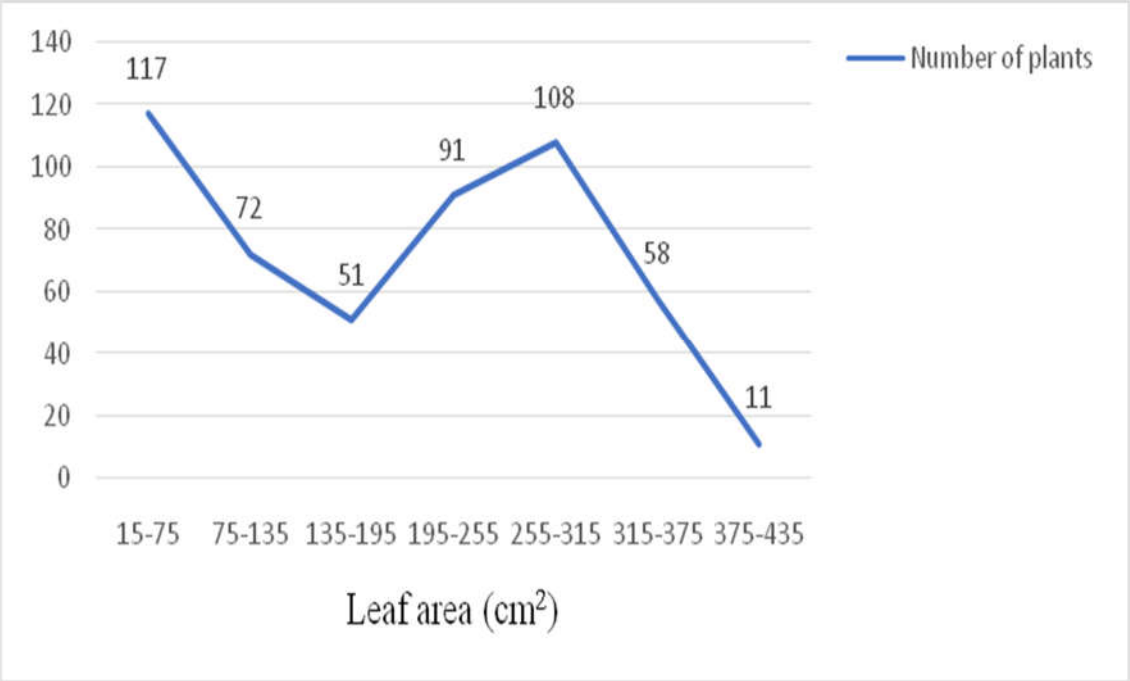


Table 4.7. Frequency distribution of yield per plant.

Yield per plant (g)	Number of plants
20-90	137
90-160	146
160-230	109
230-300	77
300-370	26
370-440	9
440-510	2
510-580	2
580-650	1

Fig. 4.7. Frequency curve of yield per plant.

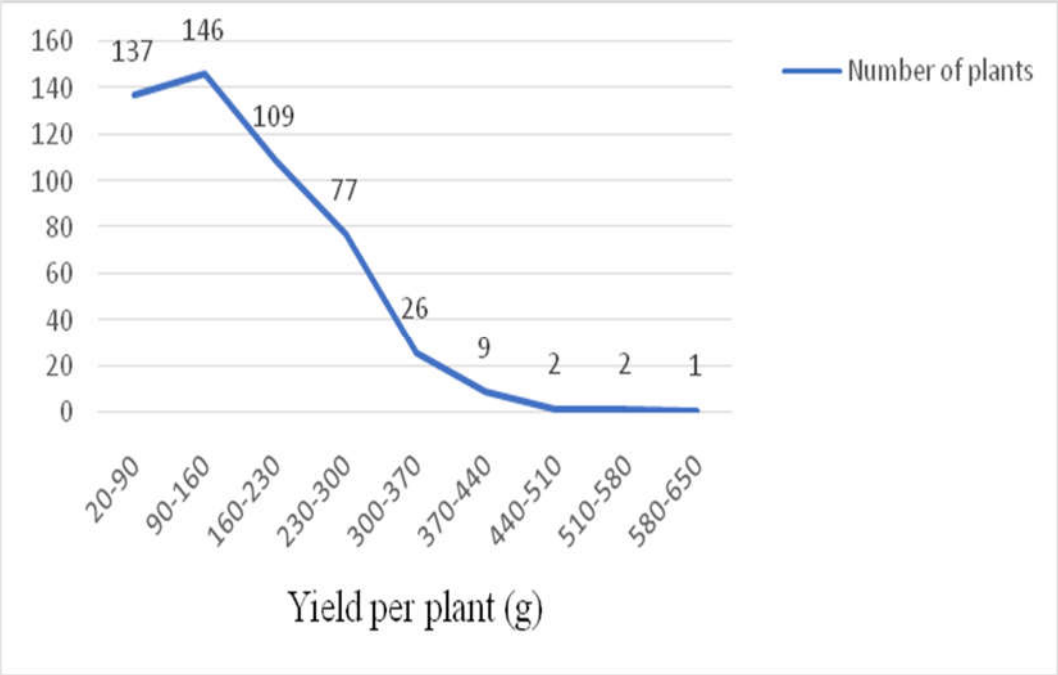


Table 4.8. Frequency distribution of number of primary fingers.

Number of primary fingers	Number of plants
1-3	176
3-5	282
5-7	50

Fig. 4.8. Frequency curve of number of primary fingers.

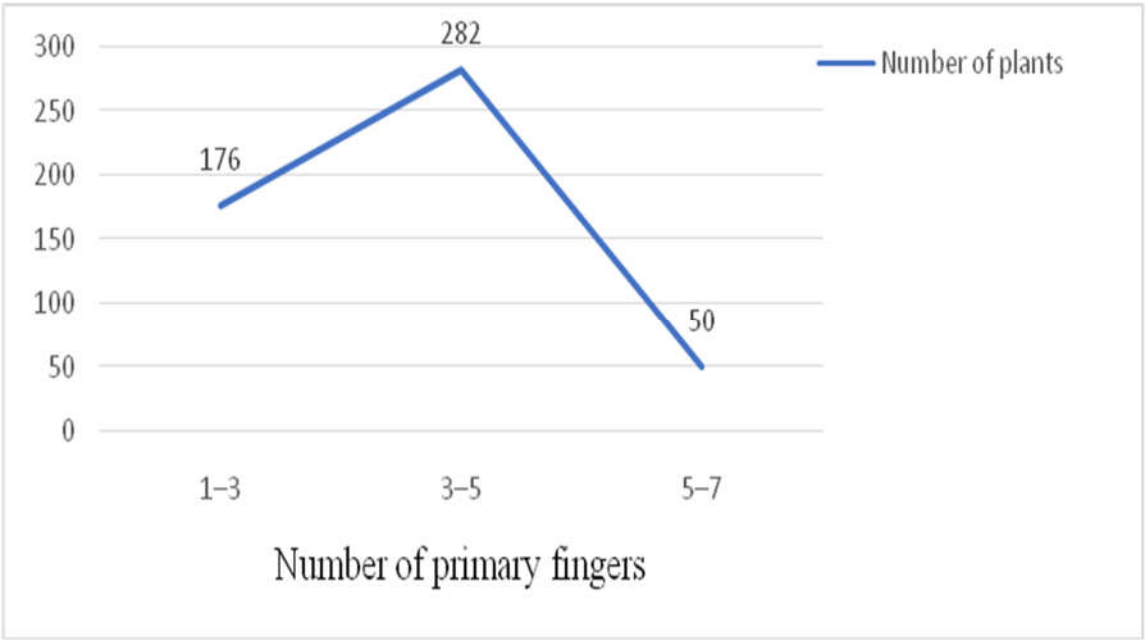


Table 4.9. Frequency distribution of number of secondary fingers.

Number of secondary fingers	Number of plants
1-5	14
5-10	98
10-15	114
15-20	99
20-25	89
25-30	50
30-35	27
35-40	14
40-45	3

Fig. 4.9. Frequency curve of number of secondary fingers.

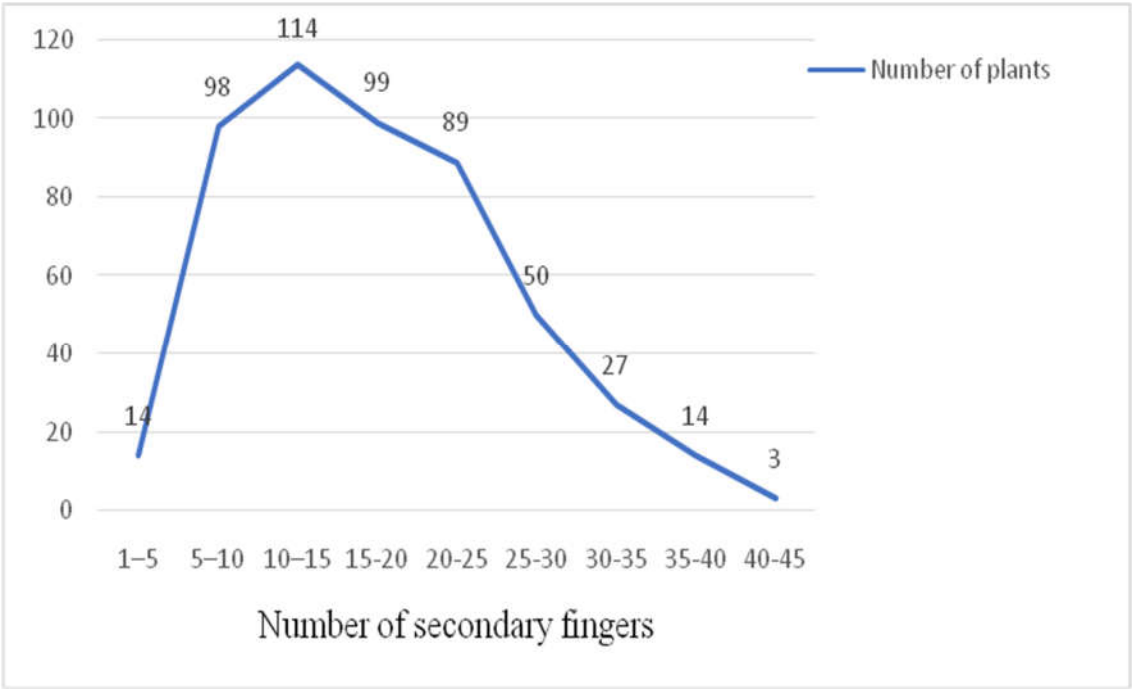


Table 4.10. Frequency distribution of length of primary finger.

Length of primary finger (cm)	Number of plants
2-5	18
5-8	66
8-11	83
11-14	119
14-17	136
17-20	70
20-23	10
23-26	4
26-29	2

Fig. 4.10. Frequency curve of length of primary finger.

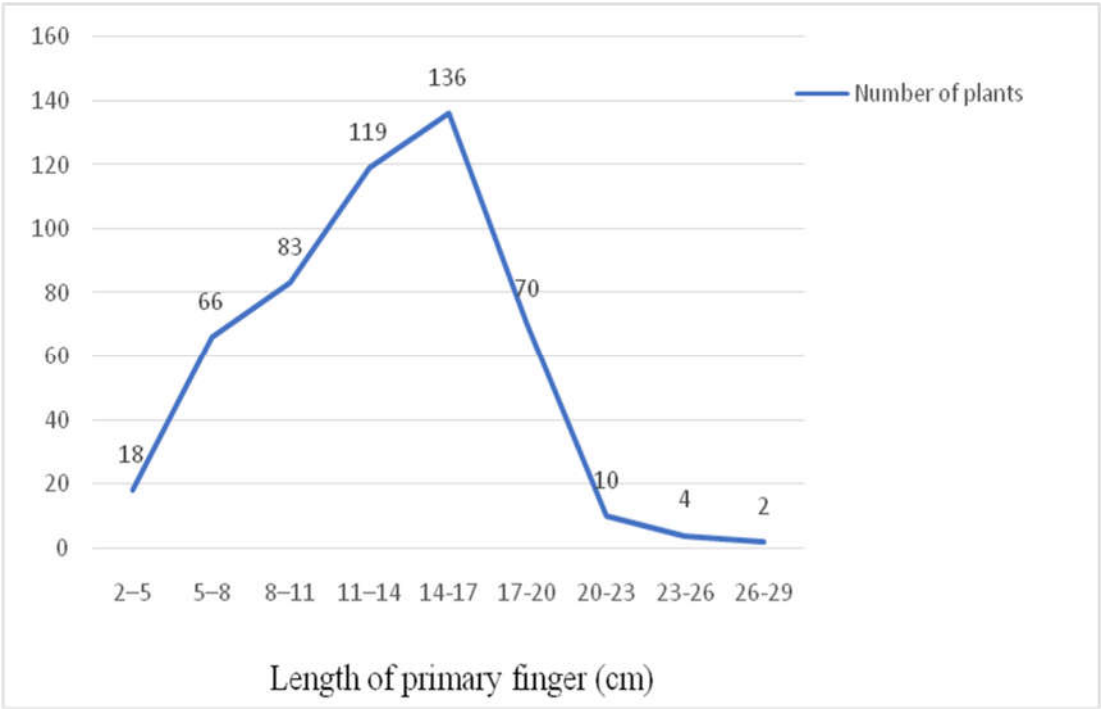


Table 4.11. Frequency distribution of circumference of primary finger.

Circumference of primary finger (cm)	Number of plants
0.5-1.5	1
1.5-2.5	12
2.5-3.5	78
3.5-4.5	93
4.5-5.5	178
5.5-6.5	86
6.5-7.5	41
7.5-8.5	16
8.5-9.5	3

Fig. 4.11. Frequency curve of circumference of primary finger.

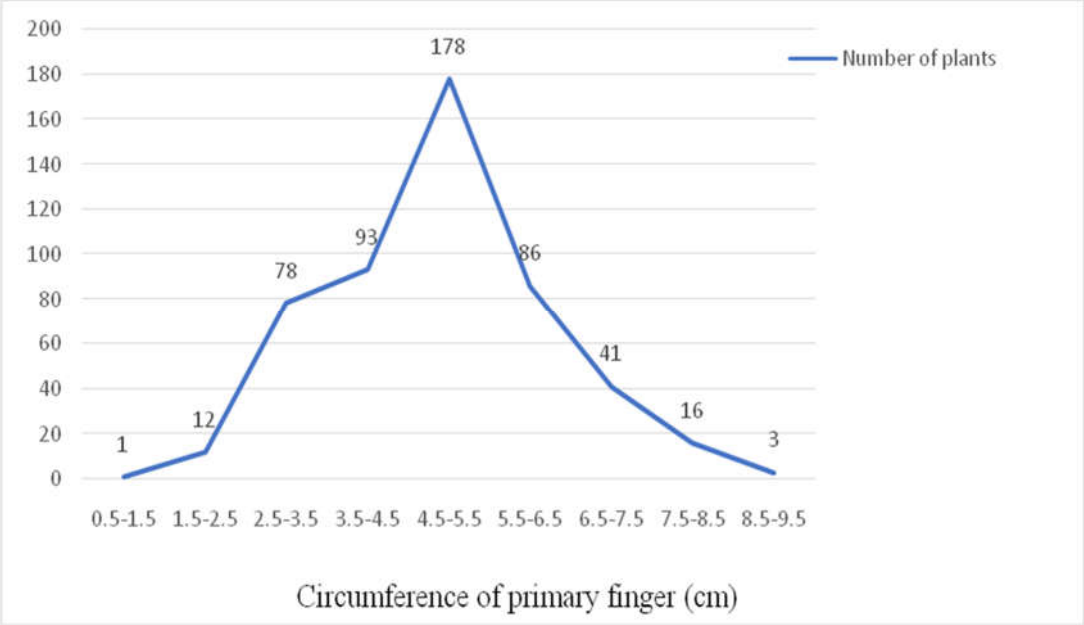


Table 4.12. Frequency distribution of length of secondary finger.

