STUDIES ON GENETIC DIVERSITY IN TURMERIC (*Curcuma longa* L.) GENOTYPES USING MORPHOLOGICAL AND MOLECULAR MARKERS

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DECLARATION

I, **Mrs. K.MAMATHA** hereby declare that the thesis entitled **"STUDIES ON GENETIC DIVERSITY IN TURMERIC (*Curcuma longa* L.) GENOTYPES USING MORPHOLOGICAL AND MOLECULAR MARKERS"** submitted to Dr. Y.S.R. Horticultural University, Venkataramannagudem, West Godavari for the degree of **DOCTOR OF PHILOSOPHY IN HORTICULTURE (PLANTATION, SPICES, MEDICINAL AND AROMATIC CROPS)** is the result of original research work done by me. I declare that no material contained in the thesis has been published earlier in any manner.

Date: **(K.MAMATHA)**

Place: VENKATARAMANNAGUDEM

CERTIFICATE

Mrs. K.MAMATHA has satisfactorily prosecuted the course of research and that the thesis entitled **"STUDIES ON GENETIC DIVERSITY IN TURMERIC (*Curcuma longa* L.) GENOTYPES USING MORPHOLOGICAL AND MOLECULAR MARKERS"** submitted is the result of original research work and is of sufficiently high standard to warrant its presentation to the examination.

I certify that neither the thesis nor its part thereof has been previously submitted by her for a degree of any University.

Place: VENKATARAMANNAGUDEM. (**Dr**. **M. B. NAGESWARA RAO**)

Date: Major Advisor

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LIST OF SYMBOLS AND ABBREVIATIONS

T	:	Absolute temperature
P.M	:	After noon / Post Meridian
a.i.	:	Active ingredient
vs.	:	Against
ARI	:	Agricultural Research Institute
ANOVA	:	Analysis of variance
et al.	:	And others
etc.	:	And so on; other people/ things
per se	:	As such with mean
@	:	At the rate of
A.M.	:	Before noon/ Post Meridian
cm	:	Centimeter
χ^2	:	Chi-square
r	:	Correlation Co-efficient
CD (5%)	:	Critical Difference at 5 per cent level
Dr.Y.S.R.H.U	:	Dr. Y.S.R. Horticultural University
DAT	:	Days After Transplanting
^{0}C	:	Degree Celsius
df	:	Degrees of freedom
ER	:	Effective Rainfall
ET	:	Evapo- Transpiration
Fig.	:	Figure
F. test	:	Fisher Test
F_1	:	First Filial Generation
e.g.	:	For example, for instance
GA	:	Genetic Advance
gca	:	General combining ability
GAM	:	Genetic Advance as per cent of Mean
GCV	:	Genotypic Co- efficient of variation
r_g	:	Genotypic correlation co-efficient
$S^2{}_g$	:	Genotypic variance
X	:	Grand mean
$h^2{}_b$	:	Heritability in broad sense
IIHR	:	Indian Institute of Horticulture Research
Max	:	Maximum

m	:	Metre
ms^{-1}	:	Metre per second
m^2	:	Metre square
µl	:	Microlitre
µm	:	Micrometer
mg	:	Milligram
ml	:	Millilitre
M T	:	Million Tonnes
Min	:	Minimum
min	:	Minute
NBPGR	:	National Bureau of Plant Genetic Resources
R	:	Multiple correlation co-efficient
viz.,	:	Namely
NS	:	Non- significant
H_0	:	Null hypothesis
No.	:	Number
ppm	:	Part per million
L^{-1}	:	Per liter
m^{-2}	:	Per meter square
%	:	Percent
PCV	:	Phenotypic co-efficient of variation
PJTSAU	:	Prof. Jayashankar Telangana State Agricultural University
RBD	:	Randomized block design
s	:	Second
sca	:	Specific combining ability
SKLTSHU	:	Sri Konda Laxman Telangana State Horticultural University
cm^2	:	Square centimeter
SE (d)	:	Standard error of difference
SE (m)	:	Standard Error of mean
i.e.	:	That is
t	:	Tonne
$t\ ha^{-1}$	:	Tonne per hectare
$\sigma^2 GCA$	:	Variance due to General combining ability
$\sigma^2 SCA$	:	Variance due to Specific combining ability
Var.	:	Variety
wt	:	Weight

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ABSTRACT

Eighty three turmeric (*Curcuma longa* L.) genotypes were evaluated to elucidate the extent of genetic diversity and relationship among the accessions using morphological and molecular markers at Horticultural Research Station, Kovvur, Dr Y S R Horticultural University, Venkataramannagudem during the years 2013-2015.

Among all the accessions, CLL-335 performed well with high growth and yield, whereas, T Sundar possessed high curcumin content. Analysis of variance for twenty one characters indicated that considerable level of variability was displayed by various genotypes. The highest phenotypic and genotypic coefficient of variation was observed for number of secondary rhizomes, followed by length of leaf petiole and yield per plant. High heritability coupled with high genetic advance in per cent mean was recorded for curcumin content, number of secondary rhizomes and number of tillers per plant.

Path coefficient analysis projected basal diameter, number of primary rhizomes, girth of mother rhizome, leaf width and number of secondary rhizomes per plant as the major contributors towards yield due to high positive effects.

The genetic diversity assessed by D^2 statistic for 21 quantitative traits revealed significant contribution of the characters towards variability and grouped the entire germplasm into ten distinct clusters. The cluster mean in conjunction with inter and intra cluster distances interpreted yield, leaf area, plant height and number of secondary rhizomes as major contributing characters towards genetic divergence.

The principal component analysis (PCA) revealed 75.86 per cent of total variability of which curcumin content, leaf area and number of primary rhizomes contributed for more variation.

Genetic diversity was also assessed using twenty one RAPD and nineteen SSR markers to study the genetic variation at DNA level. Among the twenty one RAPD primers, all the primers showed the PIC value more than 0.5 except the RAPD primer OPC 16 (0.483) with the product sizes ranging from 190 bp to 1500 bp. Similarly, all the SSR primers used in the present study produced discrete, scorable and unambiguous bands. The PCR product size obtained by the amplification of SSR primers ranged from 110bp to 440bp with a total of 38 fragments. All the SSR markers used in the genetic analysis were polymorphic with banding pattern ranging from 1 to a maximum of 3 alleles per individual in all the loci.

In the present study, the SSR markers were found to be reliable markers to identify the duplicates in the germplasm whereas by using RAPD markers only closely related ones can be identified. Synonymous entities with genetic similarity coefficient value of 1.0 were identified by the SSR markers, which is utmost important for precise assessment of cultivar type and genetic diversity in turmeric. Among the 83 accessions studied, nine sets of synonymous entities were identified, indicating that SSRs are more accurate and reliable than RAPDs to study the genetic diversity in turmeric. The SSR analysis confirms the fact that collecting turmeric accessions based on vernacular identity could result in adding duplicates in the germplasm collection .The accessions which were having wide diversity can be used in the future breeding programmes.

CHAPTER I

INTRODUCTION

Turmeric (*Curcuma longa* L. Syn. *C. domestica* Valet.) known as "golden spice" as well as "spice of life" and has been used in India as medicinal plant and held sacred from time immemorial (Ravindran *et al.* 2007). Turmeric belongs to the family *Zingiberaceae* and is considered to have been originated from South East Asia and is found throughout South and South East Asia with a few species extending to China, Australia and South Pacific. The highest diversity is concentrated in India and Thailand with at least 40 species in each area followed by Myanmar, Bangladesh, Indonesia and Vietnam (Velayudhan *et al* .2012).

Turmeric is gaining importance globally as the potential source of new drugs to combat a variety of ailments as the species contain molecules credited with anti-inflammatory, hypocholestraemic, choleratic, antimicrobial, insect repellent, antirheumatic, antifibrotic, antivenomous, antiviral, antidiabetic, antihepatotoxic as well as anticancerous properties. Turmeric oil is also used in aromatherapy and in the perfume industry. Though the traditional Indian Ayurvedic system of medicine and Chinese medicine long ago recognized the medicinal property of turmeric in its crude form, the last few decades have witnessed extensive research on the biological activity and pharmacological actions of *Curcuma*, especially the cultivated species. Turmeric powder obtained from rhizomes of *Curcuma longa* or related species, is extensively used as a spice, food preservative and colouring material, in religious applications as well as a household remedy for bilary and hepatic disorders, anorexia, diabetic wounds, rheumatism and sinusitis in India, China and South-East Asia and in folk medicine. Cucuminoids, the biologically active principles from *Curcuma*, promise a potential role in the control of rheumatism, carcinogenesis and oxidative stress-related pathogenesis.

Apart from its religious, cultural and magical uses, it has been an inseparable part of Ayurvedic system of medicine in India and in Chinese system (Sasikumar, 2005).

India is the major producer (93.7% of the world production),consumer (92% of the consumption) and exporter of turmeric. During the year 2014-15, turmeric was cultivated in an area of 2.33 lakh hectare with a production of 1190 million tonnes (National Horticulture data base 2015). During 2014-15, 86,000 tonnes of turmeric was exported earning Rs.744.35 crores registering an increase of 11 per cent in volume and 12 per cent in value from the previous year's export (Spices Board Annual report, 2014-15). India is having the largest share in world exports. The top export destinations of Indian turmeric are U.A.E, Bangladesh, Malaysia, Iran, U.K and U.S.A. Many of the developed countries like U.S.A, U.K and Japan are taking much interest in purchasing Indian turmeric due to high degree of quality and high curcumin content. In Andhra Pradesh, it is cultivated in an area of 0.15 lakh hectares with a production of 95,000 MT.

Though India enjoys the monopoly in turmeric production and export, because of its ever increasing demand in both food and pharmaceutical industries, there is pressing need to further increase the productivity of turmeric. However to increase the productivity, information regarding the crop genetic diversity is essential. Turmeric is a cross pollinated triploid (2n=3x=63),which can be vegetatively propagated using its underground rhizomes (Sasikumar, 2005). Although more than 80 cultivars are known in the country, it is difficult to discriminate the cultivars solely on rhizome morphology. Since hybridization is ineffective in most of the cases, genetic improvement is often limited to germplasm selection and mutation breeding (Ravindran , *et al.*2007). Collection of accessions, especially land races and cultivars are done mainly based on the vernacular names and there is every possibility that a particular cultivar or race is known by different vernacular names in different places. This fact coupled with lack of clear-cut distinguishing morphological features could have resulted in the accumulation of duplicates in the collection (Shamina *et al.*1998).

Even though, germplasm collection represent the main source of variability for turmeric genetic improvement, studies aimed at characterizing these collections are scarce and mostly restricted to phenotypic evaluation of different accessions from India, which is the major turmeric grower of the world. However, the use of phenotypic traits in germplasm characterization is limited due to small number of descriptors available and the influence of genotype x environment interactions (Khan, *et al.* 2013). Characterization of promising turmeric cultivars/accessions by morphological data and qualitative traits like curcumin, oleoresin and essential oil content are not sufficient as these characters often change under varying environmental conditions, thus raising problem in proper identification and elimination of synonyms. Predominance of synonyms possess problem in identification and charcterization of germplasm. Lack of clear-cut morphological traits among turmeric cultivars coupled with vernacular identity of the germplasm collection results in accumulation of duplicates in the germplasm taxing heavily on conservation cost and hampering the crop improvement work. (Sasikumar, 2005).