Length of secondary finger (cm)	Number of plants
1-2.5	54
2.5-4	73
4-5.5	50
5.5-7	61
7-8.5	77
8.5-10	67
10-11.5	45
11.5-13	24
13-14.5	13
14.5-16	7
16-17.5	3
17.5-19	1
19-20.5	1

Fig. 4.12. Frequency curve of length of secondary finger.

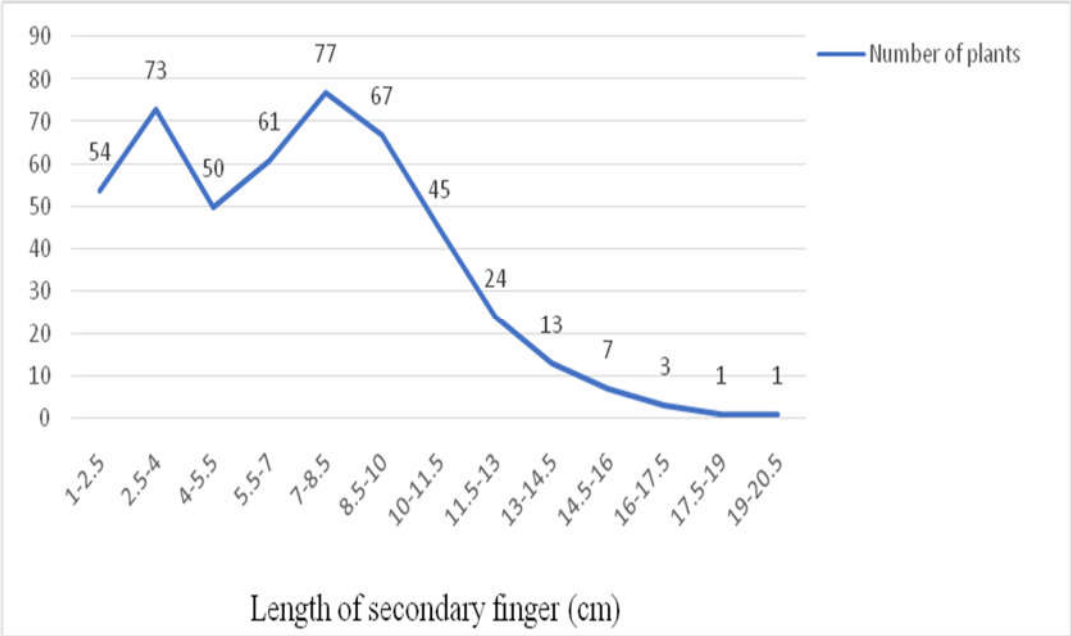


Table 4.13. Frequency distribution of circumference of secondary finger.

Circumference of secondary finger (cm)	Number of plants
0.5-2	37
2-3.5	134
3.5-5	222
5-6.5	73
6.5-8	10

Fig. 4.13. Frequency curve of circumference of secondary finger.

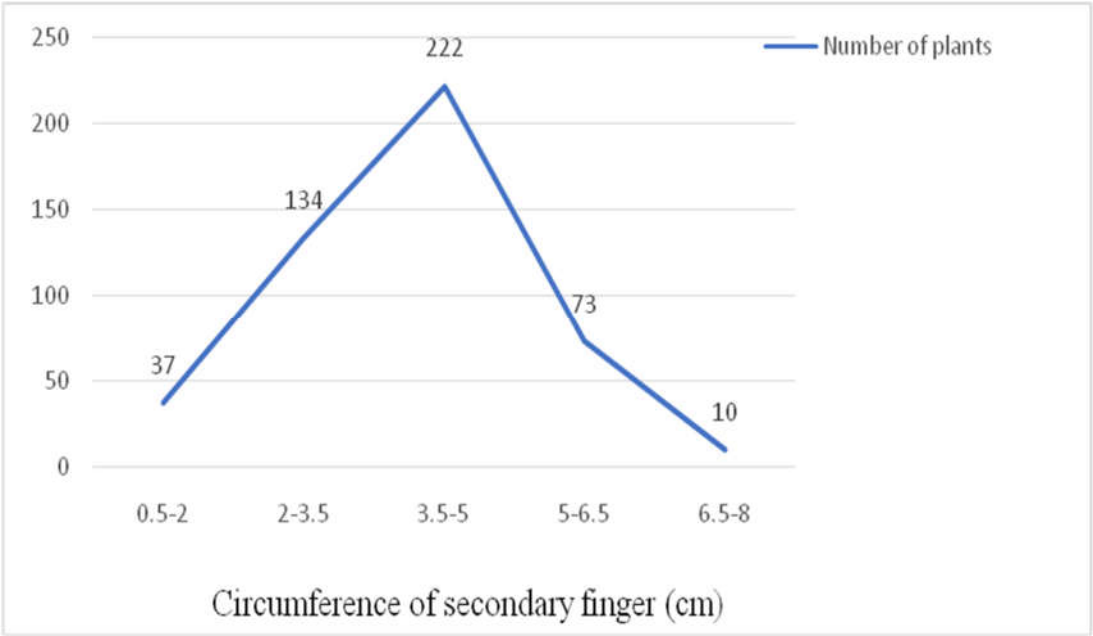


Table 4.14. Frequency distribution of length of mother rhizome.

Length of mother rhizome (cm)	Number of plants
2-3	50
3-4	119
4-5	181
5-6	110
6-7	32
7-8	15
8-9	1

Fig. 4.14. Frequency curve of length of mother rhizome.

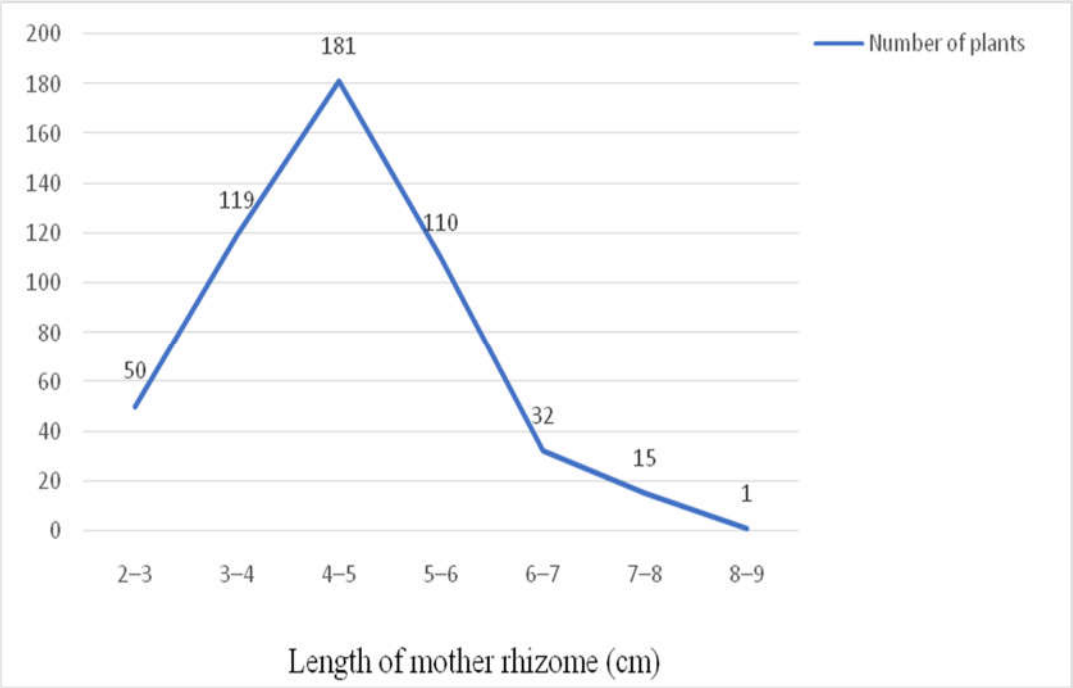
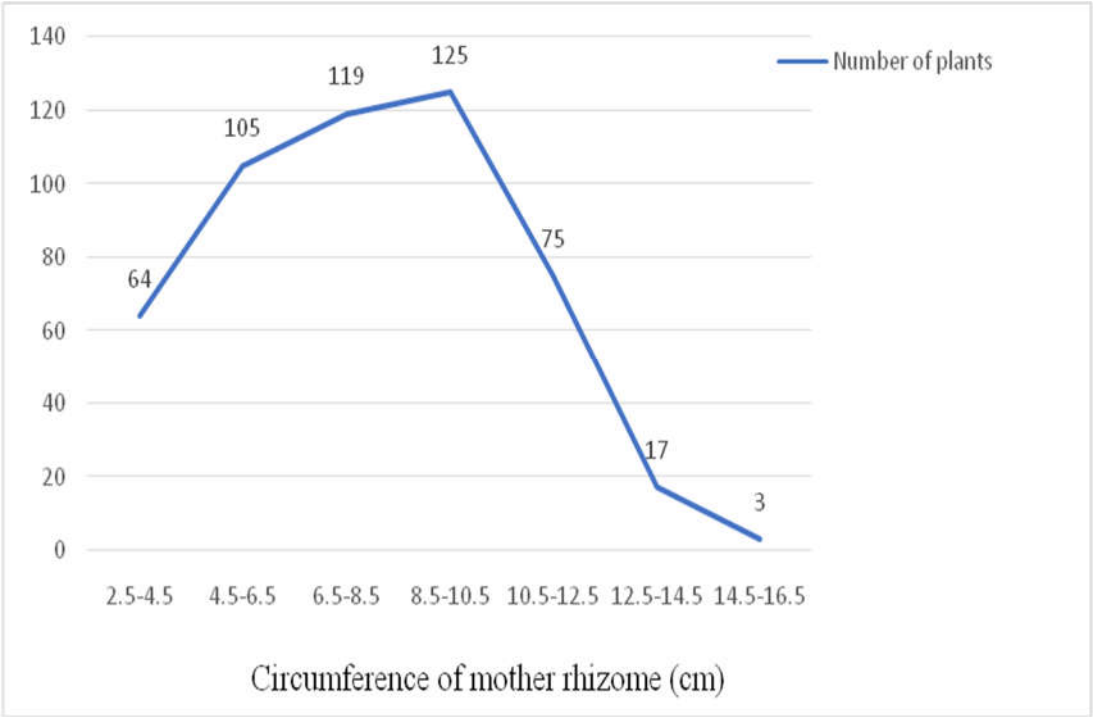


Table 4.15. Frequency distribution of circumference of mother rhizome.

Circumference of mother rhizome (cm)	Number of plants
2.5-4.5	64
4.5-6.5	105
6.5-8.5	119
8.5-10.5	125
10.5-12.5	75
12.5-14.5	17
14.5-16.5	3

Fig. 4.15. Frequency curve of circumference of mother rhizome.



According to this analysis, of the six growth characters, two exhibit skewness towards the distal side of the frequency distribution, whereas the other four exhibit skewness towards the proximal side of the distribution. All nine of the yield characters studied show skewness towards the proximal side of their distributions, indicating that recessive alleles are largely accumulated in their gene pools, which will seriously limit the crop yield due to the presence of higher frequency of recessive contributing factors.

The analyses show that the genetic base of *Curcuma zedoaria* in the study area is comparatively broad and the natural and locally cultivated populations of this species are not threatened by narrowing of genetic diversity. However, marginal crops such as *C. zedoaria*, are severely threatened in their natural habitats and traditional homestead habitats due to crop conversion, industrialised agriculture and shift to monocropping. It is important that the potential diversity of the species is maintained and the unexplored potential of this crop is available to future generations of humans to study and develop plant-based drugs and nutraceuticals that significantly raise the quality of human life.

Although the agronomic characters of the germplasm under study possess good genetic potential, there is relatively high frequency of recessive alleles in the germplasm and the parent populations from which the plants were collected, which limits the crop's yield significantly. It is therefore necessary to initiate further selection programmes in order to develop high yielding and improved varieties of this species. This will ensure that farmers can cultivate this species in an organized manner and are not forced to abandon the cultivation of this important species for economic reasons.

This method of analysis has previously been used to study the frequency distributions of phenotypes for genetic analysis in a variety of crops, including coffee (Raghu *et al*., 2003), *Cassia tora* (Chandramohanan and Mohanan,

2005), vanilla (Umamaheswari and Mohanan, 2004) and *Maranta arundinacea* (Shintu *et al*., 2016). Studies like these have become key to evaluating genetic diversity and initiating future breeding programmes.

4.1.2. Phenotypic and genotypic variability

In order to assess the extent of phenotypic and genotypic variability exhibited by the populations of zedoary in the study area, variability studies of 57 accessions of the species based on six growth and nine yield characters have been carried out. Analysis of variance revealed statistically significant differences among the studied accessions. The observations are listed in Table 4.16. and Table 4.17.

It was found that all the growth characters studied showed statistically significant variation at 1% level of significance. The mean value of plant height was 66.86 cm, with a range of 13 to 113 cm and a coefficient of variation of 33.36%. The range of number of tillers varied from 1 to 3, with a mean value of 1.20 and a coefficient of variation of 12.5%. Leaves per tiller ranged from 2.5 to 11 cm with a mean value of 6.79 and the coefficient of variation was 10.01%. With the mean value of 44.06 cm and coefficient of variation of 30.84%, leaf length ranged from 8.3 to 75.3 cm. Leaf breadth had a mean value of 6.50 cm, coefficient of variation of 32.15%, and a range of 1.83 to 11.16 cm. Leaf area ranged from 9.25 cm^2 to 422.05 cm^2, with a mean of 189.14 cm^2 and a coefficient of variation of 51.40 %. Leaf area had the highest coefficient of variation of all the growth characters examined, followed by plant height (33.36%). The number of tillers (10.01%) had the lowest coefficient of variation (Table 4.16).

Table 4.16. Genetic variability of the growth characters in *Curcuma zedoaria* studied.

Accession number	Plant height **	Number of tillers **	Number of leaves per tiller **	Leaf length **	Leaf breadth **	Leaf area **
CUW1	83.50±5.32	1.44±0.22	6.27±0.73	53.25±3.11	7.87±0.49	255.45±27.32
CUW2	92.38±2.90	1.22±0.11	7.05±0.46	58.57±1.61	8.09±0.44	285.36±13.76
CUW3	90.99±1.36	1.22±0.22	7.94±0.57	58.34±0.63	7.77±0.33	271.73±8.87
CUW4	83.16±1.74	1.22±0.22	8.05±0.28	55.23±2.94	7.73±0.53	258.45±30.56
CUW5	86.56±10.71	1.22±0.22	6.94±0.43	56.98±1.52	6.81±0.27	232.86±15.89
CUW6	87.16±1.50	1.22±0.11	7.11±0.29	55.9±0.24	7.78±0.34	261.94±10.35
CUW7	87.39±2.48	1.77±0.29	6.05±0.24	56.0±1.15	8.17±0.36	279±18.10
CUW8	81.11±2.41	1.44±0.11	6.50±0.10	51.38±0.20	7.84±0.37	242.31±10.54
CUW9	100.1±4.47	1.22±0.11	7.0±0.20	60.41±2.21	8.48±0.25	307.92±12.81
CUW10	95.55±1.62	1.44±0.11	6.55±0.79	59.38±0.84	8.80±0.17	309.84±4.21
CUW11	90.83±2.37	1.22±0.11	6.44±0.68	55.46±2.35	8.65±0.18	288.02±16.82
CUW12	87.10±2.06	1.44±0.11	5.88±0.28	53.62±1.33	8.15±0.43	262.44±13.62
CUW13	56.16±6.27	1.33±0.19	6.44±0.55	36.73±3.65	6.15±0.64	144.87±26.20
CUW14	75.42±4.79	1.33±0.19	6.16±0.17	52.23±6.70	7.22±0.23	235.4±30.80
CUW15	82.57±4.97	1.11±0.11	7.33±0.88	52.81±3.02	8.74±0.64	282.14±32.55
CUW16	75.83±3.87	1.33±0.19	6.83±0.48	50.45±1.59	8.83±0.64	267.73±27.25
CUW17	75.27±4.08	1.33±0.19	6.66±0.66	48±3.75	7.71±0.63	230.12±30.94
CUW18	90.11±2.84	1.22±0.22	7.55±0.55	57.59±2.05	8.06±0.26	279.34±15.28
CUW19	86.83±3.19	1.11±0.11	6.83±0.35	56.23±2.07	7.75±0.33	259.22±18.65
CUW20	78.83±4.67	1.22±0.22	6.66± 0.69	52.14±2.07	7.7±0.32	243.13±14.69

CUW21	72.82±10.28	1.11±0.11	7.11±0.49	54.73±5.71	8.49±0.86	286.90±54.50
CUW22	87.97±3.50	1.55±0.29	6.50±0.58	57.31±1.40	8.29±0.29	288.16±14.97
CUW23	90.11±1.22	1.33±0	6.50±0.54	58.95±1.21	7.92±0.45	280.76±16.69
CUW24	80.27±3.70	1.11±0.11	7.55±0.68	49.83±1.35	8.82±0.29	264.27±15.98
CUW25	90.66±2.55	1±0	7.22±0.68	57.07±2.26	8.87±0.39	301.05±28.08
CUW26	89.57±4.40	1.11±0.11	7.44±0.22	56.63±2.80	8.40±0.05	288.98±11.90
CUW27	90.81±5.17	1.11±0.11	7.44±0.40	58.01±4.02	9.26±0.28	314.19±41.68
CUW28	84.44±1.70	1±0	8.22±1.16	55.40±1.17	8.81±0.37	291.77±11.85
CUW29	80.33±3.51	1.22±0.11	6.55±0.14	51.09±2.85	7.79±0.63	242.65±30.46
CUW30	89.9±5.76	1±0	8.22±0.68	60.72±3.73	7.09±0.44	260.42±27.05
CUW31	37.24±1.53	1.22±0.11	7±0.58	24.76±0.29	3.63±0.17	54.47±7.82
CUW32	33.66±1.25	1.11±0.11	6.72±0.79	20.10±1.30	3.50±0.21	42.88±5.05
CUW33	30.33±0.17	1±0	7.89±0.11	19.81±0.16	3.45±0.04	41.23±1.02
CUW34	74.2±3.03	1.33±0	6.34±0.35	45.96±1.43	8.04±0.02	227.88±12.57
CUW35	33.22±1.22	1.22±0.22	6.63±1.05	21.48±0.43	3.48±0.17	45.58±3.16
CUW36	32.22±1.66	1.11±0.11	7.55±0.62	20.57±0.84	3.38±0.09	42.45±2.66
CUW37	28.5±1.76	1.11±0.11	7.55±0.68	18.51±1.02	3.08±0.32	35.97±5.65
CUW38	28.00±1.83	1.11±0.11	7.50±0.75	17.69±1.41	3.32±0.16	36.07±4.46
CUW39	30.61±0.40	1.22±0.22	7.33±0.66	19.14±0.48	3.03±0.12	35.37±0.64
CUW40	30.61±0.78	1.11±0.11	6.55±0.62	20.28±0.66	3.58±0.06	43.69±1.67
CUW41	64.60±4.25	1.11±0.11	6.16±0.68	43.67±3.21	6.82±0.13	184.79±10.93
CUW42	77.16±1.77	1.11±0.11	6.83±0.25	48.2±1.53	7.84±0.35	228.35±10.38
CUW43	75.63±3.16	1.2±0.20	6.92±0.55	52.5±2	7.76±0.14	255.85±3.18
CUW44	57.61±4.01	1.11±0.11	6.33±0.39	40.68±3.40	5.66±0.23	141.46±14.47

CUW45	41.83±2.78	1.11±0.11	5.05±0.48	33.15±2.43	4.22±0.57	86.46±16.96
CUW46	53.38±2.27	1.33±0.19	7.28±0.36	36.46±0.46	7.75±0.10	170.53±4.05
CUW47	60.11±1.38	1.33±0	6.94±0.14	39.46±0.59	7.94±0.66	191.38±16.07
CUW48	48.69±2.16	1.11±0.11	6.83±0.54	37±1.81	4.86±0.38	112.94±9.40
CUW49	48.61±2.77	1±0	6.44±0.40	33.81±1.03	4.39±0.32	90.46±9.36
CUW50	52.00±0.92	1±0	6.22±0.22	36.64±1.08	4.67±0.09	106.49±1.58
CUW51	43.55±2.48	1.22±0.11	5.17±0.44	31.21±2.07	4.05±0.33	80.65±13.34
CUW52	47.78±2.22	1.11±0.11	6.16±0.35	34.57±1.76	3.86±0.21	81.38±8.35
CUW53	48.27±3.87	1.33±0.19	5.78±0.14	35.06±2.18	4.32±0.45	93.48±14.95
CUW54	47.55±0.79	1±0	6.22±0.11	37.36±0.67	4.14±0.13	94.05±3.87
CUW55	46.00±1.00	1±0	6±0.39	35.85±0.34	3.94±0.25	85.41±6.03
CUW56	47.03±1.03	1.28±0.14	5.97±0.18	32.25±1.13	3.70±0.58	72.09±4.47
CUW57	47.72±2.72	1±0	6.44±0.22	34.84±1.54	3.91±0.14	83.58±6.95
Mean	66.86	1.20	6.79	44.06	6.50	189.14
Range	13-113	1-3	2.5-11	8.3-75.3	1.83-11.16	9.25-422.05
CV	33.36	12.5	10.01	30.84	32.15	51.40
SD	22.30	0.15	0.68	13.59	2.09	97.22
CD at 5%	9.92	0.08	1.45	6.25	1.01	50.39

**: Significant variation at 1% level.

Among yield characters, number of primary fingers ranged from 1 to 8 with a coefficient of variation of 20.60% and a mean value of 3.01. Length of primary finger varied between 2.1 and 28.7 cm with a mean of 12.74 cm and a coefficient of variation of 24.96%. The circumference of primary finger varied between 1.6 and 9 cm with a mean of 4.84 cm and a coefficient of variation of 21.49%. The mean size of secondary fingers was 16.77 cm with a coefficient of variation of 35.30%. The number of secondary fingers varied from 3 to 43. The length of secondary finger showed a mean value of 6.71 cm with a range of 1 to 18.7 cm and the coefficient of variation exhibited was 37.70%. The circumference of secondary finger ranged from 0.8 to 8.1 cm with a mean value of 3.79 cm and a coefficient of variation of 24.46%. The length of mother rhizome varied from 2 cm to 8.6 cm; the mean value was 4.46 cm with a coefficient of variation of 19.73%. Circumference of mother rhizome had a mean value of 7.73 cm and it ranged from 2.8 cm to 16.4 cm. The value of coefficient of variation was 31.55%. The range of yield per plant was 20 g to 635 g, with a coefficient of variation of 46.88%. The mean value for the character was 156.44 g (Table 4.17). Among the yield characters studied the length of the mother rhizome showed the lowest coefficient of variation, while yield per plant and the length of secondary finger showed the highest coefficient of variation (Table 4.17).

It can be concluded from the study that leaf area, yield per plant and length of secondary finger are the most variable characters in the *C. zedoaria* accessions studied. While carrying out selection, due consideration should be given to these characters. The studied characters are all statistically significant, indicating that they can be used for selecting promising genotypes as part of future improvement programmes.

Table 4.17. Genetic variability of the yield characters in *Curcuma zedoaria* studied.

Accession number	Number of primary fingers**	Length of primary finger** (cm)	Circumference of primary finger** (cm)	Number of secondary fingers**	Length of secondary finger** (cm)	Circumference of secondary finger** (cm)	Length of mother rhizome** (cm)	Circumference of mother rhizome** (cm)	Yield per plant** (g)
CUW1	3.33±0.39	15.86±1.16	5.33±0.32	22.89±2.59	7.77±1.10	3.87±0.18	4.58±0.31	7.66±0.30	216±48.65
CUW2	3.44±0.29	15.65±1.91	6.24±0.24	22.66±2.52	10.82±0.78	4.93±0.27	5.11±0.07	10.04±0.23	268.33±15.05
CUW3	3.77±0.11	17.60±0.17	5.99±0.35	26.88±1.90	9.93±0.35	4.54±0.37	4.64±0.27	9.35±0.93	324.44±10.30
CUW4	3.77±0.11	17.01±0.62	5.81±0.10	27.44±3.14	9.98±0.11	5.56±0.53	5.48±0.17	8.72±0.34	263.33±18.76
CUW5	3.66±0.34	17.62±0.61	4.79±0.17	30.22±1.83	10.16±1.38	3.96±0.16	4.84±0.06	8.17±0.27	195±11.1
CUW6	2.66±0.20	14.66±1.02	5.79±0.12	17.22±0.29	8.97±1.20	4.63±0.02	5.33±0.34	9.22±0.39	216.66±25.10
CUW7	3.22±0.11	14.81±0.65	4.96±0.52	19.55±1.64	8.75±0.60	4.25±0.29	4.30±0.25	7.34±0.61	202.78±19.80
CUW8	3.11±0.11	15.32±0.22	5.2±0.06	23.33±1.35	9.69±0.73	4.22±0.20	4.27±0.20	4.27±0.20	172.22±10.61
CUW9	4.0±0.20	20.18±4.03	6.76±0.60	28.22±3.53	13.38±1.72	6.17±0.62	6.24±0.28	12.19±0.50	362.78±62.68
CUW10	4.33±0.19	13.94±0.54	5.46±0.35	18.44±0.59	7.41±0.86	4.71±0.36	4.96±0.52	9.63±0.35	235.55±18.08
CUW11	4.10±0.51	14.51±1.76	4.94±0.42	25.55±2.99	6.84±0.79	4.16±0.14	4.13±0.30	7.17±0.45	186.67±26.86
CUW12	3.55±0.59	14.23±0.49	5.23±0.06	23.33±3.87	7.07±0.66	3.75±0.09	4.57±0.25	8.55±0.49	191.66±24.58
CUW13	2.11±0.23	12.84±2.14	4.12±0.50	10.89±1.57	5.52±1.12	3.36±0.11	3.38±0.34	5.11±0.48	87±24.30
CUW14	3.11±0.45	14.08±2.52	4.91±0.95	17.22±3.57	7.12±1.71	3.29±1.05	4.32±0.47	7.46±1.02	154.44±16.83
CUW15	3.33±0.34	14.25±1.40	5.7±0.40	18.00±1.90	7.78±1.02	4.44±0.49	4.74±0.45	8.93±0.76	195±18.95

CUW16	3.66±0.69	15.13±0.84	4.91±0.38	20.55±3.77	8.01±0.44	3.95±0.36	4.15±0.31	7.67±0.61	182.22±35.14
CUW17	3.22±0.77	13.24±1.31	5.16±0.31	23.89±6.60	7.58±0.91	4.22±0.24	4.31±0.11	8.13±0.50	186.66±30.96
CUW18	2.99±0.34	14.79±0.71	6.33±0.24	17.89±1.94	8.18±1.03	4.77±0.17	5.53±0.50	9.89±0.68	235±4.42
CUW19	4.11±0.45	12.01±1.43	5.09±0.16	18.66±1.39	7.79±1.86	4.07±0.30	4.93±0.05	9.28±0.72	182.78±31.43
CUW20	3.11±0.59	12.92±0.31	5.43±0.32	17.44±4.14	9.08±1.06	4.45±0.34	5.66±0.03	11.17±1.44	199.44±16.75
CUW21	2.77±0.59	11.84±2.63	5.58±0.86	15.77±4.06	5.91±1.78	4.51±0.76	5.89±0.30	10.43±0.47	174.44±45.62
CUW22	3.44±0.29	14.20±1.24	5.46±0.09	23.66±2.34	8.25±0.31	3.84±0.03	4.87±0.10	8.93±0.27	195.55±3.65
CUW23	3.22±0.11	14.40±0.69	5.10±0.35	21.66±0.39	7.92±0.34	4.34±0.25	4.64±0.23	7.75±0.46	184.44±23.61
CUW24	3.44±0.59	15.91±0.35	5.24±0.10	25.78±6.20	8.96±0.83	4.17±0.29	5.06±0.55	9.48±1.14	185.55±38.80
CUW25	3.55±0.55	13.66±0.51	5.07±0.28	18.11±3.13	4.83±0.44	3.79±0.31	4.42±0.21	8.28±0.39	151.66±15.14
CUW26	3.44±0.40	15.16±1.27	5.73±0.38	18.55±2.70	9.40±1.09	4.80±0.37	5.26±0.20	10.14±0.25	222.77±10.96
CUW27	2.77±0.49	13.59±0.58	5.04±0.53	16.78±1.31	8.96±0.21	4.50±0.22	4.71±0.49	9.05±0.64	203.33±34.37
CUW28	3.77±0.11	13.67±2.05	6.04±0.23	14.88±2.22	7.63±0.81	4.51±0.14	5.07±0.29	10.35±0.45	163.89±11.49
CUW29	3.59±0.26	13.95±0.78	5.23±0.32	18.77±2.57	4.49±0.94	3.58±0.24	4.22±0.10	8.20±0.28	131.11±16.42
CUW30	2.44±0.49	14.17±0.64	5.99±0.32	14.11±3.05	5.02±0.49	4.38±0.26	5.73±0.22	10.93±0.18	187.22±43.20
CUW31	2.83±0.25	15.31±0.29	6.18±0.43	16.72±1.11	6.81±0.34	4.54±0.09	5.69±0.14	10.69±0.36	218.89±10.61
CUW32	3.0±0.39	15.35±0.62	6.07±0.58	20.44±3.59	10.08±0.45	4.56±0.45	5.03±0.15	9.72±0.45	208.84±31.39
CUW33	3.44±0.11	13.60±0.71	4.66±0.24	14.89±0.29	6.86±0.30	4.07±0.20	3.44±0.06	5.78±0.21	160.22±11.08
CUW34	2.67±0.34	13.15±0.45	4.92±0.21	15.00±2.31	7.71±0.64	3.71±0.20	4.14±0.11	7.35±0.53	74.2±3.03
CUW35	2.33±0.19	12.54±1.36	5.86±0.50	15.66±1.53	8.11±0.65	4.01±0.14	5.53±0.39	10.65±0.55	189.98±30.29