Molecular marker techniques may overcome many limitations of the morphological and biochemical markers for the discrimination of the turmeric accessions by providing genetic background for the observed phenotypic variability since they are not affected by the environment or developmental stage and can detect the variation at the DNA level. DNA markers can be used to measure the genetic drift in crop germplasm and study the genetic diversity among the genotypes (Jan *et al*. 2011).

Since knowledge of the genetic variation in the turmeric germplasm is essential to increase the efficiency of selection in breeding programme as well as to direct conservation strategies in germplasm collection, the data will be useful for the sake of efficient management and differentiation of various land races. It would also be helpful for the plant breeders to select readily varied parents which will add new germplasm base

for future turmeric breeding programmes for increased curcumin, oleoresin and essential oil production to meet the ever increasing demand of turmeric for industrial and pharmaceutical uses.

Detailed information on genetic diversity and structure of the turmeric accessions suitable for coastal region of Andhra Pradesh is yet to be generated.

In view of the above facts, the investigation was taken up with eighty three accessions of turmeric collected from NBPGR Regional Station, Meghalaya, High Altitude Research Station, OUAT, Pottangi, TNAU, Coimbatore and local collections, to study the presence and pattern of genetic variability and relatedness among them at morphological as well as molecular level using DNA markers (RAPD and SSR), with the following objectives.

1. To document the information on morphological characters of different accessions of turmeric.
2. To estimate the variability parameters for rhizome yield and yield contributing characters and to determine the relationship between these characters through correlation analysis.
3. To study the direct and indirect effects on yield components on rhizome yield through path coefficient analysis.
4. To assess the diversity and varietal relationship in turmeric gene pool by D^2 analysis, RAPD and SSR markers.

CHAPTER II

REVIEW OF LITERATURE

Turmeric which belongs to the genus *Curcuma,* is often referred to as "Golden spice of life" is held sacred from time immemorial which is having strong association with social and religious life of people in Southern Asia and Oceana where the genus *Curcuma* is widely distributed. It belongs to the family Zingeberaceae and is considered to have been originated from South East Asia and can also found growing in Tropical Africa and India. It is found throughout South and South East Asia with a few species extending to China, Australia and South Pacific. The highest diversity is found in India and Thailand with at least 40 species in each area followed by Mayanmar, Bangladesh, Indonesia and Vietnam (Velayudhan *et al*. 2012).

The genus was first established by Linnaeus (1753) in his *'Species Plantarum'* in 1753 and is an important natural resource that provides many useful products such as spice, medicine, dye, food, perfume and aesthetics to people (Syamkumar, 2004). Frequent cultivation of the *Curcuma* species and targeted selection of the peculiar morphotypes have further contributed to taxonomic complexicity of the group. Correct taxonomic identity of the *Curcuma* species is important for bioprospecting as well as for conservation (Syamkumar, 2004).

The taxonomic hierarchy of the genus *Curcuma* is presented below (Kress *et al*.2002)

Kingdom	Plantae
Sub kingdom	Tracheobionta
Division	Magnoliophyta
Superdivision	Spermatophyta

Class	Liliopsida
Subclass	Zingiberidae
Order	Zingiberales
Family	Zingiberaceae
Subfamily	Zingiberoideae
Tribe	Zingibereae
Genus	Curcuma

Significance of genetic diversity of the species

The critical assessment of nature and magnitude of variability in the germplasm stock is one of the important prerequisites for formulating effective, meaningful breeding programmes, as crop improvement of any crop depends on extent of variability and heritability of economically important characters, though, under the relative influence of the environment on their expression. Being prone to sexual reproduction constraints in crops like turmeric reproduce vegetatively and thus have limited chances of crop improvement. In such species, the extent of genetic diversity is low unless the samples are drawn from diverse agroecological conditions (Ravindran *et al.* 2007) or the natural mutations preserved in the population by the clonal propagation. Irrespective of this over 70 popular turmeric types are under cultivation in India in addition to many lesser known local types (Babu *et al.* 2012).

Traditionally, morphological traits were used to assess diversity. But because of the existence of high level of variability, morphological data can lead to ambiguous interpretations. Molecular characterization is now the favored means to quantify variation within large germplasm samples. New DNA sequencing and genotyping technologies provide the power to interrogate thousands to millions of diagnostic polymorphisms, across hundreds to thousands of genotypes, thus facilitating the analysis of genetic structure and providing a rationale basis to identify and select among the underlying

lineages. Such approaches not only resolve genetic relationships at fine scale, but they also provide important measures of genetic divergence between and genetic diversity within the major genetic clusters that comprise crop germplasm.

Morphological markers are based on visually accessible traits such as growth habits, flower color, seed shape and pigmentation, and it does not require expensive technology but large tracts of land area are often required for these field experiments, making it possibly more expensive than molecular assessment in western (developed) countries and equally expensive in Asian and Middle East (developing) countries considering the labour cost and availability. These marker traits are often susceptible to phenotypic plasticity, conversely, this allows assessment of diversity in the presence of environmental variation which cannot be neglected from the genotypic variation. These types of markers are still having advantage and they are mandatory for distinguishing the adult plants from their genetic contamination in the field.

The literature available on morphological and molecular diversity in turmeric is reviewed under the following headings.

2.1. Variability and genetic parameters

2.2. Correlation and Path-coefficient analysis

2.3. Character associations

2.4. Genetic divergence (D^2)

2.1. VARIABILITY AND GENETIC PARAMETERS

Greater variability in the initial breeding material ensures better chances of producing desired forms of a crop plant. Thus, the primary objective of germplasm conservation is to collect and preserve the genetic variability in indigenous collection of

crop species to make it available to present and future generations. The estimates of heritability act as predictive instrument in expressing the reliability of phenotypic value. Therefore, high heritability helps in effective selection for a particular character. The genetic advance is an useful indicator of the progress that can be expected as result of exercising selection on the pertinent population. Heritability in conjunction with genetic advance would give a more reliable index of selection value (Johnson *et al.* 1955).

Existence of wide variability among the cultivars with respect to growth parameters, yield attributes and quality parameters reported by various workers are briefed here under.

Ratnambal (1986) studied the variability in quality parameters in over 100 accessions of turmeric collected from all over India belonging to both *longa* , *aromatica* and some wild types and found that dry recovery percentage varied between 13.5 to 30.8 and curcumin content varied from 2.7 to 10.9 per cent.

Pathania *et al.* (1990) observed greatest variation for curcumin content (0.28 to 8.76 per cent) while examining 23 collections of C*urcuma longa* for agronomic and quality characters. Variation for volatile oil content was also determined to classify genotypes into four groups.

The essential oil from five *Curcuma* spp. including *C. domestica* and *C. aromatica* were analysed by Zwaving and Bos (1992). The results showed that *C. domestica* yielded 3.50 per cent and *C. aromatica* yielded 9.40 per cent of essential oil.

Indiresh *et al.* (1992) studied the genetic variability in turmeric and revealed that highly significant variations were observed in characters such as plant height, petiole length, fresh rhizome yield, length of primary and secondary fingers per plant, girth of primary and secondary finger and weight of mother rhizome .

Babu *et al.* (1993) studied the genetic variability of 108 accessions of turmeric and reported that significant variability was observed for all the traits viz., plant height, girth, number of leaves and tillers leaf length and width especially for yield and yield attributing traits like weight and number of primary and secondary rhizomes. They also reported that the phenotypic coefficient of variation estimates were higher than genotypic coefficient of variation estimates for all the morphological traits and the magnitude of the difference between PCV and GCV were less for traits like plant height, girth, number of leaves, leaf length, leaf width, number and weight of mother, primary and secondary rhizomes. They revealed that high GCV and broad sense heritability estimates coupled with very good genetic gain were observed for weight of mother and finger rhizomes as well as number and weight of secondary rhizomes indicting the predominance of additive genetic variance in the expression of these traits.

Yadav *et al.* (1996) evaluated 17 genotypes grown in the dry season and observed significant differences for all the traits, *viz.*, plant height, girth, number of leaves, leaf length, number and weight of mother, primary and secondary rhizomes evaluated with exception for leaf width.

Lynrah *et al.* (1998) found that large genetic variation was observed among twenty five genotypes of turmeric for curcumin content ranging from 0.08% to 8.6% suggesting that there was a large diversity for this quality character. Similar findings were also reported by Pathania *et al.*(1998) who also reported high genetic variation and heritability for curcumin content.

Twenty seven *Curcuma longa* land races from different parts of North Indian plains were evaluated by Garg *et al.* (1999) for essential oils curcumin and oil quality and revealed that the accessions differed widely in the essential oil content which varied from 0.16 % to 1.94% and there was a variation of 0.61% to 1.45% in the dried rhizome.

Lynrah and Chakrabarthy (2000) evaluated the performance of 25 genotypes of turmeric including *C. longa*, *C. aromatica* and *C. caesia* and reported that significant variation was observed with respect to growth, yield and quality parameters among the genotypes. Among the genotypes, black turmeric (*C. caesia*), a semi wild type, showed the most vigorous growth and yield with highest number of tillers, leaves and leaf area per clump.

Hazra *et al.*(2000) carried out an investigation to study the genetic variability and revealed that leaves per clump at 180 days after planting emerged as an important yield component of turmeric.

Shamnugasundaram *et al.* (2000) envisaged that among fifteen divergent genotypes of turmeric, highest phenotypic and genotypic variations were recorded for leaf area followed by fresh rhizome yield per plant and the least in number of tillers.

Narayanpur and Hanashetti (2003) analyzed 16 cultivars of turmeric for morphological and yield characteristics and expressed that plant height, number of leaves, number of tillers and leaf area index differed significantly. Also highly significant variations were noticed among the cultivars for fresh and cured rhizome yield for which the reason was attributed to genetic characteristics and their response to particular agro climatic conditions.

Singh *et al.* (2003) studied the genetic variability among 65 exotic and indigenous genotypes of turmeric and revealed that there was larger influence of environment for the expression of the characters and the phenotypic coefficient of variability was maximum for weight of secondary rhizomes per plant, number of secondary rhizomes, length of secondary rhizomes and the PCV was minimum for plant height.

Chattopadhyay *et al.*(2004) evaluated ten promising germplasm of turmeric, for 17 growth and yield parameters at genotypic and phenotypic levels along with their interactions with the environment and expressed that the genotypes varied significantly among themselves with respect to different growth characters such as plant height, leaf length and leaf breadth and also with respect to different rhizome characters and the maximum genotypic coefficient of variation and phenotypic coefficient of variation was observed for weight of secondary rhizome, which also recorded the maximum heritability (0.885) followed by plant height (0.838), yield (0.799) and number of secondary finger (0.721).

An investigation was carried out by Sinker *et al.* (2005) to identify a promising turmeric variety for cultivation in Konkan region and reported that maximum phenotypic and genotypic coefficient of variation was observed for leaf area followed by leaf length, leaf width and plant height and the Salem variety was found to be significantly superior over all the varieties.

Singh *et al.* (2008) studied the genetic variability among 33 genotypes of turmeric and revealed that higher estimates of phenotypic and genotypic coefficients of variation was recorded for weight of mother rhizome per plant, followed by number of tillers per clump and weight of fresh rhizomes per plant.

The total curcuminoid content was found to be lower in six months old rhizomes than the rhizomes which were analyzed before planting, when grown in same environment (Thaikert and Yingyong, 2009).

Jadhav *et al.*(2009) evaluated the performance of growth, quality and yield parameters of four genotypes (Krishna, Suvarna, Salem and Wagaon) of turmeric and concluded that genotype Wagon was significantly better genotype in terms of all growth parameters and quality parameters and can be recommended for commercial production in Vidarbha region of Maharashtra State.

Nair *et al.* (2010) studied the variation among the open pollinated seedling progenies of turmeric and expressed that average yield varied between the mother genotype and progenies as well as among progenies.

Jan *et al.*(2012) revealed that among twenty genotypes of turmeric largest variation was observed for plant height, leaf length, leaf width, total leaves and number of fresh leaves.

Singh *et al.*(2012) opined that among the seventeen turmeric genotypes studied, high heritability accompanied with high genetic advance was recorded for weight of mother rhizome and weight of fresh rhizome per plant while high heritability along with moderate genetic advance was recorded for number of tillers for per clump, plant girth, length and width of mother rhizome, number and weight of fresh rhizome per plant, number and weight of secondary rhizome per plant rhizome girth and dry matter percentage and also revealed that estimates of heritability ranged from 72.9% to 81.2%.

Roopdarshini and Gayatri (2012) carried out an investigation to isolate somaclones through callus phase of vegetative bud among 105 regenerants from *C.longa* and identified that somaclones with narrow elongated leaf and thick short pseudostem were found to be highly superior with regard to morphological and biological traits.

Jilani *et al.*(2012) evaluated different cultivars of turmeric under Dera Ismail Kahn conditions in Pakistan and expressed that all the turmeric cultivars depicted significant differences for all the parameters studied and among all the varieties, Krishna showed the supremacy over the other varieties followed by Zedory and Duggirala. Similar findings were reported by Chaudary *et al.* (2006), who reported that Krishna produced more rhizomes, maximum length and width of rhizomes compared to other cultivars.

Rajyalakshmi *et al.* (2013) studied the genetic variability and character association among eight popular genotypes of turmeric in Andhra Pradesh and revealed that high estimates of heritability were observed for plant height, number of tillers per plant, number of leaves per plant and rhizome yield indicating that yield and its components were highly heritable with moderate to high level of genetic advance.

Among 50 accessions of mango ginger all the growth and yield characters showed significant variation (Jayasree *et al.* 2014).They have reported that among the characters studied, yield per plant showed highest variability followed by tiller number, leaf area, and plant height. Accession no CUM 34 showed the highest performance index with 494.44 g yield /plant followed by CUM 35 with 450 g/plant.

Prajapati *et al.* (2014) studied the genetic variability among thirty three leading turmeric genotypes of Orissa and concluded that there was wide variation among qualitative and quantitative characters.

Kumari *et al.* (2014) conducted a study on bioactive components in ten cultivars of turmeric of western UP and revealed that there was lot of variation in curcumin content (3.70 to 5.90 per cent) with maximum in NDH-1 (5.90 g/100g) and essential oil content (3.32 to 8.19 per cent) with a maximum in NDH-8 (8.19%).

Kandiannan *et al.* (2015) attempted a study to know the influence of flowering on yield and quality of eleven turmeric varieties and concluded that fresh and dry yield, dry recovery and curcumin content did not differ between flowered and non-flowered plants and it is concluded that flowering in turmeric would not have any impact on yield and quality.

Geethanjali *et al.* (2016) studied the curcumin content in turmeric samples from nine states of India and observed high curcumin content from the samples collected from

Odisha and Andhra Pradesh. They also revealed that the geographical location apart from other factors like soil, climate, method of cultivation and rainfall drastically effects the curcumin content in turmeric.

2.2 CORRELATION AND PATH COEFFICIENT ANALYSIS

Studies on correlation indicate the magnitude of association between any pair of characters hence aid in selection.

Radhakrishnan *et al.*(1995) evaluated six turmeric cultivars and reported that cultivars with high indices of green matter yield presented low curcumin content after drying indicating a negative correlation between the traits.

Hazra *et al.* (2000) carried out an investigation to elucidate the role of different growth characters on rhizome yield and expressed that leaves per clump at 180 days after planting registered maximum direct contribution to rhizome yield.

Tomar *et al.* (2005) revealed that plant height and leaf length had maximum significant positive association among themselves followed by number of primary rhizomes and thickness of primary rhizome. They also expressed that thickness of secondary rhizome followed by plant height, leaf length, thickness of primary rhizome and number of secondary rhizomes showed positive correlation with yield where as

number of tillers and number of mother rhizomes showed negative correlation with rhizome yield.

The rhizome yield exhibited highly significant and positive correlation with length of mother rhizome, weight of fresh rhizome per plant, width of primary rhizome and weight of primary rhizome per plant (Singh *et al.* 2008).

A study was conducted by Roy *et al.* (2011) to determine the pattern of distribution of morphological variation for ten quantitative characters in eighty nine turmeric genotypes collected from five states of North eastern India and opined that almost all the characters, *viz.,* plant height, number of leaves, leaf length, leaf width, width of primary rhizome exhibited significant positive correlation with yield per plot. They have also expressed that tall plants, having longer and broader leaves with higher number of suckers per plant will be an ideal plant type for high rhizome yield.

Ten varieties of turmeric (*Curcuma longa* L.) were evaluated by Shoba *et al.* (2011) and revealed that number of tillers, number of leaves, leaf breadth, weight of mother rhizome, weight of primary rhizome, length of mother rhizome, leaf length and plant height had significant and positive correlations with that of yield which indicates that these components are amenable for simultaneous improvement with that of yield. Path coefficient analysis revealed positive and direct effect on rhizome yield was maximum for weight of primary rhizome (0.666), followed by leaf length (0.405), number of leaves (0.3828) and weight of mother rhizomes (0.292).

Jan *et al.*(2012) studied the correlation and genetic diversity among twenty genotypes of turmeric collected from different locations of Pakistan and expressed that highest coefficient of variation was observed for number of dry leaves while the lowest was showed by spike width of germplasm which was ranged from 9.1 to 53.2%.

2.3 CHARACTER ASSOCIATION STUDIES

Understanding the nature and extent of the associated relationship of the quantitative characters among the germplasm accessions greatly facilitates the simultaneous improvement of several characters in a genotype in selection operations. Correlation studies also reveal the genetic barriers, if any, which are going to hurdle the enhancement of the genotype. Many yield components and growth parameters have been extensively studied by many breeders for effective selections or using germplasm material.

Chattopadhyay *et al.*(2004) evaluated ten promising germplasm of turmeric, for 17 growth and yield parameters at genotypic and phenotypic levels along with their interactions with the environment and expressed that the genotypes varied significantly among themselves with respect to different growth characters such as plant height, leaf length and leaf breadth and also with respect to different rhizome characters and indicated that leaf length, leaf breadth, weight of primary finger had substantial positive effect on yield at genotypic level but weight of primary and secondary fingers had positive direct effect on yield at phenotypic level.

Prajapathi *et al.* (2014) studied the genetic variability among thirty three leading turmeric genotypes of Orissa and revealed that there was a wide variation among qualitative and quantitative characters suggesting that there was a good deal of association among dry yield and its various attributes .

Singh and Ramakrishna (2014) evaluated twenty seven genotypes of turmeric (*Curcuma longa* L.) for estimating the extent of variability and inheritance pattern, so that suitable breeding strategies can be suggested to identify productive genotypes and indicated that there was a large variation among genotypes. They also revealed that

positive direct effects on rhizome yield were high for leaf length (2.070) and curcumin content (0.617) which is higher or fairly close to its significant correlation coefficients indicating that a direct selection based on leaf length and curcumin content would be the most effective and reliable tool to identify productive and curcumin-rich genotypes of turmeric.

2.4 GENETIC DIVERGENCE

Genetic improvement in any crop mainly depends upon the amount of genetic variability present in the population. The importance of genetic diversity in crop plants was first realized by Darwin (1859) and the term "morphism" employing genetic morphs was given by Huxley (1955) which means the existence of distinct genetic forms in a population. Mahalanobis's D^2 analysis is a powerful tool in quantifying the degree of divergence between biological populations at genetic level and provides a quantitative measure of association between geographic and genetic diversity based on generalized distance (Mahalanobis, 1936).

Estimation of degree of divergence between populations and contribution of different characters to total divergence is done by Mahalanobis D^2 statistic (Maurya and Singh, 1977) which is more reliable method in selection of parents for hybridization programme. A brief review of studies on morphological and molecular divergence in turmeric is presented here under.

2.4.1 Morphological divergence

A detailed study was made by Cintra *et al.* (2005) to know the genetic divergence of twenty one Brazilian *Curcuma longa* genotypes and grouped into five clusters and expressed that curcuminoid content and dry weight were the traits that contributed most to genetic divergence and allowed the selection of best accessions for breeding programmes to develop high yielding genotypes with high curcuminiod content.

Rao *et al.* (2005) evaluated 54 turmeric accessions and grouped into six clusters based on yield and curcumin content and revealed that all the clusters were distantly related except cluster III with cluster IV and cluster V with cluster VI indicating the wide divergence in genetic material.

Vijayalatha and Chezhiyan (2008) studied the genetic divergence between 223 accessions of turmeric by using d^2 statistic and identified eight variable parameters to group the accessions into five clusters and opined that on ranking the characters for relative contribution, yield followed by girth of secondary rhizomes, weight of primary and secondary rhizomes and days to maturity were the highest contributions to total divergence while plant height, number of leaves and number of tillers contributed least to the divergence.

Roy *et al.* (2011) evaluated eighty three turmeric accessions from five north eastern states of India and divided the accessions into five clusters based on ten morphological traits and reported that the accessions exhibited large genetic variation for rhizome weight/plant, suckers/plant, yield/plot and plant height and expressed that North Eastern region of India represents a geographical diverse area of varying altitude and temperation and the variability can be utilized for selecting high yielding genotypes.

Genetic divergence among Brazilian turmeric accessions was studied by Sigirist *et al.* (2011) using seven morpho agronomic descriptors and opined that overall genetic divergence was low although some divergent genotypes were identified and divided them into four main groups based on diversity level. They also revealed that based on canonical variable analysis some descriptors were more important to discriminate the accessions.

Jan *et al.* (2012) evaluated twenty genotypes of *Curcuma longa* to study the agro morphological characterization and revealed that considerable level of polymorphism was displayed by various genotypes for some of the quantitative characters.

Prajapati *et al.*(2014) studied the genetic divergence among thirty genotypes of turmeric and divided into five clusters based on D^2 statistics and reported that dry weight of clump, weight of primary fingers, leaf area and weight of secondary fingers have contributed more towards genetic divergence. They have observed that Thodupuzha and PTS -62 were more divergent types which were grouped under Cluster I and VI respectively.