CUW36	2.66±0	13.28±0.95	5.45±0.14	13.33±1.17	7.24±1.08	3.99±0.24	4.59±0.16	9.70±0.16	225±29.09
CUW37	2.66±0.34	13.04±1.44	4.59±0.18	12.11±2.63	6.24±1.58	3.83±0.60	5.74±0.62	9.53±0.66	146.11±37.24
CUW38	2.66±0.20	11.79±0.83	4.62±0.35	14.78±2.15	4.97±0.49	3.57±0.20	4.43±0.15	7.86±0.32	127.22±12.05
CUW39	2.0±0.20	11.74±1.51	4.14±0.53	11.55±0.29	4.79±1.47	3.35±0.52	4.65±0.18	8.65±0.75	113.33±17.42
CUW40	2.55±0.29	12.15±0.34	4.98±0.14	12.22±1.13	8.7±0.84	4.25±0.61	5.52±0.27	11.56±0.57	162.89±16.10
CUW41	2.89±0.29	12.49±0.43	4.97±0.12	12.88±1.18	8.33±1.79	3.99±0.53	4.82±0.13	9.87±0.18	140±7.65
CUW42	3.44±0.22	13.42±0.9	4.66±0.17	22.33±2.35	6.84±0.40	3.83±0.14	3.91±0.12	6.79±0.12	77.16±1.77
CUW43	3±0.58	14.67±1.12	5.33±0.22	14.66±2.89	6.29±0.66	3.98±0.23	4.68±0.33	8.46±0.60	142.75±21.34
CUW44	2.47±0.33	11.27±1.33	3.29±0.34	10.81±1.82	4.33±0.23	2.98±0.37	3.67±0.24	2.89±1.06	110.67±10.49
CUW45	1.55±0.29	7.41±2.00	3.17±0.50	7.55±2.55	2.92±0.81	2.69±0.80	3.32±0.34	4.22±0.29	49.44±2.42
CUW46	3.21±0.55	9.09±0.43	4.08±0.20	13.11±0.87	2.95±0.33	2.04±0.26	3.99±0.20	4.98±0.24	72.78±9.70
CUW47	3.22±0.22	8.23±0.55	4.33±0.18	16.44±1.25	2.55±0.31	2.46±0.40	4.46±0.16	5.40±0.10	97.77±4.01
CUW48	2.72±0.15	7.82±0.83	3.46±0.32	11.89±0.73	3.35±0.35	2.6±0.16	3.07±0.32	4.58±0.32	63.89±6.55
CUW49	3.00±0.66	6.24±0.31	2.94±0.05	7.89±1.42	2.87±0.22	1.75±0.20	3.64±0.14	4.74±0.10	45.55±6.42
CUW50	2.44±0.45	10.16±0.93	3.91±0.13	13.44±2.30	3.56±0.32	2.93±0.10	3.33±0.02	4.32±0.38	75.55±5.48
CUW51	1.77±0.29	9.55±0.71	3.55±0.11	7.55±1.54	4.53±0.77	2.93±0.13	2.96±0.06	4.01±0.24	57.22±3.90
CUW52	2.55±0.29	8.64±0.85	3.08±0.17	12.00±2.53	4.14±0.81	2.72±0.13	2.73±0.13	3.82±0.20	58.33±8.57
CUW53	2.77±0.11	6.44±0.69	3.13±0.11	8.44±0.95	3.22±0.58	2.35±0.31	3.01±0.23	5.18±0.61	42.22±5.31
CUW54	2.00±0	7.16±0.15	2.32±0.14	7.11±0.11	2.44±0.12	1.99±0.02	3.10±0.02	3.45±0.06	43.33±1.67
CUW55	2.78±0.23	7.83±0.57	3.25±0.23	10.44±1.31	2.52±0.15	2.5±0.18	3.19±0.10	4.68±0.09	61.66±0.97

CUW56	2.23±0.18	5.19±0.31	2.65±0.20	5.71±0.45	2.73±0.37	1.67±0.06	3.5±0.25	4.39±0.30	37.55±2.17
CUW57	1.77±0.11	7.63±0.50	3.36±0.29	8.55±1.16	4.15±0.62	2.75±0.43	2.79±0.20	4.01±0.44	56.66±9.19
Mean	3.01	12.74	4.84	16.77	6.71	3.79	4.46	7.73	156.44
Range	1-8	2.1-28.7	1.6-9	3-43	1-18.7	0.8-8.1	2-8.6	2.8-16.4	20-635
CV	20.60	24.96	21.49	35.30	37.70	24.46	19.73	31.55	46.88
SD	0.62	3.18	1.04	5.92	2.53	0.93	0.88	2.44	73.34
CD at 5%	1.01	3.35	0.96	6.91	2.47	1.00	0.78	1.46	62.14

**: significant at 1% level.

The phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV) and heritability (broad sense) of characters can be used to estimate the degree to which the environment impacts the agronomic characters. In the current study, PCV was larger than GCV for all characters, signifying the additive nature, polygenic control and varying degrees of environmental impacts on the characters under investigation. The PCV and GCV of the observed characters ranged from 14.80 % to 53.17 % and 4.17 % to 50.49 %, respectively. The highest PCV and GCV were recorded for leaf area followed by yield per plant. The lowest PCV was observed for the number of leaves per tiller and the lowest GCV for number of tillers (Table 4.18).

Leaf area showed a PCV of 53.17% and marked the highest among the six growth characters studied. The lowest PCV was obtained for number of leaves per tiller (14.80%). Plant height showed a GCV of 32.92% and a PCV of 34.19% and in the case of leaf breadth those were 31.69% and 33.08% respectively. PCV for leaf length was 31.68% and for number of tillers it was 20.83% while GCV of these characters were 30.41% and 4.17% respectively. The GCV of number of leaves per tiller and leaf area were 6.48% and 50.49%. Of the nine yield characters studied the maximum GCV and PCV were recorded for yield per plant followed by length of secondary finger and number of secondary fingers. GCV for yield per plant was 44.64%, for length of secondary finger it was 35.32% and for number of secondary fingers it was 31.96% whereas PCVs were 51.07%, 42.18% and 41.09% respectively. The lowest GCV was shown by number of primary fingers (16.61%) and the lowest PCV was observed for length of mother rhizome (21.75%).

In comparison to growth characters, both PCV and GCV values were relatively higher among yield characters. Characters with high differences between PCV and GCV indicate relatively higher environmental influence on the phenotypic expression of the character, while characters with low difference

indicate limited environmental influence. It makes it possible to directly select genotypes for further crop improvement based on their phenotypic performance, if their heritability and genetic advance values are also high. Among the tested characters, the differences between PCV and corresponding GCV were higher for number of tillers, number of primary fingers and number of secondary fingers, while in the case of the other characters those were lower.

Similar studies on genetic variability of different species of economically important plants have been done by earlier workers like Shintu *et al*. (2016) in West Indian arrowroot, Mishra *et al*. (2015) in strawberry, Jayasree *et al*. (2014a) in mango ginger, Radhakrishnan *et al*. (2006a) in cardamom, Lal *et al*. (2015) in turmeric, Soorya *et al*. (2016a) in *Curcuma aeruginosa* and Azimi *et al*. (2017) in wheat.

4.1.3. Heritability (broad sense) of the agronomic characters

Heritability is the measure of inherited portion of variability (Khan, 2000). An evaluation of heritability (in broad sense) provides information about a specific trait that will be passed on to successive generations and is important for plant breeders when choosing parent plants for crop improvement programmes. Heritability in broad sense refers to the contribution of all forms of gene actions of a character including dominant, additive, maternal, paternal and epistatic effects to the phenotypic variance of a population. The value of broad sense heritability also reveals how much environmental influences have an impact on that character. A higher heritability value indicates a very low extent of the environment's influence on the character.

Table 4.18. Genotypic variance, phenotypic variance, GCV, PCV, heritability and genetic advance in the case of the growth and yield characters of *Curcuma zedoaria*.

Character	Genotypic variance	Phenotypic variance	Genotypic coefficient of variation	Phenotypic coefficient of variation	Heritability (broad sense) (%)	Genetic advance (%)
Plant height (cm)**	484.32	5522.76	32.92	34.19	92.65	65.26
Number of tillers**	0.003	0.25	4.17	20.83	4.76	2.04
Number of leaves per tiller**	0.19	1.00	6.48	14.80	18.81	5.71
Leaf length (cm)**	179.55	194.78	30.41	31.68	92.18	60.17
Leaf breadth (cm)**	4.24	4.64	31.69	33.08	91.38	62.26
Leaf area (cm^2)**	9120.56	10111.95	50.49	53.17	90.20	98.79
Yield per plant (g)**	4875.91	6383.67	44.64	51.07	76.38	80.36
Number of primary fingers**	0.25	0.65	16.61	26.91	38.46	21.32
Length of primary finger (cm)**	8.66	13.03	23.08	28.34	66.46	38.79
Circumference of primary finger (cm)**	0.96	1.32	20.25	23.76	72.73	35.60
Number of secondary fingers**	28.77	47.42	31.96	41.09	60.67	51.35
Length of secondary finger (cm)**	5.63	8.01	35.32	42.18	70.29	61.07
Circumference of secondary finger (cm)**	0.73	1.12	22.43	27.97	65.18	37.55
Length of mother rhizome (cm)**	0.70	0.94	18.83	21.75	74.47	33.36
Circumference of mother rhizome (cm)**	5.69	6.52	30.92	32.99	87.27	59.31

Fifteen agronomic characters of zedoary have been studied presently for broad sense heritability. The heritability values of growth characters ranged from 92.65% for plant height to 4.76% for number of tillers (Table 4.18). Leaf length showed 92.18% heritability, leaf breadth exhibited 91.38% heritability, leaf area showed 90.20% heritability and the number of leaves per tiller exhibited a heritability of 18.81%. Heritability of the yield characters ranged from 38.46% (number of primary fingers) to 82.27% (circumference of mother rhizome). Number of secondary fingers showed the heritability of 60.67%, circumference of secondary finger exhibited a heritability of 65.18%, length of primary fingers showed 66.46 % of heritability, length of secondary finger exhibited 70.29% of heritability, circumference of primary finger showed a heritability of 72.73%, length of mother rhizome exhibited 74.47% of heritability and yield per plant showed 76.38% of heritability.

Heritability is considered as low below 30%, medium between 30% and 60%, and high above 60% by Babu *et al*. (2012). According to this classification, four growth characters, plant height, leaf length, leaf breadth, and leaf area showed the highest heritability. In contrast, number of tillers and number of leaves per tiller showed the lowest heritability. Among the nine yield characters, eight including yield per plant, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome showed high heritability. Number of primary fingers, on the other hand, showed only medium heritability. The present study indicates that plant height, leaf length, leaf breadth, leaf area and circumference of mother rhizomes with very high heritability will respond better to selection and could be improved through direct selection.

There have been similar studies carried out by earlier workers on heritability and the influence of environment on phenotypic variation in

different crops such as cowpea (Omoigui *et al*., 2006), coriander (Tripathi *et al.,* 2000), brinjal (Shekar *et al*., 2012), soybeans (Johnson *et al*., 1955), coffee (Nikhila *et al*., 2002) and bottle gourd (Damor *et al*., 2016).

4.1.4. Genetic advance

Genetic advance is another selection parameter which indicates the quantum of improvement of a character that is possible through selection (Allard, 1960). Estimating genetic advance along with heritability is helpful in predicting the gain under selection in crop improvement programmes. The genetic advance of *Curcuma zedoaria* analysed for six growth and nine yield characters in this study ranged from 2.04% (number of tillers) to 98.79% (leaf area).

Among the growth characters, the highest genetic advance was recorded for leaf area (98.79%) followed by plant height (65.26%), leaf breadth (60.26%), leaf length (60.17%), number of leaves per tiller (5.71%) and the lowest genetic advance by number of tillers (2.04%). Among the yield characters, the highest genetic advance was exhibited by yield per plant (80.36%) followed by length of secondary finger (61.07%) and the circumference of mother rhizome (59.31%). The lowest genetic advance was shown by number of primary fingers (21.32%). Values of genetic advance observed for other characters were 51.35%, 38.79%, 37.55%, 35.60% and 33.36% for number of secondary fingers, length of primary finger, circumference of secondary finger, circumference of primary finger and the length of mother rhizome respectively.

The selection potential of characters with high heritability and genetic advance is higher than those with low heritability and genetic advance. Hence genetic advance is a great selection parameter for breeders when developing superior varieties through a selection programme.

Earlier workers have worked on genetic advance in crops like banana (Kavitha *et al*., 2008), elephant foot yam (Anil *et al*., 2011), sesame (Kebede *et al*., 2014), rice (Adhikari *et al*., 2018), small cardamom (Hrideek *et al*., 2015) and linseed (Singh *et al*., 2019). The study of genetic advance of agronomic characters in *C. zedoaria* is new to science, and this research will provide better guidance on genetic gains possible in this crop through selection.

All agronomic characters of *C. zedoaria* examined show significant variation between accessions, indicating that the crop has a strong genetic base in the experimental area. It is very important to take advantage of this variability to conserve and improve the species due to its value as a medicinal plant.

4.2. Correlation of agronomic characters

Characters controlled by polygenes show varying levels of interrelationships due to common sharing of genes between the characters. The relationship between two or more variables under consideration is called correlation. Correlation analysis is an excellent tool to determine the degree of relationship between two or more variables, as well as the direction of that relationship. During the selection process, breeders always consider yield related characteristics, and it is crucial to assess the relative contribution of such characters to yield in both direct and indirect ways.

Accordingly, correlation analysis has been conducted using the data recorded from the experimental population raised from fifty seven accessions of *C. zedoaria* to find out how the fifteen agronomic characters of zedoary are related. In the present experiment, seven characters, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger and length of secondary finger showed significant correlation with the maximum number of characters and number of tillers revealed

significant correlation with ten characters (Tables 4.19 and 4.20). Correlation studies of various growth and yield characters of *C. zedoaria* is new to science. Thus, the present experiment should provide a better understanding of the association between different agronomic characteristics in this crop plant.

Plant height showed significant correlation with 13 other characters such as number of tillers, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and the circumference of mother rhizome. It indicates that these characters possess some inherent interrelationship with plant height.

Number of leaves per tiller exhibited significant correlation with 12 other characters namely number of tillers, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome. Number of tillers showed significant correlation with ten characters namely plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger and the length of secondary finger.

Leaf length showed significant correlation with plant height, number of tillers, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome. Leaf area exhibited significant correlation with all the 14 characters such as plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area,

yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome.

Leaf breadth showed significant correlation with plant height, number of tillers, number of leaves per tiller, leaf length, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome.

Circumference of mother rhizome showed significant correlation with thirteen characters such as plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome. Length of mother rhizome revealed significant correlation with plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger and circumference of mother rhizome. Yield per plant exhibited significant correlation with all the other fourteen characters like plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome.

Number of secondary fingers revealed significant correlation with plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, length of primary finger,

circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome. Length of primary finger showed significant correlation with plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome. Length of secondary finger showed significant correlation with plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome.

Circumference of secondary finger revealed significant correlation with thirteen characters like plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, length of mother rhizome and circumference of mother rhizome. Number of primary fingers showed significant correlation with plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome. Circumference of primary finger exhibited significant correlation with thirteen other characters like plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome.

Table 4.19. Correlation of agronomic characters in the case of *Curcuma zedoaria* studied.

Characters	Plant height	Number of tillers	Number of leaves per tiller	Leaf length	Leaf breadth	Leaf area	Yield per plant	Number of primary fingers	Number of secondary fingers	Length of primary finger	Circumference of primary finger	Length of secondary finger	Circumference of secondary finger	Length of mother rhizome	Circumference of mother rhizome
Plant height	1														
Number of tillers	0.338428*	1													
Number of leaves per tiller	0.195582	-0.3011*	1												
Leaf length	0.984894*	0.297355*	0.149751	1											
Leaf breadth	0.931577*	0.364298*	0.241755*	0.912625*	1										
Leaf area	0.979212*	0.341788*	0.244655*	0.968117*	0.977508*	1									
Yield per plant	0.562078*	0.24077*	0.509825*	0.484811*	0.488487*	0.554535*	1								
Number of primary fingers	0.67024*	0.257408*	0.372028*	0.60466*	0.680941*	0.677569*	0.615611*	1							
Number of secondary fingers	0.683243*	0.367683*	0.341234*	0.606109*	0.647523*	0.667106*	0.767406*	0.770118*	1						
Length of primary finger	0.592264*	0.268539*	0.47728*	0.504353*	0.551742*	0.591734*	0.876551*	0.611892*	0.842495*	1					
Circumference of primary finger	0.535383*	0.19022	0.563894*	0.4528*	0.537099*	0.565293*	0.884836*	0.566452*	0.702082*	0.866833*	1				
Length of secondary finger	0.530393*	0.271844*	0.35452*	0.444494*	0.473516*	0.518368*	0.864017*	0.540405*	0.766054*	0.875909*	0.810519*	1			
Circumference of secondary finger	0.547751*	0.132133	0.528114*	0.470695*	0.502488*	0.560137*	0.900416*	0.54722*	0.713052*	0.894039*	0.913651*	0.888697*	1		
Length of mother rhizome	0.376563*	0.093285	0.554451*	0.314021*	0.381564*	0.415378*	0.778784*	0.409466*	0.520965*	0.690478*	0.850071*	0.688232*	0.790912*	1	
Circumference of mother rhizome	0.359634*	0.03737	0.534166*	0.286975*	0.349743*	0.3934*	0.778524*	0.435114*	0.496894*	0.691616*	0.868622*	0.711087*	0.811455*	0.92*	1

*: significant at 5% level

Table 4.20. Details of the characters correlated.