Singh and Ramakrishna (2014) evaluated 27 genetically diverse genotypes of turmeric including 11 released varieties and 16 germplasm collections which were categorized into two groups *viz*., North East Indian collections and other parts of India and opined that there was sufficient variation with respect to plant growth, maturity, yield and curcumin content. They also revealed that the North East Indian collections were significantly superior to other parts of India collections with respect to shoot length, leaf length, shoot thickness, leaf area, rhizome yield, dry matter yield, curcumin content and curcumin yield revealing the genetic potential of North East Indian collections over other parts of India which have 34.0–52.4% yield advantages of economic produce *i.e*. dry rhizome and curcumin yield.

Mishra *et al.* (2015) conducted a study to screen sixty-five germplasms of *Curcuma longa* L. for high rhizome yield and reported considerable amount of natural and genetic variability in thirteen agro-morphological traits and revealed that direct contribution towards total rhizome yield was highest for dry weight of rhizome followed by length of stipulated tuber, respectively.

2.4.2 Molecular Divergence

Among various techniques for studying the genetic variability of the crop germplasm, the molecular approach seems to be more effective as it allows direct access to the hereditary material and makes it possible to understand the relationship between the plants (Williams *et al.*1990).

Shamina *et al.* (1998) studied isozyme polymorphism in a germplasm collection of *C.longa* and used acid phosphatase, superoxide dismutase, esterase, polyphenol oxidase peroxidase and catalase which showed good polymorphism in fifteen accessions studied.

RAPD markers were used to detect the DNA polymorphism within and among *C. wenyujin, C.sichuanensis* and *C. aromatica* from China by Chen *et al.*(1999). They observed that it was difficult to differentiate between *C. wenyujin* and *C. sichuanensis* at the molecular level. The relationship between *C. wenyujin* and *C. aromatica* was also analyzed. Based on the morphological and biochemical data, it was suggested that these two species should be combined into one and the classification based on peduncle central or peduncle lateral may not be right.

Isozyme markers were used to resolve the taxonomic confusion prevailing in the genus *Curcuma* in Thailand by Apavatjrut *et* al. (1999). Out of twenty one isozymes tested, eight isozymes namely phospho glucomutase, glutamic oxaloacetate transaminase, diaphorase, aconitase, esterase, shikimate dehydrogenase, leucine amino peptidase and isocitrate dehydrogenase which showed reliable polymorphism were used to discriminate the *Curcuma* species such as C. *zedoaria*, C. *xanthorrzhiza, C. rubescens, C elata, C. aeruginosa* along with two unidentified *Curcuma* species. The analysis based on the isozyme data showed that the species *C. rubescens* is distinct from the other species which was in agreement with the morphological observation.

Salvi *et al.* (2001) attempted to utilize fourteen RAPD primers to assess the genetic variation among the *in vitro* regenerated plants and noticed that the primers

yielded 231 scorable bands including 36 noval polymorphic bands which were absent in control but present in some of the regenerants with a polymorphism of 16.5 percent.

Paisooksantivatana *et al.* (2001) investigated the genetic diversity of *C.alismatifolia Ganep* in Thailand using allozyme polymorphism and found higher levels of genetic diversity within a population. In addition an overall higher genetic diversity in the population of *C.alismatifolia* was observed which may be due to wide range of ecological conditions within the distribution area of *C.alismatifolia* populations in Thailand.

Pinheiro *et al.* (2003) attempted to use RAPD markers to screen 20 turmeric accessions from Brazil and showed a small divergence between the accessions collected in Mara rosa, suggesting that the producers of the township used a mixture of genotypes for commercial planting.

Random amplified polymorphic DNA analysis was performed on regenerated plantlets of *C.amada* to assess the clonal fidelity which revealed 103 scorable bands using 10 primers was attempted by Prakash *et al.* (2004) and revealed that during this study even though the observed genetic diversity was low, most of the regenerated plantlets were similar to the mother plants.

Nayak *et al.* (2005) attempted to study the genetic variation among sixteen promsing cultivars of ginger by using twenty random decamer primers and found that a relatively high genetic variation was detected among the ginger cultivars which were evident from high number of polymorphic markers and unique bands with 101 polymorphic loci.

Praveen (2005) studied the genetic fidelity analysis of micropropagated plants of *C. longa* using RAPD markers and observed that plants regenerated from vegetative buds

showed high genetic fidelity, while the callus regenerated plants showed lack of genetic fidelity.

Application of single nucleotide polymorphism analysis based on species specific nucleotide sequence was developed by Sasaki *et al.* (2004) to identify the plants and drugs derived from *C. longa, C. phaeocaulis, C. zedoaria* and *C.aromatica.* Based on the difference in the nucleotide positions at 177, 645, 724 and a 4 base indel on the *trnK* gene obtained using three different lengths of 26 mer, 30mer & 34 mer and the reverse primers helped to identify the four *Curcuma* species studied. They concluded that the SNP analysis method developed become a useful method for the identification of botanical origins of *Curcuma* drugs used in Chinese medicine, which is difficult to identify morphologically and phytochemically.

Xia *et al.* (2005) used 5s rRNA spacer domain specific primers to ensure the quality of *Rhizoma curcumae* derived from three species of *Curcuma* namely *C.phaeocaulis, C. wenyujin* and *C. kwangensis* from their common adulterants and revealed that *C longa* and *C. chanyujin* are identified as the common adulterants used in the *Rhizoma curcumae.* The phylogenetic tree generated by comparing the sequence identity among the five *Curcuma* species showed that C. *phaeocalis, C. kwangensis* and *C. wenyujin* formed a single group with closest homology between *C. phaeocaulis* and *C. wenyujin, C. longa* and *C. chanyujin* showed only 50 - 55% DNA similarity between them.

Nayak *et al.* (2006) used twenty random decamer primers for analyzing the genetic diversity among seventeen promising cultivars of turmeric and revealed that high amount of genetic variability among the accessions and highest amount of polymorphism was found in ACC 31 with 98.6 percent polymorphic bands and lowest (35.6%) was observed in PTS-51.

Jatoi *et al.* (2006) demonstrated that rice -SSR primer sets can be used as RAPD markers for detection of polymorphism and assessment of genetic diversity in different Zingeberaceae taxa. Among the 141 bands, that could be scored, 140 (99.5%) were polymorphic. On the average, each microsatellite primer set amplified 17.6 DNA fragments. They have reported that based on the cluster analysis genotypes from the *Curcuma* and *Alpinia* genera were grouped into clusters I and II, respectively. Clusters III and IV comprised genotypes from the genus *Zingiber*. Based on the results they suggested that the rice microsatellite primers were useful for assessing the genetic diversity among genera in the family Zingiberaceae.

Significant variation among seventeen elite cultivars of *C.longa* was determined using cytological and RAPD markers (Nayak *et al.*2006) and investigated a single nucleotide sequence of *trnk* gene to identify *C.longa, C.phaseocaulis, C.zedoraia* and *C.aromatica.* The inter cultivar polymorphism ranged from 35.6 % to 98.6% among the seventeen cultivars studied and the amplification fragments per primer ranged from four to seventeen with fragment size ranging from 0.4 kb to 3 kb.

High level of genetic diversity within *C.zedoria* populations was observed by using RAPD analysis (Islam *et al.*2007) which revealed that hilly populations maintain higher genetic diversity and also found to be distinct from plain land and plateau land populations.

Syamkumar and Sasikumar (2007) made an attempt to characterize fifteen economically important curcuma species by using thirty nine RAPD primers and fourteen ISSR primers and revealed that a total of 376 scorable bands were observed out of which 352 were polymorphic with the polymorphism ranged from 75-100% whereas, among ISSR primers six gave 100 % polymorphic bands with an average of 94.61% polymorphism.

Skornickova *et al.* (2007) carried out an investigation to assess the chromosomal and genome size variation in 161 individual plants belonging to 51 taxonomic entries and reported that six different chromosome counts (2n = 22, 42, 63, 70, 77 and 105) were found, the last two representing new generic records. The 2C-values varied from 1.66 pg in *C. vamana* to 4.76 pg in *C. oligantha*, representing a 2.87-fold range and concluded that the basic chromosome number in the majority of Indian taxa belonging to subgenus Curcuma is x = 7.

A total of eleven unique decamer primers were used by Anuntalabhochai *et al.* (2007) to characterize twenty *Curcuma* cultivars and found that a total of 220 distinct polymorphic band profiles were produced with fragments ranging from 100 to 2500 bp in which based on that SCAR markers were developed to characterize and detect the hybrids of *Curcuma*. A robust sequence characterized amplified region with 600 bp in length was present in all ' *C. alismatijolia*' varieties and hybrids, which did not amplify in an additional series of 24 distinct *Curcuma* species used as an independent test. The marker produced was useful for cost effective morphologically independent characterization of *Curcuma* hybrids.

Genetic diversity in in-vitro conserved germplasm of *Curcuma* at NBPGR, New Delhi using RAPD markers was studied by Hussain *et al*. (2008) in a set of thirty accessions of five *Curcuma* species including *C.latifolia, C.malabarica, C.manga, C.raktakanta* and 13 *C.longa* conserved morphotypes and reported that among 200 RAPD primers studied, twenty one primers were found to be polymorphic and a total of 172 amplification products were scored with an average frequency of 18.19 bands per primer with the molecular size ranging from 200 to 3640 base pairs. Based on that, they have concluded that variability was high in twenty five accessions of *C.longa* as the similarity values were at a par with those of all accessions including wild species which indicated that RAPD analysis has proven to be successful in revealing the diversity within and among the species of *Curcuma.*

Angel *et al.* (2008) conducted a study to identify the level of genetic variation among eleven starchy *Curcuma* species using random amplified polymorphic DNA markers to verify whether RAPD can be used as a tool for differentiating and identification of promising starchy species and found that 264 polymorphic bands were identified and divided into three clusters and among all *C. harita* was found to be genetically distinct from all the other *curcuma* species.

Thaikert and Yinyong (2009) also reported genetic divergence in turmeric by using 19 random primers , which produced 184 scorable bands out of which 166 were polymorphic.

Loasatit *et al.* (2009) attempted to use AFLP and morphological markers to study the genetic diversity of thirty three samples of *Curcuma comosa* Roxb. and were amplified with nine *Eco*RI, *Mse*I primer combinations that generated a total of 161 bands, 63.35% of total bands were polymorphic and based on the UPGMA clusters derived from AFLP they were classified into two major groups based on the inflorescence.

Ahmad *et al.*(2009) examined the genetic variation in 22 accessions belonging to 11 species in four genera of the Zingiberaceae, mainly from Myanmar, by PCR-restriction fragment length polymorphism analysis to investigate their relationships within this family and reported that two of 10 chloroplast gene regions (trnS-trnfM and trnK2-trnQr) showed differential PCR amplification across the taxa. Restriction enzyme digestion of the PCR products revealed interspecific variability and the results showed that the gene region trnS-trnfM appeared to display interspecific variability among most of the species.

Das *et al.* (2010) conducted a detailed study to assess the genetic stability of regenerated plants of *Curcuma* species using 10 RAPD primers and revealed that nine

selected RAPD primers utilized to study *C.amada* gave rise to a total of 46 bands ranging from 500 -2000 base pairs with an average of five bands per primer and seven primers utilized to study *C. longa* gave rise to a total of 36 scorable bands ranging from 500 -2000 base pairs with an average of five bands per primer.

Siju *et al.* (2010) attempted a study to develop micro satellite markers in turmeric to assess the genetic diversity of twenty turmeric accessions across the country. Based on that they have separated into five groups at the Jaccards similarity coefficient of 0.79 and the genetic similarity coefficients ranged from 0.1 to 1.0 with 0.62 as the average discriminatory power of micro satellite markers ensuring the future utility of the generated micro satellite markers for genetic diversity studies in turmeric.

Keeratinijakal *et al.* (2010) attempted to use the amplified fragment length polymorphism (AFLP) marker to identify and elucidate the phylogenetic relationships among 97 accessions of 'Waan chak modlook' (*C. comosa)* collected throughout Thailand and revealed that nine AFLP primer combinations generated a total of 202 bands, of which, 158 bands were polymorphic, with an average of 17.56 bands per primer pair. Pair wise similarity estimated between all samples ranged from 0.39 to 1.00 with an average of 0.67. they also revealed that based on phylogenetic tree 'Waan chak modlook' accessions were divided into five clusters.

The psbA-trnH intergenic region was studied for authenticating *Curcuma longa* and its two related species (*Curcuma sichuanensis* and *Curcuma chuanhuangjiang*) by Deng *et al.* (2011) by analysing the sequences using Neighbor-Joining to improve the phylogenetic resolution of these three *Curcuma* species. They reported that genetic diversity of these three species is 0.009 to 0.014 (<0.05). The results show that partial population specimens of *C. sichuanensis* originate from the cultivated mutation of *C. longa* and retain the *C. chuanhuangjiang* as an individual species. They concluded that the differentiation is engendered between the wildness and cultivated specimens within *C. longa* species.

Molecular genetic finger prints of nine *Curcuma* species from North East India were developed by Das *et al* (2011) using PCR based markers to elucidate intra and inter specific genetic diversity for utilization, management and conservation, revealing that the primers produced 266 polymorphic fragments among which ISSR confirmed maximum polymorphism of 98.85% followed by AFLP (97.27%) and RAPD (93.22) . Marker index and polymorphic information content varied in the range of 8.64-48.1, 19.75-48.14, and 25-28 and 0.17-0.48, 0.19-0.48, and 0.25-0.29 for RAPD, ISSR, and AFLP markers, respectively and concluded that high genetic polymorphism documented is significant for conservation and further improvement of *Curcuma* spp.

Jan *et al*. (2011) attempted to use ten RAPD primers to study the genetic diversity among twenty genotypes of *C.longa* from three locations and generated ninety five reproducible and scorable amplification products out of which, ninety two were polymorphic with 96.84 percent polymorphism and confirmed that most of the genotypes of turmeric were genetically different from each other.

Genetic relatedness among six medicinal species of *Curcuma* from different geographical locations was assessed by Zou *et al.*(2011) using thirty RAPD primers and based on the dendrogram they have revealed that the genetic relationship of these *Curcuma* species was not associated with geographical distribution and there was no separation of cultivated populations from wild populations.

Sigrist *et al.* (2011) attempted to study the genetic variability among fifty seven accessions of turmeric by using 17 micro satellite markers and the results indicated that all the SSR markers were polymorphic with banding pattern ranging from one to three alleles per individual in all the loci and based on the similarities the accessions were divided into five clusters.

Zaveska *et al.* (2011) studied the phylogenetic relationships among three previously revealed genome size groups and among species of the highly polyploid genus *Curcuma* using AFLP and suggested two main lineages which includes hexaploids and higher polyploids of the previously recognized genome size group I, and the second one includes mainly hexaploids of genome size groups II and III. They found that hexaploid species exhibited significantly higher genetic diversity than higher polyploids (9x, 15x) suggesting that this genetic diversity pattern is largely influenced by the mode of reproduction, as higher polyploids reproduce exclusively vegetatively, whereas hexaploids reproduce mainly sexually.

Mohanty *et al.* (2012) studied the genetic stability of micro propagated mango ginger using RAPD and ISSR markers and based on the absence of polymorphism in all the regenerants reported that out of 25 RAPD and 10 ISSR primers screened, 19 RAPD primers and 8 ISSR primers gave 3100 and 3300 bands, respectively. RAPD and ISSR analysis revealed monomorphic bands showing the absence of polymorphism in all regenerants analyzed, confirming their genetic uniformity suggesting that *in vitro* multiplication method developed is appropriate and applicable for clonal mass propagation of *Curcuma amada*.

Genetic diversity in fifty five turmeric accessions and five cultivars collected from ten agro climatic zones of India was investigated by Singh *et al.* (2012) using RAPD and ISSR markers and observed high genetic distances and highest number of effective alleles which revealed relatively high genetic variation among the accessions which may be due to wide range of ecological conditions within the distribution area of its population.

Taheri *et al.* (2012) used eighteen ISSR primers to study the genetic relationship among five varieties of *C.alismatifolia* and concluded that among all, sixteen ISSR primers have exhibited polymorphism with a total of 139 amplification fragments varying from 3 to 18 fragments per primer ranging from 180 to 2151 base pairs in size. The

overall polymorphism for the 16 primers was 77% suggesting that ISSR markers were polymorphic markers suitable to detect the genetic diversity of *Curcuma* varieties at DNA level.

Khan *et al.* (2013) studied the genetic variability among fifty accessions of turmeric collected from five geographical locations using 22 RAPD primers and reported that the polymorphism of turmeric genotypes using OPA-03 (38.3%) was found to be the highest followed by OPE-07 (25.0%) and OPC-01 (21.74%) .They reported that out of a total of 141 bands, the molecular size of the bands ranged from 10 kb to 50 kb. Un-weighed Pair Group Arithmetic Averages (UPGMA) analysis has clustered 50 turmeric genotypes into 6 groups showing their differentiation on the basis of their locality.

Ashraf *et al.* (2014) conducted an investigation to assess the genetic diversity among twelve accessions of ginger collected from different regions of India using twenty decamer primers and reported that a total of 275 amplified fragments were scored among which 261 (ie 94.9%) were polymorphic and based on that dendrogram was constructed by grouping them into two clusters.

Twenty deca nucleotide primers were used for the RAPD analysis to confirm the genetic integrity of micro rhizome induced plants of three turmeric cultivars by Archana *et al.* (2013) and confirmed that all the micro rhizome induced plants were monomorphic and similar to the mother plant.

Moon *et al.* (2013) collected 52 germplasms of five representative species belonging to *Curcuma* genus (*C. longa, C. aromatica, C.zedoaria, C. phaeocaulis and C. kwangsiensis*) from different farmhouse in Korea and China to elucidate the genetic diversity among the species, using amplified fragment length polymorphism (AFLP) and obtained a total of 643 DNA fragments and 349 polymorphic bands with the 54.3% ratio of polymorphism. In the analysis of similarity coefficient using Un-weighed Pair

Group Arithmetic Averages (UPGMA), 52 *Curcuma* germplasm lines were ranged from 0.60 to 0.99 which were clustered into five distinct groups according to the species and collected geographical levels. They have also concluded that the principal coordinate analysis (PCA) by multi-variate analysis has shown significantly greater differences among species than geographical origins based on AFLP profiling data of the samples.

Using 12 RAPD primers Ghosh *et al*. (2013) attempted a study to know the genetic integrity of *in vitro* regenerated plants from rhizomal explants and respective naturally occurring field grown donor plant of *Curcuma longa* L .and observed that each primer produced a unique set of amplification products ranging in size from 100 bp to 3 kb and all the scorable bands were monomorphic in nature, indicating homogeneity among the culture regenerates and genetic uniformity with that of the donor plants.

Phurailatpam *et al.* (2013) studied on DNA finger printing of turmeric accessions using ten decamer primers and based on that reproducible banding pattern for each cultivar was obtained even in different populations.They reported that Lakadong and Shilot cultivars which have golden-yellow colour also reveal darker RAPD bands at 900 bp using OPAC-19 primer. Meitei-yaingang cultivar which is light-yellow has no such band and other cultivars which have intermediate colour have lighter bands at 900 bp using the same primer suggesting the possibility of developing a molecular marker for antioxidant character of curcumin using the primer OPAC-19.

Pradhan *et al.* (2014) carried out an investigation to study the genetic diversity among five varieties of ginger through RAPD markers and revealed that a total of 104 clear, reproducible and scorable fragments ranging from 150-13000 bp were generated from 21 primers and 99% were found polymorphic. Cluster analysis using UPGMA algorithm placed the 40 accessions into three main clusters and the genetic dissimilarity matrix between genotypes ranged from (0.74994-5.33209) between the 5 cultivars studied**.**

Taheri *et al.* (2014) studied the genetic diversity among gamma irradiated *C.alismatifolia* by using SSR markers and opined that all the SSR primer pairs were highly polmorphic ensuring the utility of micro satellite markers for genetic variation studies in *C.alismatifolia* varieties .

Basak (2014) studied the intra and inter varietal relationship between 19 local cultivated varieties of *curcuma longa* from North Eastern states using 20 RAPD and 20 ISSR primers and revealed that among all the RAPD primers OPA 18 was found to be very informative with PIC value 0.43 and among all ISSR primers HB 13 and 811 exhibited the highest polymorphism percentage and based on the UPGMA dendrogram these 19 varieties were divided into two clusters .They have opined that the genetic similarity among the varieties of each state was found to be varying using two marker systems and their combinations.

Verma *et al.* (2015) attempted a study to estimate the genetic variability in indigenous turmeric germplasm using Directed Amplification of Minisatellite DNA (DAMD) and Inter Simple Sequence Repeats (ISSR), methods and revealed that cumulative data analysis for DAMD (15) and ISSR (13) markers resulted into 478 fragments, out of which 392 fragments were polymorphic, revealing 82 % polymorphism across the turmeric genotypes. The UPGMA dendrogram generated using cumulative data showed significant relationships amongst the genotypes and all the 29 genotypes were grouped into two clusters irrespective of their geographical affiliations with 100 % bootstrap value except few genotypes, suggesting considerable diversity amongst the genotypes.

Elvira *et al.* (2015) studied the existence of genotypic diversity among turmeric accessions in Mindanao, Philippines using ten random amplified polymorphic DNA (RAPD) markers from which a total of 209 amplification products were generated. They

revealed that almost all the bands generated were polymorphic indicating genetic diversity among the accessions and six primers (OPD 08, OPB 07, OPA 11, OPA 12, OPC 05 and OPN 16) had polymorphic information content values of almost 0.5, thus, were able to discriminate between the turmeric genotypes.

1. Singh *et al.* (2015) investigated on genetic diversity of 10 turmeric genotype using SSR primers and revealed that SSR markers showed distinct polymorphism among the genotypes grouping them into two major distinct clusters, which showed a significant genetic variation ranging between 0.60 and 0.98 among the different genotypes. Based on this study, they have concluded that the highest PIC value of 0.98 for the SSR loci was associated with higher level of polymorphism and their findings distinctly identified and characterized 10 genotypes using 10 different SSR markers which can be used in background selections during backcross breeding.