Character	Number of characters showing significant correlation	Characters correlated
Plant height	13	Number of tillers, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Number of tillers	10	Plant height, Number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, length of secondary finger.
Number of leaves per tiller	12	Plant height, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Leaf length	13	Plant height, number of tillers, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Leaf breadth	14	Plant height, number of tillers, number of leaves per tiller, leaf length, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.

Leaf area	14	Plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Yield per plant	14	Plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Number of primary fingers	14	Plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Number of secondary fingers	14	Plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Length of primary finger	14	Plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome circumference of mother rhizome.
Circumference of primary finger	13	Plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary

		fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Length of secondary finger	14	Plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
Circumference of secondary finger	13	Plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, length of mother rhizome, circumference of mother rhizome.
Length of mother rhizome	13	Plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, circumference of mother rhizome.
Circumference of mother rhizome	13	Plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome.

Considering the present study, all the studied characters contribute to the yield of *C. zedoaria* and therefore plant breeders can work with them to improve zedoary varieties.

Researchers have previously investigated the correlation between agronomic characters in different crops. In wheat, Sokoto *et al*. (2012) found

that plant height, number of spikes, number of spikelets per spike, leaf area index, crop growth rate, number of grains per spike, total aerial phytomass yield, grain yield, harvest index and 1000 grain weight were the major contributors to grain yield due to their high correlation, and as a result, direct selection for these characters should be a top priority for plant breeders. Studies of Vijaya (2009) in turmeric showed that plant height, weight of mother rhizome, number of leaves, number of secondary fingers, leaf length, leaf breadth, girth of mother rhizome, number of primary fingers and weight of secondary finger were positively correlated with yield under coconut shade condition. Studies on correlation were also conducted by Walle *et al*. (2018) in cowpea, Paul and Bari (2013) in elephant foot yam, Rohman *et al*. (2003) in mung bean, Khan *et al*. (2009) in pointed guard, Taha *et al*. (2003) in *Cucumis*, Tena *et al*. (2016) in sugarcane and Rao *et al*. (2008) in *Curcuma amada*.

4.3. Character association

Due to the influence of the same sets of alleles on different characters, polygenic characters exhibit varying degrees of association among themselves. Such characters can be grouped based on their association with each other. This is an effective method for grouping of variables, reducing the inconvenience of managing a large number of variables so as to find out lead characters that can be used in conducting breeding experiments (Nikhila *et al*., 2008). Factor analysis is an effective method for determining character association, reducing data by identifying lead characters and grouping different variables into different factors. Lead characters could be defined as those with higher factor loading. Based on this, promising genotypes could be selected in a way that other agronomic characters associated with the lead characters would also be selected automatically (Hrideek *et al*., 2008).

Character association in *C. zedoaria* has been analysed with the help of factor analysis using fifteen variables. Three factors with Eigen values greater than one could be extracted by the analysis. The fifteen agronomic characters of *C. zedoaria* under study could be classified based on their factor loadings

into two factor groups (Tables 4.21 to 4.23). The first factor group consisted of fourteen variables such as plant height, number of leaves per tiller, number of primary fingers, yield per plant, leaf length, leaf breadth, leaf area, circumference of mother rhizome, number of secondary fingers, length of secondary finger, length of mother rhizome, length of primary finger, circumference of secondary finger and circumference of primary finger. The second factor group was occupied by one variable: the number of tillers.

Table 4.21. Factor analysis in the case of *Curcuma zedoaria* – Factor loadings.

Characters	Factor I	Factor II
Plant height	**-0.797462**	-0.551170
Number of tillers	-0.302264	**-0.426869**
Number of leaves per tiller	**-0.503986**	0.466086
Leaf length	**-0.727442**	-0.604942
Leaf breadth	**-0.771250**	-0.550792
Leaf area	**-0.810158**	-0.531359
Yield per plant	**-0.901459**	0.264853
Number of primary fingers	**-0.753553**	-0.224172
Number of secondary fingers	**-0.857825**	-0.109344
Length of primary finger	**-0.904189**	0.179063
Circumference of primary finger	**-0.901615**	0.320560
Length of secondary finger	**-0.855075**	0.222244
Circumference of secondary finger	**-0.898083**	0.314990
Length of mother rhizome	**-0.768486**	0.458947
Circumference of mother rhizome	**-0.763184**	0.493237

Table 4.22. Factor analysis in the case of *Curcuma zedoaria* – Eigen values, percentage of total variance.

Factors	**Eigen value**	**% of total variance**	**Cumulative Eigen value**	**Cumulative % of variance**
1	9.219195	61.46130	9.21920	61.46130
2	2.524616	16.83077	11.74381	78.29208

Table 4.23. Factor analysis in the case of *Curcuma zedoaria* - characters showing association as per factor analysis.

Factors	Characters
I	Plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, circumference of mother rhizome.
II	Number of tillers.

The first factor included 14 characters such as plant height, number of leaves per tiller, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome with factor loadings of -0.797462, -0.503986, -0.727442, -0.771250, -0.810158, -0.901459, -0.753553, -0.857825, -0.904189, -0.901615, -0.855075, -0.898083, -0.768486 and -0.763184 respectively. There was only one character associated with the second factor group showing higher factor loading when compared with the other factor group namely number of tillers with a factor loading of -0.426869.

Characters belonging to the same factor group share common alleles to a significant extent in their expression, and those with the highest factor loadings are said to be the lead characters. By improving these lead characters, all its associated characters will also get improved. It is clear from this analysis that the lead characters in the first factor are length of primary finger, circumference of primary finger and the yield per plant, which have the highest

factor loadings. In the second factor, number of tillers can be considered the lead character. The improvement of these lead characters will simultaneously result in the improvement of the other agronomic characters associated to them and hence significant consideration should be given to them while practising selection and other crop improvement programmes.

Characters like length of primary finger, circumference of primary finger, yield per plant and circumference of secondary finger with higher factor loading should be given adequate consideration for selection and subsequent crop improvement programmes in *C. zedoaria* so that the bulk of the characters for analysis could be reduced without affecting the outcome of research. Studies on the association of agronomic characters in different crops are vital for identifying relationships between quantitative morphometric characters. This is because such studies could provide genetic information for further breeding and improvement programmes.

The percentage of variance contributed by the first factor is 61.46% while the second factor contributes 16.83% of the variance. On the basis of the characters studied, these two factors cumulatively contribute 78.29% of the total variance of the present experimental population (Table 4.22).

In order to group variables and to analyse character association prevailing in crop plants, factor analysis has been widely used by earlier researchers, including Umamaheswari and Mohanan (2011) in vanilla, Radhakrishnan *et al*. (2004) and Hrideek *et al*. (2008) in small cardamom, Denton and Nwangburuka (2011) in *Solanum anguivi,* Soorya *et al*. (2018) in *Curcuma aeruginosa* and Yol *et al*. (2010) in sesame.

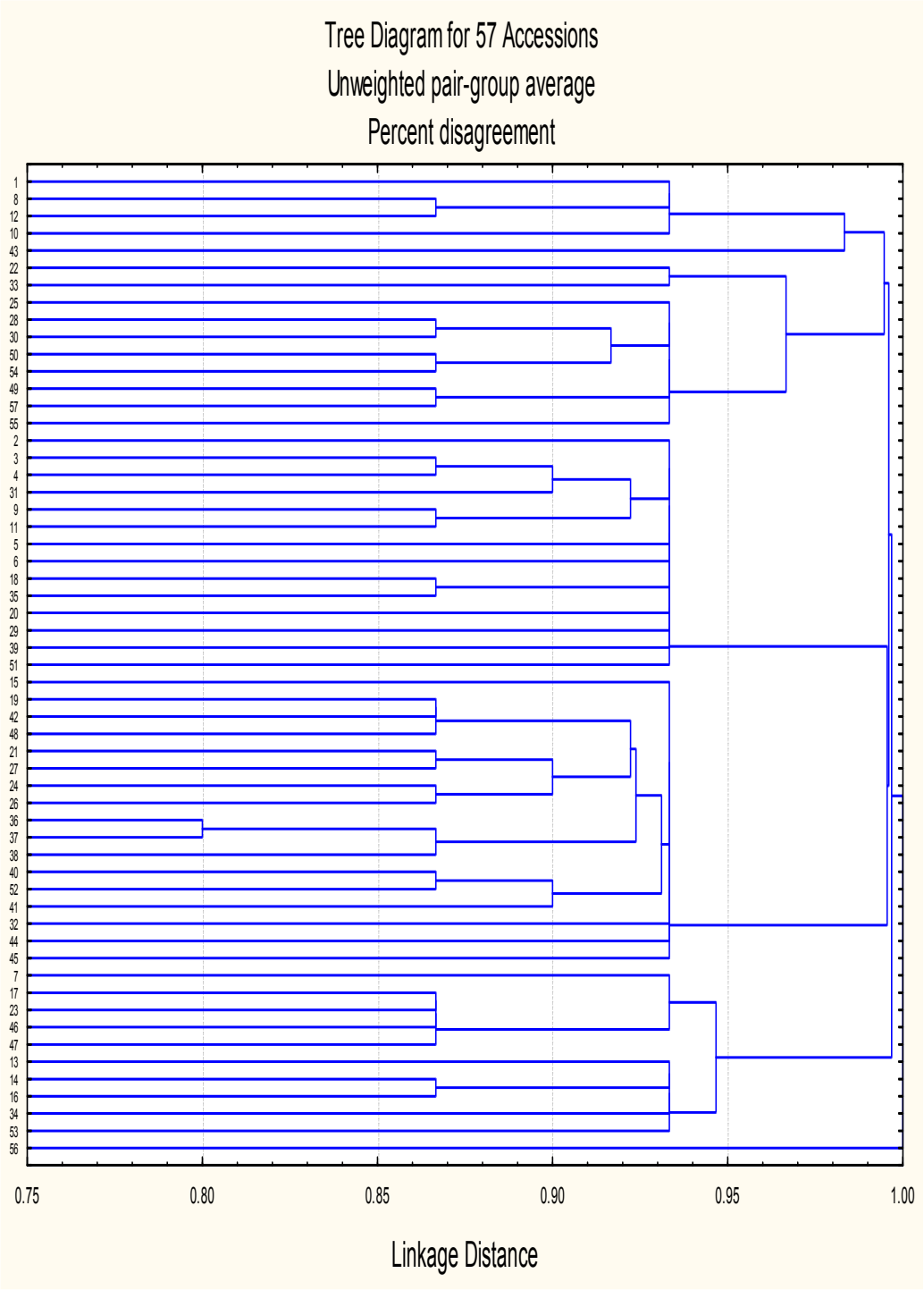
4.4. Genetic divergence

Due to the similarities and differences in their genetic constitution, different genotypes of a plant species display varying degrees of genetic divergence. Analysis of genetic divergence assesses the extent of genetic diversity present in the selected genotypes, which facilitates the process of choosing diverse genotypes as parents for future breeding programmes. Considering this, the genetic divergence of fifty seven accessions of *C. zedoaria* has been explored using STATISTICA software, following the method of UPGMA (Unweighted Pair Group Method with Arithmetic Mean) to determine distances and closeness pertaining among the accessions based on fifteen agromorphometric characters.

Cluster analysis grouped the entire accessions into two major clusters at a linkage distance of 0.999 (Fig 4.16). Cluster I is occupied by fifty six accessions and Cluster II is occupied by a single accession. The first cluster is bifurcated again into two sub clusters at a linkage distance of 0.998. The small cluster has 10 accessions and the large cluster has 46 accessions. The smaller cluster splits in to two at a linkage distance of 0.948. Each subcluster splits again at a linkage distance of 0.934.

As far as the characters subjected to the study are considered, genotypes CUW 36 and CUW 37 are genetically more related. This group belongs to the first cluster and these two bifurcate at the linkage distance of 0.80. There were genotypes from all the six districts namely Kottayam, Ernakulam, Alappuzha, Thrissur, Kozhikode and Malappuram in Cluster I. The genotype in Cluster II was from Thrissur district. As Cluster I contain genotypes collected from different geographical areas in different districts, it is clear that geographical separation is not a major criterion in determining genetic distance and closeness between the accessions studied. There is higher degree of similarity between genotypes belonging to the same cluster, which indicates that they show genetic

Fig. 4.16. Dendrogram showing the diversity of the fifty-seven accessions of *Curcuma zedoaria* studied.



proximity. Genotypes belonging to different clusters are genetically distant from each other with higher levels of genetic divergence in their genetic makeup.

Accessions that are distantly related can be regarded as genetically diverse. Therefore, there is a greater opportunity to select genetically divergent genotypes for crossing in order to raise promising and improved planting materials. Since the accessions showing closeness might have developed from similar parental lines there is lesser scope for selection process.

The present study reveals the genetic distances and genetic affinity between the different accessions of *C. zedoaria* collected. These results indicate that majority of the genotypes collected presently belong to the same cluster, indicating limited genetic variability. It may be difficult for breeders to develop a superior variety of crop due to its limited variability. If adequate care is not taken to conserve the existing diversity of *C. zedoaria*, loss of genetic diversity will prove critical.

Table 4.24. Clustering of the genotypes studied in *Curcuma zedoaria*.

Cluster Number	**Sub cluster Number**	**Accessions**
I	IA	CUW 7, CUW 17, CUW 23, CUW 46, CUW 47, CUW 13, CUW14, CUW 16, CUW 34, CUW 53
	IB	CUW 1, CUW 8, CUW 12, CUW 10, CUW 43, CUW 22, CUW 23, CUW 25, CUW 28, CUW 30, CUW 50, CUW 54, CUW 49, CUW 57, CUW 55, CUW2, CUW 3, CUW 4, CUW 31, CUW 9, CUW 11, CUW 5, CUW 16, CUW 18, CUW 35, CUW 20, CUW 29, CUW39, CUW 51, CUW 15, CUW 19, CUW 42, CUW 48, CUW 21, CUW 27, CUW 24, CUW 26, CUW 36, CUW 37, CUW 38, CUW 40, CUW 52, CUW 41, CUW 32, CUW 44, CUW 45
II		CUW 56

Earlier workers used cluster analysis to study genetic divergence in different crops like chilli (Janaki *et al*., 2016), tomato (Reddy *et al*., 2013), cardamom (Radhakrishnan *et al*., 2006b), chick pea (Syed *et al*., 2012) and wheat (Sharma *et al*., 2018). Studies like these have proved useful in determining the genetic distance and genetic closeness of different genotypes of crops. Based on the present study, it has been possible to determine the extent of genetic distance between different accessions of zedoary collected.