CHAPTER III

MATERIALS AND METHODS

An experiment entitled "**Studies on genetic diversity in turmeric (*Curcuma longa,L.*) genotypes using morophological and molecular markers**" was carried out during 2013-15 at Horticultural Research Station, Kovvur, West Godavari District, Andhra Pradesh. The materials and methods adopted during the investigation are presented in this chapter.

3.1 LOCATION OF THE EXPERIMENTAL SITE

Horticultural Research Station, Kovvur is located at 17^0. 00 N latitude $81^0.43^1$E.longitude and an altitude of 15.66 m above mean sea level. The experimental area has got a tropical monsoon climate with south-west monsoon from June to September, north-east monsoon in October – November and dry spell from December to May with summer showers in April and May. Average temperature varies from 23^oC to 39^0C and the annual rainfall is about 110 cm.

2.

3.2. SOILS

The soils are alluvial which are endowed with good drainage. The soil properties of the experimental site are as follows:

Soil P^H	-	7.5
Organic carbon content	-	0.72
Electrical conductivity (dSm^{-1})	-	0.48
Available N	-	228.48 kg ha^{-1}
Available P	-	21.60 kg ha^{-1}
Available K	-	520.30 kg ha^{-1}

3.3 EXPERIMENTAL DETAILS

The present experiment was carried out in two phases.

1. Diversity studies in turmeric accessions using morphological markers.
2. Diversity studies in turmeric accessions using molecular markers (RAPD and SSR markers)

3.3.1 DIVERSITY STUDIES IN TURMERIC ACCESSIONS USING MORPHOLOGICAL MARKERS

A total of 83 turmeric genotypes including checks (Table 1, Plate 1 and 2) were collected from different sources *ie.*, NBPGR Regional Station, Meghalaya, High Altitude Research Station, OUAT, Pottangi, TNAU, Coimbatore including 15 local accessions and evaluated based on morphological attributes as per the descriptors given by NBPGR.

3.3.1.1 EXPERIMENTAL DESIGN:

The experiment was laid out in an augmented block design consisting of 10 augmented blocks in which three checks and ten entries were planted. Each accession was raised in a single row plot of 4.5 m length at a spacing of 22.5 cm between plants accommodating 20 plants in each row and five plants were selected and tagged from each accession for collection of data.

3.3.1.2 CULTURAL PRACTICES:

The standard packages of practices were followed throughout the study.

3.3.1.3 OBSERVATIONS RECORDED:

Five tagged plants from each accession were used for recording the observations and the mean values were subjected for statistical analysis. The following observations were recorded.

3.3.1.3.1 QUANTITATIVE CHARACTERS

3.3.1.3.1.1 GROWTH PARAMETERS

3.3.1.3.1.1.1 Plant height (cm)

The plant height was measured at 180 days from the collar region of the pseudostem upto the tip of the fully opened top most leaf and its mean value was expressed in cm per plant.

3.3.1.3.1.1.2 Basal stem diameter (cm)

The basal stem diameter was measured in two directions and its average per plant was given in cm.

3.3.1.3.1.1.3 Number of leaves per plant

The number of fully opened leaves from the five tagged plants was noted and the average was computed as total number of leaves per plant.

3.3.1.3.1.1.4 Leaf length (cm)

The length of the leaf was taken from the leaf base to the tip of the leaf along the mid rib and expressed in cm.

3.3.1.3.1.1.5 Leaf width (cm)

The leaf width was recorded at the widest point of the leaf lamina

and expressed in cm.

3.3.1.3.1.1.6 Leaf area (cm^2)

The leaf area was computed by multiplying the product of leaf length and leaf width with conversion factor 0.72 (Rao and Swamy 1984) to arrive the actual leaf area.

3.3.1.3.1.1.7 Number of tillers per plant

The numbers of tillers produced by each of the five tagged plants was counted and expressed as number of tillers per plant.

3.3.1.3.1.1.8 Length of leaf petiole (cm)

The length of the leaf petiole was measured and presented in cm.

3.3.1.3.1.2 YIELD PARAMETERS

3.3.1.3.1.2.1 Number of mother rhizomes

The number of mother rhizomes was counted and the mean was expressed as number per plant.

3.3.1.3.1.2.2 Length of mother rhizome

The length of mother rhizome was expressed in cm.

3.3.1.3.1.2.3 Thickness of mother rhizome

The girth at the broadest point of the mother rhizome was measured using a non stretchable string and scale from the five tagged .plants and the mean was expressed in cm.

3.3.1.3.2.4 Number of primary rhizomes

The primary rhizomes arising from the mother rhizomes from the

five tagged plants were counted and the mean was expressed as number per clump.

3.3.1.3.1.2.5 Length of primary rhizome

The total length of primary rhizomes from the five tagged plants was counted and their mean was expressed as length of primary rhizome per plant.

3.3.1.3.1.2.6 Girth of primary rhizome

The total girth of primary rhizomes from the five tagged plants was counted and their mean was expressed as girth of primary rhizome per plant.

3.3.1.3.1.2.7 Number of secondary rhizomes

The secondary rhizomes arising from the primary rhizomes from the five randomly selected clumps were counted and their mean was expressed as number per clump.

3.3.1.3.1.2.8 Length of secondary rhizome

The Length of secondary rhizome was measured from the five tagged plants using a scale and their mean was expressed in cm.

3.3.1.3.1.2.9 Girth of secondary rhizome

The girth of secondary rhizome was measured from secondary rhizomes of five tagged plants using the tape and the mean was expressed in cm.

3.3.1.3.1.2.10 Inner core colour

Inner core colour of the primary rhizomes was recorded visually.

3.3.1.3.1.2.11 Yield

The fresh rhizomes harvested from each accession were weighed and the mean was expressed as kg/plant and also as tonnes per ha. (t ha^{-1}).

3.3.1.3.1.2.12 Days to maturity

Number of days taken to complete the senescence of above ground parts was recorded as days to maturity**.**

3.3.1.3.1.2.13 Curing (%)

One kilogram of fresh rhizomes from each accession was boiled in pure water for 45-60 minutes till the rhizomes become soft for finger pressing and froth came out with white fumes giving out a typical turmeric odour. After boiling, the rhizomes were dried under sun and rubbed manually on cement floor to remove the adhering particles and the curing percentage was worked out as below.

$$\textbf{Curing \%} = \frac{\text{Fresh weight of rhizomes (kg)- Dry weight of rhizomes after curing}}{\text{Fresh weight of rhizomes}} \mathbf{X} 100$$

3.3.1.3.1.2.14 Curcumin Content

Curcumin content in the rhizomes of the turmeric cultivars was estimated by the method given by Manjunath *et al.* (1991). The plant samples were powdered in a plant sample analyzer and finely ground turmeric sample, weighing 0.1 g was extracted by refluxing over water cooled condenser with 40 ml of alcohol for 2 1/2 hr . The extract was cooled and filtered quantitatively into 100 ml volumetric flask. The residue was then transferred to the filter, washed thoroughly and volume was made upto 100 ml with alcohol. Then 5 ml of this aliquot was pipetted out into 100 ml volumetric flask and the volume was

made up with the alcohol. The diluted extract was mixed well and its absorbance was read at 425 nm using alcohol in Spectrophotometer(Citizen).

The Curcumin content was calculated as follows.

$$\text{Curcumin content (\%)} = \frac{0.00025 \times A\,425 \times 100 \times 100}{\text{Absorbance of standard (0.42)} \times \text{weight of samples} \times 5}$$

(0.42 absorbance at 425 nm corresponds to 0.00025 g curcumin)

3.3.1.3.2 QUALITATIVE CHARACTERS

3.3.1.3.2.1 Leaf disposition pattern

The leaf disposition pattern varies in three ways and presented as

1. Horizontal
2. Semi erect
3. Erect

3.3.1.3.2.2 Colour of the dorsal surface of the leaf

The colour of dorsal surface of the leaf was recorded as

1. Green
2. Dark green

3.3.1.3.2.3 Spatial arrangement of veins on leaves:

The spatial arrangement of veins on the leaves is presented as

1. Close
2. Distant

3.3.1.3.2.4 Prominence of leaf venation

The prominence of leaf venation is recorded as

1. Less prominent
2. Prominent

3.3.1.3.2.5 Flowering

Flowering was recorded as

1. Present

2. Absent

3.3.1.4 STATISTICAL ANALYSIS OF DIVERSITY STUDIES IN TURMERIC ACCESSIONS.

3.3.1.4.1 Genetic variability

The efficiency of selection depends on the extent of genetic variability observed in the case of any species. Genetic improvement is normally achieved by selecting the genotypes with desirable qualities from the available population. Moreover, the unthreatened occurrence of the species on the earth surface is ensured only if the species show high genetic variability.

Analysis of variance and components of genetic variability such as, genotypic and phenotypic coefficient of variance (GCV and PCV respectively), heritability and genetic advance as per cent mean (GAM) were estimated using standard procedures. Multivariate analysis (D^2 statistics), phenotypic and genotypic correlations and Path-coefficient analysis were performed on the data obtained as detailed here under.

3.3.1.4.2 Analysis of variance

Analysis of variance for each character was carried out by using the Augmented Design II method as per the procedure outlined by Federer (1956). The significance test was carried out by referring to ‘F’ table value given by Fisher and Yates (1963).

3.3.1.4. 3 Genotypic and phenotypic variances

The genotypic and phenotypic variances were computed based on the expected mean sum of squares as follows:

$$\sigma^2 g = \frac{M_2 - M_3}{r}$$

$$\sigma^2 p = \sigma^2 g + \sigma^2 e \quad \text{or} \quad \frac{M_2 - M_{3} + M_3}{r}$$

Where, $\sigma^2 g$ = Genotypic variance (GV)

$\sigma^2 p$ = Phenotypic variance (PV)

$\sigma^2 e$ = Environmental variance (EV)

M_2 = Treatment mean sum of squares

M_3 = Error mean sum of squares

3.3.1.4. 4.Coefficient of variation

Genotypic and phenotypic coefficients of variation were computed according to Burton (1952) based on the estimate of genotypic and phenotypic variance as follows:

$$\text{Genotypic variance } (\sigma^2_g) = \frac{\text{(Mean sum of squares due to treatments – Mean sum of squares due to error)}}{\text{Number of replications}}$$

Phenotypic variance $(\sigma^2_p) = (\sigma^2_g) + (\sigma^2_e)$

(σ^2_e) = Error variance

The genotypic and phenotypic coefficients of variation were calculated according to the formula given by Falconer (1967).

$$\text{Phenotypic Co- efficient of variation (PCV \%)} = \frac{\sqrt{\text{Phenotypic variance}}}{\text{Grand mean}} \times 100$$

$$\text{Genotypic Co- efficient of variation (GCV \%)} = \frac{\sqrt{\text{Genotypic variance}}}{\text{Grand mean}} \times 100$$

As suggested by Siva - Subramanian and Menon (1973), GCV and PCV were categorised,

Low = Less than 10%

Moderate = 10-20%

High = More than 20%

3.3.1.4.5 Heritability

Heritability in broad sense refers to the proportion of genetic variance to the total observed variance in the population. It has been estimated as per the formula given by Lush (1940). The extent of variation due to genetic differences among the genotypes can be used to estimate the relative contribution of the genotype and environment in the form of heritability

$$h^2 (b) = \frac{\text{Genotypic variance } (\sigma^2 g)}{\text{Phenotypic variance } (\sigma^2 p)} \times 100$$

Where, σ^2 g and σ^2 p are the genotypic and phenotypic variances. Further, the range of heritability in broad sense was classified as suggested by Johnson *et al*. (1955).

Less than 30%	:	Low
30 – 60%	:	Moderate
More than 60%	:	High

3.3.1.4.6 Genetic Advance as per cent Mean (GAM)

Genetic advance is the genetic improvement of the progeny possible through selection over the original population.

Genetic advance as per cent mean was worked out for each character adopting the formula given by Johnson *et al*. (1955)

$$GAM = \frac{GA \times 100}{\bar{X}}$$

Where,

GA = Genetic advance = k x σ^2 p x h^2

k :Selection differential which is equal to 2.06 at 5% intensity

of selection (Lush, 1940)

$\sigma^2 p$	:	Phenotypic standard deviation
h^2	:	Estimated heritability, and
$\overline{X}$	:	General mean

The range of genetic advance as per cent of mean was classified according to Johnson *et al*. (1955)

Low	:	Less than 10%
Moderate	:	10 – 20%
High	:	More than 20%

3.3.1.4.7 Correlation coefficient analysis

3. Characters of organisms show different degrees of interrelationships. The systematic interrelationship between the variables is termed correlation. To determine the degree of association of characters with yield and also among the yield components, the correlation coefficients were calculated.

Both genotypic and phenotypic coefficients of correlation between two characters were determined by using the variance and covariance components as suggested by Al-Jibouri *et al.* (1958).

$$r_g(xy) = \frac{Cov_g(xy)}{\sqrt{\sigma_g^2(x)\ .\ \sigma_g^2(y)}} \qquad r_p(xy) = \frac{Cov_p(xy)}{\sqrt{\sigma_p^2(x)\ .\ \sigma_p^2(y)}}$$

Where,

r_g (xy), r_p (xy) are the genotypic and phenotypic correlation coefficients respectively.

Cov_g, Cov_p are the genotypic and phenotypic covariance of xy, respectively.

$\sigma^2{}_g$ and $\sigma^2{}_p$ are the genotypic and phenotypic variance of x and y, respectively.

The calculated value of ‘r’ was compared with table ‘r’ value with n-2 degree of freedom at 5% and 1% level of significance, where, n refers to number of pairs of observation.

3.3.1.4.8. Path coefficient analysis

Path coefficient analysis was carried out using phenotypic correlation values of yield components on yield as suggested by Wright (1921) and illustrated by Dewey and Lu (1959). Standard path coefficients which are the standardized partial regressing coefficients were obtained using statistical software packages called GENRES. These values were obtained by solving the following set of 'p' simultaneous equation using the above package.

$$P_{01} + P_{02}\, r_{12} + \text{---------} + P_{0P}\, r_{1P} = r_{01}$$

$$P_{02}\, r_{21} + P_{02} + \text{---------} + P_{0P}\, r_{2P} = r_{02}$$

$$\downarrow$$

$$P_{01} r_{P1} + P_{02}\, r_{2P} + \text{---------} + P_{0P} = r_{0P}$$

Where, P_{01}, P_{02}, ------------- P_{0P} are the direct effects of variables 1,2,--------p on the dependent variable 0 / r_{12}, r_{13}, ------------r_{1P}--------- $r_{P(P-1)}$ are the possible correlation coefficients between various independent variables / r_{01}, r_{02}, r_{03} ---- r_{0P} are the correlation between dependent and independent variables.

The indirect effects of the ith variable *via* j^{th} variable is attained as P_{oj} x r_{ij}. The contribution of remaining unknown factor is measured as the residual factor, which is calculated and given below.

$$P^2ox = 1-[P^2_{01} + 2P_{01}P_{02}r_{12} + 2P_{01}P_{03}r_{13} + \text{------------} + P^2_{02} + 2P_{02}P_{03}r_{13} + \text{--------} + P^2_{0P}]$$

Residual factor = $\sqrt{(P^2_{ox})}$

Direct or indirect effects were categorized as suggested by Lenka and Mishra (1973) are given below:

Negligible - 0.00 to 0.09

Low - 0.10 to 0.19

Moderate - 0.20 to 0.29

High - 0.30 to 0.99

Very high - 1.00.

3.3.1.4.9 Genetic divergence analysis

The genetic diversity in 83 accessions for twenty one characters were analyzed through Mahalanobis's D^2 statistic technique.

3.3.1.4.9.1 Mahalanobis distance (or) Generalised squared distance (D^2)

Genetic diversity between accessions was estimated by using D^2 analysis given by Mahalanobis (1936). The D^2 value between i^{th} and j^{th} accessions for P characters was calculated as

$$D^2_{ij} = \sum_{t=1}^{P} (Y_{it} - Y_{jt})^2$$

Where

Y_{it} = Uncorrelated mean value of ith accession for 't' character.

Y_{jt} = Uncorrelated mean value of jth accession for't' character

D^2_{ij} = D^2 value between i^{th} and j^{th} accessions.

P = No. of characters

3.3.1.4.9.2 Test of significance

Variances were calculated for all the twenty one characters and test of significance was done. Analysis of covariance for the character pairs was estimated on the basis of mean values. A dispersion table was prepared from the estimates. After testing the differences between accessions for each of the character, a simultaneous test of significance of difference between the mean values of a number of correlated variables was done (Rao, 1952) by using 'V statistic, which in turn utilizes Wilk's criterion (Wilks,

1932). The sum of squares and sum of products of error and error plus variety, variance - covariance matrix were used for this purpose (Panse and Sukhatme, 1961).

The estimate of ‘λ’ (Wilk’s Criterion) was done using the following formula.

$$\wedge = \frac{|E|}{|E+V|}$$

Where,

$|E|$ = Determinant of error matrix

$|E+V|$ = Determinant of error + varieties matrix.

The significance of ‘λ’ was tested by

$V(Stat) = -m \log_e \wedge$

$= n-[(P+Q+1)/2] \log_e \wedge$

Where,

m = n-[(P+Q+1)/2]

n = Degrees of freedom for error + varieties

$\log_e \wedge = 2.3026 \log_{10} \wedge$

P = Number of variables or characters *i.e.*, (26)

Q = Number of varieties – 1(or df for populations) *i.e.,* (84-1)=83

V (Stat) is distributed as χ^2 with PQ (26 x 83) degrees of freedom

3.3.1.4.9.3 Transformation of correlated variables

In the present model, computation of D^2 value was reduced to simple summation of differences in mean values of various characters of two accessions i.e. $\sum d_i^2$. Therefore, transformation of correlated variables into standardized uncorrelated ones was done before working out the D^2 values. Transformation was done using pivotal condensation method in computation of D^2 values.

3.3.1.4.9.4 Computation of D^2 values

The D^2 value between i^{th} and j^{th} accessions for p characters was calculated as:

$$D^2_{ij} = \sum_{t=1}^{P} (Y_{it} - Y_{jt})^2$$

3.3.1.4.9.5 Testing the significance of D^2 values

The D^2 values obtained for a pair of accessions was taken as the values of 'X^2' and tested against tabulated 'X^2' at 'p' degree of freedom where 'p' is the number of characters considered.

3.3.1.4.9.6 Grouping of accessions into various clusters

Grouping of the accessions into various clusters was done by using Ward's minimum variance method as described by Rao (1952). The criterion used in clustering by this method is that any two variables belonging to the same cluster should on an average show a smaller D^2 value than those belonging to different clusters. The combinations of each accession were arranged in ascending order of their magnitude in a tabular form as described by Singh and Chaudhary (1977). To start with, two populations having the smallest distance with each other were considered, to which a third population having smaller D^2 value from the first two populations was added. Similarly, the next nearest fourth population was considered and this procedure was continued. At certain stage, when it was felt that after adding a particular population if there was rise in the average D^2, then, that population was not considered for inclusion in that cluster. The groups of the first cluster were then excluded from the group and the rest were treated in a similar way. This process was continued till all the populations were included into one (or) the other cluster. After the formation of the cluster, the average inter and intra cluster distances (divergence) were calculated.

3.3.1.4.9.4 .7 Average intra - cluster distance

For the measurement of intra - cluster distances the formula used is $\sum D_i^2$ / n Where, is $\sum D_i^2$ was the sum of distances between all possible combinations (n) of the populations included in a cluster.

3.3.1.4.9.8 Average inter-cluster distance

Clusters were taken one by one and their distances from other clusters were calculated. The distance between two clusters was the sum of D^2 values between the members of one cluster to each of the members of the other cluster divided by the product of number of genotypes in both the clusters under consideration.

$$\text{Average inter-cluster distance} = \frac{D^2}{(n_1 \text{ X } n_2)}$$

Where,

n_1 and n_2 are number of accessions of two clusters.

3.3.1.4.9.9 Cluster diagram

The clusters and their mutual relationships were presented diagrammatically. The square root of average D^2, which was an approximate measure of divergence between groups, was used to denote the distance.

3.3.1.4.9.10 Contribution of individual characters towards divergence

In all the combinations, each character was ranked on the basis of their contribution towards divergence between two entries ($d_i = y_{it} - y_{ij}$). Rank I was given to the highest mean difference and rank ‘p’ to the lowest mean difference, where P is the total number of characters considered. The number of cases where a particular character ranked first was continued, the proportion of this to the total number of combinations expressed in percentage was quantified as the contribution of character to the overall genetic divergence between the accessions.

$$\text{Percentage contribution of the character} = X = \frac{N \times 100}{M}$$

Where,

X = Per cent contribution of character.

N = Number of accession combinations where the character ranked first.

M = All possible combinations of the accessions concerned.

3.3.1.4.9.11 Principal Component Analysis (PCA)

Principal component analysis was carried out according to procedure described by Banfield (1978) using WINDOWSTAT software package version 8.1. It is defined as a method of data reduction to clarify the relationships between two or more characters and to divide the total variance of the original characters into limited number of uncorrelated new variables. The main objectives of PCA are:

i. To discover or to reduce the dimensionality of the data set.

ii. To identify new meaningful under laying variables.

PCA can be performed on two types of data matrices *viz.,* variance – covariance matrix and correlation matrix. With characters of different scale, a correlation matrix standardizing the original data set is preferred. If the characters are of same scale, a variance – covariance matrix can be used. In the present study, PCA was performed on the correlation matrix of traits, thereby removing the effects of scale (Jackson, 1991). The main reasons for the use of the correlation matrix are :

1. The presence of original variables in different units.
2. Existence of differences in the variances, in case of original variables having the same units.

3.3.1.4.9.12 Eigen values and eigen vectors

The eigen values and eigen vectors were computed from data matrix. Eigen values define the amount of total variation that is displayed on principal components. The proportion of variation accounted for each principal component (PC) is expressed as the eigen value divided by the sum of the eigen values.

$$\text{Per cent variance explained for } PC_1 = \frac{\text{Eigen value (PC1)}}{\text{Sum of eigen values}}$$

The eigen vector (loading) defines the correlation of each variable with the principal components.

The principal components were identified by the following procedure.