4.5. Performance analysis of the *Curcuma zedoaria* Rosc. accessions collected

Performance analysis of fifty seven *C. zedoaria* accessions has been done currently based on the performance indices of major growth and yield characters. For further crop improvement programmes, the most promising genotypes with the desired features can be identified using the cumulative performance index calculated as described earlier. Among the fifty seven accessions of *C. zedoaria*, accession number CUW 9 ranked first with a cumulative performance index of 22.76. CUW 3 and CUW 4 with cumulative performance indices of 19.98 and 19.67 respectively, have been ranked second and third. With cumulative performance indices of 19.60, 19.35, 18.91, 18.67, 18.53, 18.25, and 18.20, respectively, the accessions CUW 2, CUW 27, CUW 10, CUW 26, CUW 18, CUW 5, and CUW 22 were rated from 4 to 10 (Table 4.25 and 4.26; Figs. 4.17- 4.26). When compared to other accessions, these superior accessions have significantly higher values of agronomic characters. Further breeding experiments can be performed on these accessions to produce genotypes with more promising agronomic characteristics and can be made available to farmers.

Similar studies on performance analysis have been conducted on various crops in order to choose superior genotypes from the available germplasm by earlier workers like Ahmad *et al*. (2007) in tomato, Elavarasan *et al*. (2013) in cabbage, Salimath *et al*. (2014) and Chaudhary *et al*. (2006) in turmeric, Hrideek *et al*. (2011) in small cardamom and Chandramohanan *et al*. (2016) in rice.

Table. 4.25. Performance analysis of the accessions of *Curcuma zedoaria* studied – Mean values of the characters.

Accession No.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
CUW 1	83.50	1.44	6.27	53.25	7.87	255.45	216.00	3.33	22.89	15.86	5.33	7.77	3.87	4.58	7.66
CUW 2	92.38	1.22	7.05	58.57	8.09	285.36	268.33	3.44	22.66	15.65	6.24	10.82	4.93	5.11	10.04
CUW 3	90.99	1.22	7.94	58.34	7.77	271.73	324.44	3.77	26.88	17.60	5.99	9.93	4.54	4.64	9.35
CUW 4	83.16	1.22	8.05	55.23	7.73	258.45	263.30	3.77	27.4	17.01	5.81	9.98	5.56	5.48	8.72
CUW 5	86.56	1.22	6.94	56.98	6.81	232.86	195.00	3.67	30.22	17.62	4.79	10.16	3.96	4.84	8.17
CUW 6	87.16	1.22	7.11	55.9	7.78	261.94	216.66	2.66	17.22	14.66	5.79	8.97	4.63	5.33	9.22
CUW 7	87.39	1.77	6.05	56.10	8.17	279.00	202.78	3.22	19.55	14.81	4.96	8.75	4.25	4.30	7.34
CUW 8	81.11	1.44	6.50	51.38	7.84	242.31	172.22	3.11	23.33	15.32	5.2	9.69	4.22	4.27	7.34
CUW 9	100.1	1.22	7.00	60.41	8.48	307.92	362.78	4.00	28.22	20.18	6.76	13.38	6.17	6.24	12.19
CUW 10	95.55	1.44	6.55	59.38	8.80	309.84	235.55	4.33	18.44	13.94	5.46	7.41	4.71	4.96	9.63
CUW 11	90.83	1.22	6.44	55.46	8.65	288.02	186.67	4.0	25.55	14.51	4.94	6.84	4.16	4.13	7.17
CUW 12	88.00	1.44	5.88	53.62	8.15	262.4	191.66	3.55	23.33	14.23	5.23	7.07	3.75	4.57	8.55
CUW 13	56.16	1.33	6.44	36.73	6.15	144.87	87.00	2.11	10.89	12.84	4.12	5.52	3.36	3.38	5.11
CUW 14	75.42	1.33	6.16	52.23	7.22	235.4	154.4	3.11	17.22	14.08	4.91	7.12	3.29	4.32	7.46
CUW 15	82.57	1.11	7.33	52.81	8.74	282.14	195.00	3.33	18.00	14.25	5.7	7.78	4.44	4.74	8.93
CUW 16	75.83	1.33	6.83	50.45	8.83	267.7	182.22	3.66	20.55	15.13	4.91	8.01	3.95	4.15	7.67
CUW 17	75.27	1.33	6.66	48.0	7.71	230.12	186.66	3.22	23.89	13.24	5.16	7.58	4.22	4.31	8.13
CUW 18	90.11	1.22	7.55	57.59	8.06	279.34	235.00	3.0	17.9	14.8	6.33	8.18	4.77	5.53	9.89
CUW 19	86.83	1.11	6.83	56.23	7.75	259.2	182.78	4.11	18.66	12.01	5.09	7.79	4.07	4.93	9.28

CUW 20	78.83	1.22	6.66	52.14	7.7	243.13	199.4	3.11	17.44	12.92	5.43	9.08	4.45	5.66	11.17
CUW 21	72.82	1.11	7.11	54.73	8.49	286.90	174.4	2.77	15.77	11.84	5.58	5.91	4.51	5.89	10.43
CUW 22	88.00	1.55	6.50	57.31	8.29	288.16	195.5	3.44	23.66	14.20	5.46	8.25	3.84	4.87	8.93
CUW 23	90.11	1.33	6.50	58.95	7.92	280.76	184.4	3.22	21.66	14.40	5.10	7.92	4.34	4.64	7.75
CUW 24	80.27	1.11	7.55	49.83	8.82	264.3	185.5	3.44	25.78	15.91	5.24	8.96	4.17	5.06	9.48
CUW 25	90.66	1.00	7.22	57.07	8.87	301.05	151.66	3.55	18.11	13.66	5.07	4.83	3.79	4.42	8.28
CUW 26	89.57	1.11	7.44	56.63	8.40	289.00	222.77	3.44	18.55	15.16	5.73	9.40	4.80	5.26	10.14
CUW 27	90.81	1.11	7.44	58.01	9.26	314.19	203.33	2.77	16.78	13.59	5.04	8.96	4.49	11.37	9.05
CUW 28	84.44	1.00	8.22	55.40	8.81	291.8	163.89	3.77	14.88	13.67	6.04	7.63	4.51	5.07	10.35
CUW 29	80.33	1.22	6.55	51.09	7.79	242.65	131.11	3.59	18.77	13.95	5.23	4.49	3.58	4.22	8.20
CUW 30	89.90	1.00	8.22	60.72	7.09	260.42	187.22	2.44	14.11	14.17	5.99	5.02	4.38	5.73	10.93
CUW 31	37.24	1.22	7.00	24.76	3.63	54.47	218.90	2.83	16.72	15.31	6.18	6.81	4.54	5.69	10.69
CUW 32	33.66	1.11	6.72	20.10	3.51	42.88	208.84	3.0	20.49	15.35	6.07	10.08	4.56	5.03	9.72
CUW 33	30.33	1.00	7.89	19.81	3.45	41.23	160.22	3.44	14.89	13.60	4.66	6.86	4.07	3.44	5.78
CUW 34	74.20	1.33	6.34	46.0	8.04	227.9	151.1	2.67	15.0	13.15	4.92	7.74	3.71	4.14	7.35
CUW 35	33.22	1.22	6.63	21.48	3.48	45.58	190.00	2.33	15.66	12.54	5.86	8.11	4.0	5.53	10.65
CUW 36	32.22	1.11	7.55	20.57	3.38	42.45	225.00	2.66	13.33	13.28	5.31	7.24	4.0	4.59	9.70
CUW 37	28.50	1.11	7.55	18.51	3.08	36.00	146.11	2.66	12.11	13.04	4.59	6.24	3.83	5.74	9.53
CUW 38	28.00	1.11	7.50	17.69	3.32	36.07	127.2	2.66	14.78	11.79	4.62	4.97	3.57	4.43	7.86
CUW 39	30.61	1.22	7.33	19.14	3.03	35.37	113.33	2.0	11.55	11.74	4.20	4.79	3.35	4.65	8.65
CUW 40	30.61	1.11	6.55	20.28	3.58	43.69	162.89	2.55	12.22	12.15	4.98	8.7	4.25	5.52	11.56
CUW 41	64.61	1.11	6.16	43.67	6.82	184.79	140.00	2.89	12.88	12.49	4.97	8.33	3.99	4.82	9.87
CUW 42	77.16	1.11	6.83	48.2	7.84	228.35	132.22	3.44	22.33	13.42	4.66	6.84	3.83	3.91	6.79

CUW 43	75.60	1.20	6.92	52.5	7.76	255.85	142.73	3.0	14.66	14.67	5.33	6.29	3.98	4.68	8.46
CUW 44	57.61	1.11	6.33	40.68	5.66	141.46	110.67	2.47	10.81	11.27	3.29	4.33	2.98	3.67	4.55
CUW 45	41.83	1.11	5.06	33.15	4.22	86.46	49.44	1.55	7.55	7.41	3.17	2.92	2.69	3.32	4.22
CUW 46	53.38	1.33	7.77	7.75	3.46	171.19	72.78	3.22	13.11	9.09	4.08	2.95	2.04	3.99	4.98
CUW 47	60.11	1.33	6.94	39.46	7.94	191.38	97.77	3.22	16.44	8.23	4.33	2.55	2.46	4.46	5.40
CUW 48	48.69	1.11	6.83	37.00	4.86	112.94	63.89	2.72	11.89	7.82	3.46	3.35	2.6	3.07	4.58
CUW 49	48.61	1.00	6.44	33.81	4.39	90.46	45.55	3.0	7.89	6.24	2.94	2.87	1.75	3.64	4.74
CUW 50	52.00	1.00	6.22	36.64	4.67	106.49	75.55	2.44	13.44	10.16	3.91	3.56	2.94	3.33	4.32
CUW 51	43.55	1.22	5.17	31.22	4.05	80.65	57.22	1.77	7.55	9.55	3.55	4.53	2.93	2.96	4.01
CUW 52	47.78	1.11	6.16	34.57	3.86	81.38	58.33	2.55	12.00	8.64	3.08	4.14	2.72	2.73	3.82
CUW 53	48.27	1.33	5.78	35.06	4.32	93.48	42.22	2.77	8.44	6.44	3.13	3.22	2.35	3.01	5.18
CUW 54	47.55	1.00	6.22	37.36	4.14	94.05	43.33	2.0	7.11	7.16	2.32	2.44	1.99	3.10	3.45
CUW 55	46.00	1.00	6.00	35.85	3.94	85.41	61.66	2.78	10.44	7.83	3.25	2.52	2.5	3.19	4.68
CUW 56	47.03	1.28	5.97	32.25	3.70	72.09	37.55	2.23	5.71	5.19	2.65	2.73	1.67	3.5	4.39
CUW 57	47.72	1.00	6.44	34.84	3.91	83.58	56.66	1.77	8.55	6.27	3.36	4.15	2.75	2.79	4.01
Mean	66.86	1.20	6.79	43.56	7.00	189.16	158.75	3.01	16.77	12.72	4.83	6.71	3.79	4.58	7.82

1: Plant height (cm); 2: Number of tillers; 3: Number of leaves per tiller; 4: Leaf length (cm); 5: Leaf breadth (cm);
6: Leaf area (cm^2); 7: Yield per plant (g); 8: Number of primary fingers; 9: Number of secondary fingers;
10: Length of primary finger (cm); 11: Circumference of primary finger (cm); 12: length of secondary finger (cm);
13: Circumference of secondary finger (cm); 14: Length of mother rhizome (cm); 15: Circumference of mother rhizome (cm).

Table 4.26. Performance analysis of the different genotypes of *Curcuma zedoaria* studied- Performance indices.

Acc. No.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	Total	Rank
CUW 1	1.249065	1.198602	0.923961	1.208524	1.210769	1.350587	1.380721	1.106386	1.365019	1.244898	1.102378	1.158249	1.022457	1.026745	0.99056	17.53892	19
CUW 2	1.3819	1.015482	1.038904	1.329263	1.244615	1.508724	1.715226	1.142933	1.351303	1.228414	1.290589	1.612903	1.30251	1.14556	1.298332	19.60666	**4**
CUW 3	1.361107	1.015482	1.170056	1.324043	1.195385	1.436661	2.073894	1.252575	1.602958	1.381476	1.238883	1.480234	1.199472	1.040195	1.209104	19.98153	**2**
CUW 4	1.243979	1.015482	1.186266	1.253461	1.189231	1.366448	1.683073	1.252575	1.633967	1.335165	1.201655	1.487687	1.468956	1.228507	1.127635	19.67409	**3**
CUW 5	1.294839	1.015482	1.022694	1.293178	1.047692	1.231152	1.246484	1.21935	1.802135	1.383046	0.990693	1.514519	1.046235	1.085031	1.056511	18.24904	**9**
CUW 6	1.303815	1.015482	1.047745	1.268667	1.196923	1.3849	1.38494	0.88378	1.026895	1.150706	1.197518	1.33713	1.22325	1.19488	1.192293	17.80892	12
CUW 7	1.307255	1.473281	0.891541	1.273206	1.256923	1.475098	1.296216	1.069839	1.165842	1.16248	1.025853	1.304335	1.122853	0.963974	0.949179	17.73788	14
CUW 8	1.213313	1.198602	0.957854	1.166084	1.206154	1.281115	1.100869	1.033291	1.391258	1.202512	1.075491	1.444458	1.114927	0.957249	0.949179	17.29236	21
CUW 9	1.497382	1.015482	1.031536	1.371023	1.304615	1.628	2.318972	1.328992	1.682867	1.583987	1.398139	1.994514	1.630119	1.398884	1.576361	22.76087	**1**
CUW 10	1.429319	1.198602	0.965223	1.347646	1.353846	1.638152	1.505689	1.438634	1.099648	1.094192	1.129266	1.104585	1.244386	1.111933	1.245312	18.90643	**6**
CUW 11	1.358714	1.015482	0.949013	1.258681	1.330769	1.522787	1.193237	1.328992	1.523645	1.138932	1.021717	1.019617	1.099075	0.925864	0.927195	17.61372	15
CUW 12	1.31638	1.198602	0.86649	1.216922	1.253846	1.387332	1.225134	1.17948	1.391258	1.116954	1.081696	1.053903	0.990753	1.024503	1.105651	17.4089	20
CUW 13	0.84009	1.107042	0.949013	0.833598	0.946154	0.765941	0.556124	0.701043	0.649413	1.007849	0.85212	0.822849	0.887715	0.757729	0.660804	12.33748	43
CUW 14	1.128197	1.107042	0.907751	1.185375	1.110769	1.244581	0.98696	1.033291	1.026895	1.105181	1.015512	1.061356	0.869221	0.968458	0.964697	15.71529	31
CUW 15	1.235153	0.923922	1.080165	1.198538	1.344615	1.491699	1.246484	1.106386	1.073409	1.118524	1.178904	1.15974	1.173052	1.062613	1.154791	17.548	18
CUW 16	1.134331	1.107042	1.006484	1.144978	1.358462	1.415354	1.164792	1.216028	1.225476	1.187598	1.015512	1.194025	1.043593	0.930347	0.991853	17.13588	23
CUW 17	1.125954	1.107042	0.981432	1.089374	1.186154	1.216665	1.193173	1.069839	1.424653	1.039246	1.067218	1.129927	1.114927	0.966216	1.051338	16.76316	26
CUW 18	1.347943	1.015482	1.112585	1.307022	1.24	1.476895	1.502173	0.996744	1.067446	1.161695	1.309204	1.219367	1.260238	1.239716	1.278934	18.53544	**8**
CUW 19	1.298878	0.923922	1.006484	1.276156	1.192308	1.370413	1.168371	1.365539	1.112768	0.9427	1.05274	1.161231	1.075297	1.105208	1.200052	17.25207	22
CUW 20	1.179207	1.015482	0.981432	1.183333	1.184615	1.28545	1.27461	1.033291	1.040014	1.014129	1.123061	1.353527	1.175694	1.268859	1.444459	17.55716	16
CUW 21	1.089304	0.923922	1.047745	1.242113	1.306154	1.516866	1.114804	0.920327	0.940426	0.929356	1.154085	0.880985	1.191546	1.320421	1.348765	16.92682	25
CUW 22	1.31638	1.290161	0.957854	1.300667	1.275385	1.523528	1.24968	1.142933	1.410937	1.1146	1.129266	1.229801	1.014531	1.091757	1.154791	18.20227	**10**
CUW 23	1.347943	1.107042	0.957854	1.337888	1.218462	1.484403	1.178727	1.069839	1.291669	1.130298	1.054809	1.180609	1.146631	1.040195	1.002198	17.54857	17
CUW 24	1.200748	0.923922	1.112585	1.130906	1.356923	1.397378	1.185758	1.142933	1.537361	1.248823	1.083764	1.335639	1.101717	1.134351	1.225915	18.11872	11
CUW 25	1.356171	0.832362	1.063955	1.29522	1.364615	1.591678	0.969445	1.17948	1.079969	1.072214	1.048604	0.719993	1.001321	0.990876	1.070736	16.63664	27

CUW 26	1.339865	0.923922	1.096375	1.285234	1.292308	1.527969	1.423996	1.142933	1.106208	1.189953	1.185109	1.401228	1.268164	1.179187	1.311263	18.67371	**7**
CUW 27	1.358414	0.923922	1.096375	1.316554	1.424615	1.66115	1.299732	0.920327	1.000656	1.066719	1.042399	1.335639	1.186262	2.548927	1.170309	19.352	**5**
CUW 28	1.263126	0.832362	1.211317	1.257319	1.355385	1.542773	1.047622	1.252575	0.887352	1.072998	1.249224	1.13738	1.191546	1.136593	1.33842	17.77599	13
CUW 29	1.201645	1.015482	0.965223	1.159503	1.198462	1.282912	0.838085	1.19277	1.119327	1.094976	1.081696	0.66931	0.945839	0.94604	1.060391	15.77166	30
CUW 30	1.344802	0.832362	1.211317	1.378058	1.090769	1.376864	1.196753	0.810685	0.841434	1.112245	1.238883	0.748316	1.157199	1.284552	1.413423	17.03766	24
CUW 31	0.557068	1.015482	1.031536	0.561935	0.558462	0.287988	1.399259	0.940262	0.997078	1.201727	1.27818	1.015145	1.199472	1.275585	1.382387	14.70157	36
CUW 32	0.503515	0.923922	0.990274	0.456175	0.54	0.22671	1.334953	0.996744	1.221898	1.204867	1.255429	1.502594	1.204756	1.127626	1.256951	14.74641	35
CUW 33	0.453702	0.832362	1.162688	0.449594	0.530769	0.217987	1.024163	1.142933	0.887948	1.067504	0.963806	1.022599	1.075297	0.771179	0.747446	12.34998	42
CUW 34	1.109948	1.107042	0.934276	1.043983	1.236923	1.204928	0.965866	0.887102	0.894508	1.032182	1.01758	1.153777	0.980185	0.928105	0.950472	15.44688	33
CUW 35	0.496933	1.015482	0.977011	0.487495	0.535385	0.240986	1.214523	0.774138	0.933866	0.984301	1.211996	1.208932	1.056803	1.239716	1.377215	13.75478	37
CUW 36	0.481975	0.923922	1.112585	0.466842	0.52	0.224437	1.438251	0.88378	0.794919	1.042386	1.098242	1.079244	1.056803	1.028986	1.254364	13.40674	38
CUW 37	0.426328	0.923922	1.112585	0.42009	0.473846	0.190335	0.933968	0.88378	0.722166	1.023548	0.949328	0.930177	1.011889	1.286794	1.232381	12.52114	41
CUW 38	0.418848	0.923922	1.105217	0.40148	0.510769	0.190705	0.813091	0.88378	0.881388	0.925432	0.955533	0.740862	0.943197	0.993118	1.016423	11.70377	45
CUW 39	0.457891	1.015482	1.080165	0.434388	0.466154	0.187004	0.724431	0.664496	0.688771	0.921507	0.868666	0.71403	0.885073	1.042437	1.118583	11.26908	46
CUW 40	0.457891	0.923922	0.965223	0.460261	0.550769	0.230993	1.04123	0.847232	0.728726	0.953689	1.02999	1.296882	1.122853	1.237474	1.494892	13.34203	39
CUW 41	0.966492	0.923922	0.907751	0.991103	1.049231	0.977001	0.894912	0.960197	0.768084	0.980377	1.027921	1.241727	1.054161	1.080548	1.276348	15.09978	34
CUW 42	1.154226	0.923922	1.006484	1.093913	1.206154	1.207307	0.84518	1.142933	1.331624	1.053375	0.963806	1.019617	1.011889	0.876544	0.878055	15.71503	32
CUW 43	1.13089	0.998835	1.019747	1.191503	1.193846	1.352702	0.912363	0.996744	0.874232	1.151491	1.102378	0.93763	1.051519	1.049163	1.094013	16.05706	29
CUW 44	0.86178	0.923922	0.932803	0.923245	0.870769	0.747912	0.707428	0.820653	0.644642	0.884615	0.680455	0.645459	0.787318	0.822741	0.588387	11.84213	44
CUW 45	0.625729	0.923922	0.745653	0.752349	0.649231	0.457122	0.316032	0.514984	0.450236	0.581633	0.655636	0.435275	0.7107	0.744278	0.545713	9.108493	55
CUW 46	0.798504	1.107042	1.145004	0.175889	5.609231	0.905097	0.465226	1.069839	0.7818	0.713501	0.843847	0.439747	0.53897	0.894478	0.643993	16.13217	28
CUW 47	0.899177	1.107042	1.022694	0.895556	1.221538	1.011843	0.624968	1.069839	0.98038	0.645997	0.895553	0.38012	0.649934	0.999843	0.698306	13.10279	40
CUW 48	0.728347	0.923922	1.006484	0.839726	0.747692	0.597124	0.408399	0.903715	0.709046	0.613815	0.715615	0.499374	0.686922	0.688233	0.592267	10.66068	48
CUW 49	0.72715	0.832362	0.949013	0.767328	0.675385	0.47827	0.291166	0.996744	0.470511	0.489796	0.608066	0.427822	0.462351	0.816015	0.612957	9.604936	53
CUW 50	0.777861	0.832362	0.916593	0.831556	0.718462	0.563022	0.482933	0.810685	0.801479	0.797488	0.808687	0.530678	0.77675	0.74652	0.558645	10.95372	47
CUW 51	0.651458	1.015482	0.761863	0.708547	0.623077	0.426404	0.365763	0.588079	0.450236	0.749608	0.73423	0.675273	0.774108	0.663573	0.518557	9.706258	52
CUW 52	0.714734	0.923922	0.907751	0.784576	0.593846	0.430263	0.372859	0.847232	0.715606	0.678179	0.637022	0.617137	0.718626	0.612012	0.493987	10.04775	49

CUW 53	0.722064	1.107042	0.851754	0.795697	0.664615	0.494237	0.26988	0.920327	0.50331	0.505495	0.647363	0.479995	0.620872	0.674782	0.669856	9.927289	50
CUW 54	0.711294	0.832362	0.916593	0.847896	0.636923	0.497251	0.276975	0.664496	0.423997	0.562009	0.479835	0.363723	0.52576	0.694958	0.44614	8.880212	56
CUW 55	0.688108	0.832362	0.884173	0.813626	0.606154	0.45157	0.394145	0.923649	0.622577	0.6146	0.672182	0.375648	0.660502	0.715134	0.605198	9.859628	51
CUW 56	0.703515	1.065424	0.879752	0.731923	0.569231	0.381146	0.240028	0.740913	0.340509	0.407378	0.548087	0.406952	0.441215	0.78463	0.567697	8.8084	57
CUW 57	0.713837	0.832362	0.949013	0.790704	0.601538	0.441895	0.362184	0.588079	0.509869	0.492151	0.694933	0.618627	0.726552	0.625462	0.518557	9.465763	54

1: Plant height (cm); 2: Number of tillers; 3: Number of leaves per tiller; 4: Leaf length (cm); 5: Leaf breadth (cm); 6: Leaf area (cm^2); 7: Yield per plant (g); 8: Number of primary fingers; 9: Number of secondary fingers; 10: Length of primary finger (cm); 11: Circumference of primary finger (cm); 12: Length of secondary finger (cm); 13: Circumference of secondary finger (cm); 14: Length of mother rhizome (cm); 15: Circumference of mother rhizome (cm).

Fig. 4.17. Rhizome of *Curcuma zedoaria*
Rank No. 1, Accession No. 9

Plant height (cm)	**:**	**100.1**
Number of tillers	**:**	**1.22**
Number of leaves per tiller	**:**	**7**
Leaf area (cm^2)	**:**	**307.92**
Number of primary fingers	**:**	**4**
Number of secondary fingers	**:**	**28.22**
Yield per plant (g)	**:**	**362.78**

Fig. 4.18. Rhizome of *Curcuma zedoaria*
Rank No. 2, Accession No. 3

Plant height (cm)	**:**	**90.99**
Number of tillers	**:**	**1.22**
Number of leaves per tiller	**:**	**7.94**
Leaf area (cm^2)	**:**	**271.73**
Number of primary fingers	**:**	**3.77**
Number of secondary fingers	**:**	**26.88**
Yield per plant (g)	**:**	**324.44**

Fig. 4.19. Rhizome of *Curcuma zedoaria*
Rank No. 3, Accession No. 4

Plant height (cm)	**:**	**83.16**
Number of tillers	**:**	**1.22**
Number of leaves per tiller	**:**	**8.05**
Leaf area (cm^2)	**:**	**258.45**
Number of primary fingers	**:**	**3.77**
Number of secondary fingers	**:**	**27.4**
Yield per plant (g)	**:**	**263.3**

Fig. 4.20. Rhizome of *Curcuma zedoaria*
Rank No. 4, Accession No. 2

Plant height (cm)	**:**	**92.38**
Number of tillers	**:**	**1.22**
Number of leaves per tiller	**:**	**7.05**
Leaf area (cm^2)	**:**	**285.36**
Number of primary fingers	**:**	**3.44**
Number of secondary fingers	**:**	**22.66**
Yield per plant (g)	**:**	**268.33**

Fig. 4.21. Rhizome of *Curcuma zedoaria*
Rank No. 5, Accession No. 27

Plant height (cm)	**:**	**90.81**
Number of tillers	**:**	**1.11**
Number of leaves per tiller	**:**	**7.44**
Leaf area (cm^2)	**:**	**314.19**
Number of primary fingers	**:**	**2.77**
Number of secondary fingers	**:**	**16.78**
Yield per plant	**:**	**203.33**

Fig. 4.22. Rhizome of *Curcuma zedoaria*
Rank No. 6, Accession No. 10

Plant height (cm)	**:**	**95.55**
Number of tillers	**:**	**1.44**
Number of leaves per tiller	**:**	**6.55**
Leaf area (cm^2)	**:**	**309.84**
Number of primary fingers	**:**	**4.33**
Number of secondary fingers	**:**	**18.44**
Yield per plant (g)	**:**	**235.55**

Fig. 4.23. Rhizome of *Curcuma zedoaria*
Rank No. 7, Accession No. 26

Plant height (cm)	**:**	**89.57**
Number of tillers	**:**	**1.11**
Number of leaves per tiller	**:**	**7.44**
Leaf area (cm²)	**:**	**289.00**
Number of primary fingers	**:**	**3.44**
Number of secondary fingers	**:**	**18.55**
Yield per plant	**:**	**222.77**

Fig. 4.24. Rhizome of *Curcuma zedoaria*
Rank No. 8, Accession No. 18

Plant height (cm)	**:**	**90.11**
Number of tillers	**:**	**1.22**
Number of leaves per tiller	**:**	**7.55**
Leaf area (cm^2)	**:**	**279.34**
Number of primary fingers	**:**	**3.00**
Number of secondary fingers	**:**	**17.9**
Yield per plant	**:**	**235.00**

Fig. 4.25. Rhizome of *Curcuma zedoaria*
Rank No. 9, Accession No. 5

Plant height (cm)	**:**	**86.56**
Number of tillers	**:**	**1.22**
Number of leaves per tiller	**:**	**6.94**
Leaf area (cm^2)	**:**	**232.86**
Number of primary fingers	**:**	**3.67**
Number of secondary fingers	**:**	**30.22**
Yield per plant	**:**	**195.00**

Fig. 4.26. Rhizome of *Curcuma zedoaria*
Rank No. 10, Accession No. 22

Plant height (cm)	**:**	**88.00**
Number of tillers	**:**	**1.55**
Number of leaves per tiller	**:**	**6.50**
Leaf area (cm^2)	**:**	**288.16**
Number of primary fingers	**:**	**3.44**
Number of secondary fingers	**:**	**23.66**
Yield per plant	**:**	**195.5**

4.6. Performance of *Curcuma zedoaria* Rosc. based on the status of the planting material used

In the current study, it was investigated how the fifteen morphometric characteristics of zedoary were affected by the morphological status of the planting material. Of the fifteen characters considered for this study, two of them, the number of tillers and number of leaves per tiller showed comparatively higher values in the case of plants produced by primary fingers. In the case of number of leaves per tiller, this difference was statistically significant, but not in the case of number of tillers. Plant height, leaf length, leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome, and the circumference of mother rhizome showed highest values when the plants were derived from mother rhizomes. When secondary fingers were used as seed material, not a single character showed the highest value.

Higher yield was observed when the plants were raised from mother rhizomes (263.33 g) and primary fingers (178.12 g) as planting materials. Higher yield and growth characters can be observed when mother rhizomes and primary fingers are used for planting. In contrast, the plants developed from secondary fingers produced the lowest yield (93.75g). This study suggests that farmers can use both mother rhizomes and primary fingers for large-scale cultivation of *C. zedoaria* for maximum harvest.

Various results have been obtained in the case of some other rhizomatous crops based on the status of the planting materials used. In *Curcuma longa*, Kumar and Gill (2011) found that the mother rhizome derived plants had a higher number and weight of total rhizomes per plant and the highest yields for fresh, dry and processed turmeric. In *Curcuma longa*, Manhas *et al*. (2010) observed that mother rhizomes gave higher yield than primary and secondary fingers. The yield of *Curcuma aeruginosa* was not significantly reduced using rhizomes with different statuses as seed material (Soorya *et al*., 2016b). It is recommended by Jayasree *et al*. (2014b) to use mother rhizomes as planting material since they produce 35-50% higher yield as compared to primary and secondary finger plantings of *Curcuma amada.*

Table. 4.27. Observations on the growth and yield characters of *Curcuma zedoaria* in relation to the status of the planting material used.

Sl. No.	Characters	Mother rhizome		Primary finger		Secondary finger		CD 5%
		Mean ±SE	CV	Mean±SE	CV	Mean±SE	CV	
1	Plant height (cm)	103.82±1.49	9.96	90.29±2.76	21.22	76.11±3.00	27.35	7.1
2	Number of tillers	1.33±0.08	42.10	1.67±0.062	25.75	1.27±0.070	38.58	NS
3	Number of leaves per tiller	6.22±0.193	21.54	6.27±0.210	23.29	5.46±0.20	25.46	0.57
4	Leaf length (cm)	65.42±0.88	9.34	58.60±1.53	18.10	48.42±1.73	24.80	4.04
5	Leaf breadth (cm)	10.16±0.22	15.26	8.64±0.25	20.14	6.57±0.31	32.57	0.75
6	Leaf area (cm^2)	399.11±12.11	21.02	307.88±14.00	31.52	197.72±14.86	52.09	38.77
7	Yield per plant (g)	263.33±15.16	39.89	178.12±12.82	49.87	93.75±10.57	78.16	36.74
8	Number of primary fingers	4±0.19	32.25	3.75±0.16	28.8	3.23±0.14	30.65	0.46
9	Length of primary finger (cm)	16.00±0.48	20.94	13.95±0.50	24.80	11.12±0.54	34.18	1.44
10	Circumference of primary finger (cm)	7.02±0.21	20.66	5.91±0.21	24.36	4.22±0.19	31.28	0.57
11	Number of secondary fingers	26.35±0.98	25.73	24.21±0.16	35.27	18.10±0.14	43.15	3.16
12	Length of secondary finger (cm)	7.33±0.34	32.61	6.5±0.38	40.46	4.35±0.33	51.95	1.00
13	Circumference of secondary finger (cm)	7.02±0.21	20.66	5.91±0.21	34.12	4.22±0.19	31.04	0.57
14	Length of mother rhizome (cm)	7.03±0.22	21.91	6.23±0.24	26.48	4.28±0.16	26.40	0.59
15	Circumference of mother rhizome (cm)	13.10±0.36	19.00	10.75±0.35	22.51	7.60±0.39	35.92	1.04

Chapter V

SUMMARY AND CONCLUSION

Curcuma zedoaria Rosc., a perennial herbaceous plant belonging to the family Zingiberaceae is found in the tropical regions of Asia including India, Japan and Thailand. Many human ailments are treated with the rhizomes of this plant in traditional Indian and Southwest Asian medical systems. There are several diseases for which this plant may prove to be a promising ingredient for the development of novel drugs. Due to its unique pharmacological properties, *C. zedoaria* is probably one of the most important members of the ginger family. Both processed and unprocessed rhizomes of *C. zedoaria* are consumed as food. Rhizomes contain adequate amounts of starch with a high concentration of amylose. Rhizomes are also reported to be used as an appetizer.

The genetic variability of *Curcuma zedoaria* needs to be explored and conserved. This is because its natural habitats are disappearing due to anthropogenic activities such as deforestation, habitat destruction and the replacement of peasantry farming with industrial agriculture. Besides, overexploitation and climate change also threaten the landscape ecology of its habitats. The present experiments were designed and carried out so as to investigate the variability among genotypes of *C. zedoaria* in the study area. In addition, the interrelationship and association of the growth and yield characteristics of the species were analysed using standard statistical methods. An effort has also been made to find out the best accessions of the species collected and to find out the nature of the rhizomes that could be used as planting material.

Over the period of 2018-2021, the experiment was conducted at the Genetics and Plant Breeding Division of the Department of Botany of University of Calicut, Kerala, India. Study material included fifty seven accessions of *C. zedoaria* collected from different regions of Kerala State. During December, January and February of 2018, healthy and fresh rhizomes of the species were collected and planted in the experimental plot for multiplication and preliminary screening. In the month of May 2019, before the onset of the southwest monsoon, evaluation trials and other experiments were initiated. During all the studies, healthy rhizomes of 25 g - 30 g in weight and 4-7 cm in length were used as planting material. The growth characteristics of the plants were assessed after six months of growth, while yield characteristics were assessed at nine months after reaching maturity by destructive sampling method.

Frequency distribution analysis of agronomic characters of crop plants will provide information on the mechanisms of their genetic control. Frequency distribution analysis of agronomic characters showed continuous frequency distributions with all possible intermediates, indicating their polygenic control. Among the growth characters examined, plant height and leaf breadth displayed skewness of the distribution towards the right side of the distribution curve. This indicates the accumulation of higher number of dominant contributing alleles in their gene pool. The characters such as number of tillers, number of leaves per tiller, leaf length and leaf area showed continuous distribution with skewness towards the proximal side of the distribution curve indicating that the gene pools of these characters have a higher number of recessive contributing factors.

It was found that all the yield characters studied i.e., yield per plant, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger,

circumference of secondary finger, length of mother rhizome and circumference of mother rhizome showed skewness towards the proximal side of the distribution. This indicates that the population under study has a higher number of recessive contributing alleles in the case of these characters. There were no characters that displayed a higher frequency of dominant alleles. In summary, among the fifteen agronomic characters, thirteen characters showed continuous frequency distribution with skewness towards the proximal side. In contrast, two characters showed continuous frequency distribution with skewness towards the distal side of the distribution. Due to the fact that thirteen of the fifteen agronomic traits examined display continuous frequency distributions with skewness toward the proximal side of the distribution, there is a higher accumulation of recessive contributing factors in their genetic pool. This implies that scientific selection processes should be employed to generate varieties with a higher accumulation of dominant alleles for these traits. As most of these characters contribute to yield, crop improvement programmes are necessary for better yield of this valuable crop.

Genetic variability of plants refers to heritable variations between and within populations, which is used for selection and other plant improvement programmes. In the present study of the significance of genetic variability of agronomic characters, all the characters studied showed significant variation at 1% level of significance. The variability of a character is determined by the value of its coefficient of variation. Of all the growth characters examined, leaf area had the highest coefficient of variation followed by plant height. It was the number of tillers that had the lowest coefficient of variation. A study of yield characters showed the highest coefficient of variation for yield per plant followed by length of the secondary finger. The lowest value was observed for the length of the mother rhizome.

Out of the fifteen agronomic characters, leaf area, circumference of mother rhizome and length of secondary finger were the most variable characters. Number of tillers and number of leaves per tiller were found to be the traits with the lowest variation. The possibility of choosing improved genotypes based on the specified characters in future crop improvement programmes is indicated by the statistically significant level of variability in all the studied characters.

Analysing the phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV) and heritability (broad sense) of the agronomic characters of a crop species can give an insight into the impact of environmental factors on the characters. All of the agronomic characters analysed presently exhibited higher values of phenotypic coefficients of variation than genotypic coefficients of variation, demonstrating polygenic control, additive nature, and differential degrees of environmental impact on the characters under study. Leaf area had the highest PCV and GCV out of the fifteen characters studied, followed by yield per plant. The lowest PCV was observed for the number of leaves per tiller, as well as the lowest GCV for the number of tillers. Among the yield characters, yield per plant had the highest PCV value followed by length of secondary finger. The length of mother rhizome had the lowest PCV. Yield per plant had the highest GCV, followed by secondary finger length. Number of primary fingers exhibited the lowest GCV value. Among the growth characters, yield per plant had the highest PCV and corresponding GCV, followed by plant height. The number of leaves per tiller had the lowest PCV value, whereas the number of tillers had the lowest GCV value.

There is relatively significant influence of the environment on the phenotypic expression of the characters with the highest differences between PCV and GCV values. In contrast, characters with the lowest differences indicate relatively little influence of the environment. This condition favours

direct selection of genotypes based on the phenotypic performance of the crop. In this study, the differences between PCV and corresponding GCV were maximum for the characters such as number of tillers, number of primary fingers and number of secondary fingers indicating comparatively higher impact of environment in the phenotypic expression of these three characters. The characters with the lowest differences between PCV and GCV were plant height and leaf length. This demonstrates that the environment has a limited influence on the expression of these features, supporting direct selection based on phenotypic performance of these traits for crop improvement practices.

The assessment of heritability (broad sense) provides information about a particular trait that will be passed down to future generations. The heritability of fifteen agronomic traits of *C. zedoaria* has been investigated. The heritability of growth characters ranged from 92.65% for plant height to 4.76% for number of tillers. The heritability range for yield characters ranged from 38.46% for number of primary fingers to 87.27% for circumference of mother rhizome.

Twelve of the fifteen characters studied had high heritability: plant height, leaf length, leaf breadth, leaf area, yield per plant, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome. The number of tillers and the number of leaves per tiller had the lowest heritability. Number of primary fingers demonstrated medium level of heritability.

Genetic advance is another important selection parameter which together with heritability helps the breeder to anticipate the intensities of selection and improvement of crop plants. It indicates the quantum of improvement of a character that is possible through selection. While considering the yield characters, the highest genetic advance was observed for yield per plant followed by length of secondary finger and the lowest genetic

advance for number of primary fingers. Among the growth characters, leaf area followed by plant height exhibited the highest values of genetic advance. The lowest genetic advance was shown by number of tillers.

Having a thorough understanding of genotypic and phenotypic coefficients of variation, heritability and genetic advance of the crop is crucial for any crop improvement programme. Selection may be less effective for traits with low heritability coupled with low GCV, PCV and genetic advance. However, traits with high heritability coupled with high genetic advance indicate that these traits are controlled by additive gene action. Therefore, crop improvement programmes should take these characters into account during the selection process.

Correlation analysis is an efficient method for determining the degree of association between two or more variables. Understanding the interrelationships among different agronomic characters is useful to breeders in selecting genotypes having a group of desired characters. Correlation analysis of fifteen agronomic characters of *C. zedaria* was carried out based on the data obtained from fifty seven accessions collected. Seven characters were shown to be significantly correlated with the maximum number of characters. They were leaf breadth, leaf area, yield per plant, number of primary fingers, number of secondary fingers, length of primary finger and length of secondary finger. Yield of the plant, a complex trait highly influenced by many genetic factors and environmental fluctuations was found to have significant correlation with fourteen characters like plant height, number of tillers, number of leaves per tiller, leaf length, leaf breadth, leaf area, number of primary fingers, number of secondary fingers, length of primary finger, circumference of primary finger, length of secondary finger, circumference of secondary finger, length of mother rhizome and circumference of mother rhizome. Thus, plant breeders can take advantage of these yield contributing traits to improve the yield of *C. zedoaria.*

Factor analysis can help in character association analysis, grouping variables into factors and reducing data by identifying lead characters that have higher factor loadings. This would enable promising genotypes to be selected in such a way that other characters associated with the lead characters would also get selected automatically. Based on factor loading, two factors were extracted considering the 15 agronomic characters under study in the case of zedoary.

There were fourteen variables in the first factor group, including plant height, number of primary fingers, yield per plant, leaf length, leaf breadth, leaf area, circumference of the mother rhizome, number of secondary fingers, length of secondary finger, length of mother rhizome, length of primary finger, circumference of secondary finger, circumference of primary finger and number of leaves per tiller. The second factor group was occupied by one variable namely number of tillers. Characters in the same factor group share common alleles to a significant extent in their expression. The lead characters are recognised to have the highest values of factor loadings. This study revealed that length of primary fingers, circumference of primary fingers, yield per plant and circumference of secondary fingers had the highest factor loadings. Therefore, they could be utilized for selection and subsequent crop improvement programmes in zedoary. The bulk of the characters needed for analysis could be reduced without affecting the results of the research in this way.

Genetic divergence analysis measures the extent of genetic diversity prevailing in the selected genotypes which enhances the process of selection. Cluster analysis is an effective and commonly used statistical approach for evaluating the genetic behaviour of genotypes under investigation, and as a result, genetic divergence. Cluster analysis grouped the fifty seven accessions into two major clusters at a linkage distance of 0.999. There were fifty six

accessions included in the first cluster with the maximum number of related genotypes. Cluster II was occupied by a single accession, CUW 56. The first cluster was bifurcated again into two sub-clusters at a linkage distance of 0.998. The small cluster included 10 accessions and the large cluster had 46 accessions. The smaller cluster got split into two at a linkage distance of 0.948. Each sub cluster got split again at a linkage distance of 0.934. Genotypes from all the districts were included in Cluster II and Cluster I included a single genotype from Thrissur district.

It has been found that genotypes belonging to different clusters tend to have higher levels of genetic divergence in their genetic makeup, while genotypes belonging to the same cluster tend to have higher levels of genetic similarity. Due to this, there is less scope for selecting genetically divergent parents from the same cluster since genotypes possessing closeness might have evolved from same parental lines. By selecting genotypes between clusters to be used in selection and clonal propagation programmes, novel and improved varieties of *C. zedoaria* could be developed.

An analysis of performance was conducted to find out the superior accessions of *C. zedoaria* collected. Accession number CUW 9 ranked first among the fifty seven accessions collected, with a cumulative performance index of 22.76. Accessions CUW 3 and CUW 4 were placed second and third, with cumulative performance indices of 19.98 and 19.67, respectively. The accessions CUW 2, CUW 27, CUW 10, CUW 26, CUW 18, CUW 5 and CUW 22 were rated from 4 to 10 with cumulative performance indices of 19.60, 19.35, 18.91, 18.67, 18.53, 18.25, and 18.20, respectively. Over the remaining accessions, the superior accessions have significantly higher values of agronomic characters. These can be subjected to further breeding programmes so that farmers can have access to genotypes with better agromorphologic characters.

Most farmers use mother rhizomes as planting material for the cultivation of *C. zedoaria*. But due to the lack of sufficient planting material, the large scale production of the crop is difficult. A study was conducted to find out the feasibility of using primary and secondary fingers as planting material. The experiment used mother rhizomes, primary fingers and secondary fingers as planting material. An analysis of the effects of the morphological status of planting materials on fifteen agronomic characters of the crop was performed. Plants cultivated from mother rhizomes and primary fingers produced higher yield. The yield and growth characters of plants improve when mother rhizomes and primary fingers are used for planting. Plants grown from secondary fingers, on the other hand, provided the lowest yield. According to the findings of this study, farmers can maximise harvest by cultivating *C. zedoaria* on a big scale with both mother rhizomes and primary fingers as the planting material.

Genetic diversity - the heritable variation within and between populations of crop plants - is essential for selection as well as for other plant improvement programmes. Conservation of genetic diversity is, therefore, necessary for the present and future well being of society. Genetic diversity studies should provide valuable information for the conservation and improvement of crop plants. The study is therefore significant for conserving *C. zedoaria* Rosc. and providing opportunities to plant breeders to develop new, improved varieties of this crop. Furthermore, ten superior genotypes were identified from the gene pool of *C. zedoaria* collected and these genotypes could be used for further improvement programmes.

RECOMMENDATIONS

- The study has identified superior genotypes that could be used as parental lines in further improvement programmes. They can be used to create new improved varieties of the species in the future.
- The yield of currently cultivated varieties can be increased by improving other traits contributing to it, as recommended by the study.
- Farmers can use both mother rhizomes and primary fingers as planting materials in future farming to increase yield, as suggested by the current study.

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PUBLICATIONS

1. Litty R., Mohammed Irfan T.K., Radhakrishnan V.V. and Mohanan K.V., 2022. A study on the genetic control and potential of the important agronomic characters of *Curcuma zedoaria* Rosc. *International Journal of Current Research* **14**(11): 22817-22819.

2. Litty R., Radhakrishnan V.V., Resmi L. and Mohanan K.V., 2022. A study on the genetic variability of *Curcuma zedoaria* Rosc. accessions of Kerala. *Journal of Advanced Scientific Research* **13**(6): 96-100.

SEMINAR PRESENTATIONS

1. Litty R., Radhakrishnan V.V. and Mohanan K.V., 2019. Exploratory surveys for white turmeric (*Curcuma zedoaria* Rosc.) genetic resources of Kerala. In: *Abstracts of the XLII All India Botanical Conference of the Indian Botanical Society and National Symposium on Innovations and Inventions in Plant Science Research*. November 06-08, 2019, Calicut, Kerala. p.201.

2. Litty R., Radhakrishnan V.V. and Mohanan K.V., 2019. *Curcuma zedoaria* Rosc. (White turmeric) – An insight into a rare medicinal plant. In: *Abstracts of the GMF National Seminar on Forestry, Plant Genetics and Improvement.* December 03-04, 2019, Kerala Forest Research Institute, Peechi, Kerala. p.52.