The jth principal component (Y_i) of the observations X is the linear combination given as follows:

$$Y_j = A_{1j}X_1 + \ldots\ldots\ldots. + A_{pj} X_p$$

Where,

A_{ij} are found such that Y_j is uncorrelated $Y_1, Y_{2,}\ldots\ldots.Y_j\text{-}_1$ the jth largest variance. The A_{ij} are the elements of the normalized eigen vector associated with largest j^{th} eigen value. The variance of the jth principal component of the λ_j and the total system variance trace (S) = $\lambda_1+\lambda_2 +\ldots+ \lambda_{p.}$ The importantance of the jth principal component is given by

$$\frac{\lambda_j}{\text{Trace (S)}}$$

This is informative about the proportion of total variation that can be accounted for the jth principal component. The correlation between the ith original variable X_i and the jth principal component Y_j is given by

$$\rho(X_i, Y_j) = \frac{A_{ij}\sqrt{\lambda_j}}{\sqrt{S_i}}$$

Where,

S_i is the standard deviation of X_i.

Thus, a principal component is a linear function of the test variables as described below.

Principal component = $ax_1 + bx_2 + \ldots\ldots\ldots + hx_8$

Where, a,b,…. are coefficients and x_1, x_2 ….. etc., are the variables in such a way that the principal component has a unit variance as reported by Ehrenberg (1985).

PCA scores for each accession under concerned PCs were computed and utilized to derive a 3D (dimensional) scatter plot of individuals.

3.3.1.4.9.13 CLUSTER ANALYSIS

Agglomerative hierarchical clustering technique was followed as given by Anderberg (1993).

1. Obtaining data matrix

PCA scores for 83 accessions were used as input for clustering because principal component analysis provides variable independence and balanced weighting of traits, which leads to an effective contribution of different characters on the basis of respective variation.

2. Standardizing the data matrix

To compare the similarities among the accessions the data matrix was standardized with a column standardizing function *i.e.,* Q analysis. The data matrix was standardized in cluster analysis to make the characters contribute more equally to the similarities among accessions and to nullify the arbitrarily affect of the units chosen for measuring the attributes among the accessions.

Column standardizing function CA-Q analysis was carried by the following formula.

$$Zij = \frac{\overline{X}_{ij} - \overline{X}_{j}}{s_{ij}}$$

Where

$$\overline{X}_{j} = \sum_{i=1}^{n} X_{ij} / n$$

Where

$$Sij = \sum_{i=1}^{n} \frac{X_{ij} - Xj / n}{n-1}$$

For

n= total no. of accessions *i.e.,* 84 in the case

i= i^{th} accessions

j = j^{th} variables

The resulting data after standardization is unitless and have mean zero and variance one.

3. Computing the resemblance matrix

A resemblance coefficient, which measures the overall resemblance (the degree of similarity or distance) between a pair of accessions was computed.

The data matrix was transformed to distance matrix (resemblance matrix) based on the dissimilarity coefficients using squared Euclidean distance method.

$$\text{Squared Euclidean distance } [d_{ij}] = \sum_{K=1}^{p} (X_{ik} - X_{jk})^2$$

Where,

P = number of characters

X_{ik} = value of i th accession for k characters

X_{jk} = value of j th accession for k characters

4. Execution of the clustering method

Distance matrix was converted into dendrogram by using Ward's method. Ward proposed a clustering procedure seeking to form the partitions P_n, P_{n-1}…… P_1 in a manner that minimizes the loss associated with each grouping and to quantify that loss in a form that is readily interpretable. Information loss is defined by Ward's in terms of an error sum of squares criterion, ESS.

Cluster membership is assessed by calculated total sum of squared deviations from the mean of the cluster.

For the Kth cluster, Error sum of square is defined *i.e.,*

ESSK = Sum of squared deviations from the cluster centroid

If there are 'C' clusters, define the total error sum of squares as

ESS = Sum of ESSK,

for K = 1,….C. $\left(\sum_{k=1}^{C} ESSK\right)$

Ward's minimum variance method considers the union of every possible pair of cluster and combine the 2 clusters whose combinations results in the smallest increase in ESS. In this method, the distance between two clusters is the ANOVA sum of squares between the two clusters added up over all the variables.

3.3.1.4.9.14 Multivariate analysis

Multivariate analysis was carried out by subjecting the data of 21 growth and yield related attributes to principal component analysis as suggested by Iezzoni and Pritts (1991).Principal components with their vectors and eigen roots were, identified using the correlation coefficients of 21 variables. Further, their score was used for non hierarchical Euclidean cluster analysis. The analysis was done by using SPAR 1 statistical package.

3.3.2 DIVERSITY STUDIES IN TURMERIC ACCESSIONS USING MOLECULAR MARKERS (RAPD AND SSR MARKERS).

Molecular markers have emerged as powerful tool in understanding the genetic diversity in plants and animals alike, due to their inherent advantages. Molecular diversity was standardized among 83 turmeric genotypes using RAPD and SSR markers.

The details on preparation of stock solutions and buffers was given in appendix II. Steps involved in DNA isolation are described here under.

3.3.2.1 ISOLATION OF DNA FROM YOUNG LEAVES

DNA was isolated from 83 genotypes using the modified CTAB protocol developed by Syamkumar *et al.*2003 in the following steps.

1. One gram of fresh and young leaf tissue was weighed and cut into small pieces and transferred to a pre chilled mortor and pistle.
2. Leaf tissue was frozen with liquid nitrogen and made into fine powder.
3. The powder was then transferred to 50 ml Oakridge tube and added 8 ml preheated (65^0C) CTAB extraction buffer.
4. The mixture was incubated in a water bath at 65^0 C for 60 minutes with occasional mixing by gentle swirling.
5. Equal volume of chloroform: isoamyl alcohol (24:l) v/v was added, mixed well by gentle inversions for 15 minutes and then centrifuged at 12,000 rpm at 25^0C for 10 min.
6. The clear aqueous phase was pipetted out into another centrifuge tube and equal volume of ice cold isopropanol (kept at -20^0C) was added and mixed gently by inversions for 10-20 seconds so as to precipitate the DNA and kept in freezer (4^0C) for 30 minutes.
7. The precipitated DNA was hooked out using a sterile bent pasteur, washed with 0.5ml of 70% ethanol and then, spun at 8500 rpm for 2 min.
8. The supernatant is discarded and vacuum dried for 20 min and dissolved in 375 ul of 0.1X TE buffer (1mM Tris+0.1mM EDTA pH 8.0).

3.3.2.2 PURIFICATION OF GENOMIC DNA

The isolated DNA was purified as follows:

1. The RNA contaminants were eliminated from DNA by treating the DNA with 10µg/ml of RNAase and incubated at 37°C for 1minute in a water bath.

2. Equal volume of phenol: chloroform: isoamyl alcohol (25: 24: l), was added to the DNA solution and mixed well by repeated inversions and the mixture was centrifuged at 10,000 rpm (25^{0}C) for 15 minutes.
3. The clear aqueous phase was transferred to a sterilized 2 ml eppendorf tube extracted with equal volume of chlorofom: isomyl alcohol (24: 1) and centrifuged at 10,000 rpm (25^{0}c) for 10 minutes.
4. To the aqueous phase obtained after centrifugation added equal volume of chilled 100 % ethanol and incubated at -20°C for 1 hour and centrifuged at 13,000 rpm (4^{0}C) for 5 minutes
5. The supernatant was decanted carefully, the pellet is washed with 70% ethanol, air dried and dissolved in nuclease free water.

3.3.2.3 QUANTIFICATION OF DNA

The genomic DNA was quantified using electrophoretic comparision with standard samples of known concentration (Agarose gel analysis) and Spectrophotometric determination.

3.3.2.3.1 Agarose gel analysis

Quantity of the DNA was estimated by comparing the intensity of the DNA band of the samples with that of the standard λ DNA (50 ng) marker (Genei, Bangalore, India). Agarose gel electrophoresis was carried out on 0.8 per cent agarose gel to check the quantity and quality of isolated genomic DNA.

Preparation of agarose (0.8 %) gel

1. To prepare 0.8 per cent agarose gel, 0.8 g of agarose was weighed into a clean 100 ml conical flask and 100 ml of 1X TBE buffer was added (TBE buffer 0.89M Tris base, 0.02 M EDTA, 0.89 M boric acid,pH 8.0).

2. Agarose was boiled in microwave oven until all the agarose particles are completely dissolved and then cooled (60^0C). To this, 2.5 µl of ethidium bromide (10 mg / ml) was added and mixed well.
3. The ends of gel casting tray were sealed with insulation tapes and the melted agarose was poured; comb was inserted and allowed to solidify.
4. After solidification, the tape from either side was removed and the gel was immersed in the buffer tank containing 1X TBE buffer. Then, the comb was removed carefully without damaging the wells.
5. The DNA samples (5 µl) along with 6X loading/ tracking dye (6X loading/ tracking dye- 40 % sucrose, 0.025 % bromophenol blue,0.25 % xylene cyanol) was added, mixed and loaded into the well.
6. Around 3 µl of standard uncut λ DNA (50 ng/µl) was used as marker.
7. Electrophoresis was carried out at 60-70 V for 2 to 3 hrs until the bromophenol blue dye migrated to two third of the gel.
8. The gel tray was removed and the gel was observed under UV trans-illuminator and documented using Syngene Gel Documentation System. The quantity of the DNA was determined based on the intensity of the band relative to uncut λ DNA band.

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3.3.2.3.2 Quantification of genomic DNA by spectrophotometric analysis

The DNA samples were diluted (1:600) and the absorbance was read at 260 nm and 280 nm using a Spectrophotometer (Citizen).

The amount of DNA isolated was quantified and quality was checked spectrometrically at 260 nm and 280 nm as described by Sambrook and Russel (2001).

The ratio between the readings at 260 nm and 280 nm (OD 260nm / OD 280nm) was used an estimate of the purity of DNA samples. Pure preparations of DNA recorded 260nm / 280nm OD ratio between 1.73 and 1.99.

The concentration of DNA samples was further diluted to working concentration of 100 ng / µl and stored at -20 ^{o} C for further use in Polymerase Chain Reaction (PCR).

3.3.2.4 RAPD MARKER ANALYSIS

The PCR procedure outlined by Williams *et al.* (1990) was followed with slight modifications. A single decamer of arbitrary sequence was used in each PCR reaction. PCR reaction conditions were optimized to obtain informative and reproducible amplification profiles.

3.3.2.4.1 Screening of primers

Fifty four deca-mer RAPD primers sequenced at Bioserve Technologies, Hyderabad were initially screened using template DNA of 83 turmeric genotypes to identify the suitable primers for the study. In order to ensure their producibility, the primers generating weak or ambiguous patterns were discarded. Eventually, 21 primers were selected based on their ability to produce distinct, clearly resolved and reproducible amplicon profiles. The sequence information of 21 random 10-mer primers (Operon, USA) used in the study is furnished in Appendix IIIA.

3.3.2.4.2 Reaction mixture for RAPD-PCR

A 20 µl reaction was set up in sterile 0.2 ml microfuge tubes. The volume of reaction mixture per one reaction is as follows

Sterile distilled water	9.67µl
10 X reaction buffer	2µl
$MgCl_2$ (50 mM)	0.5µl
dNTP mix (2.5 mM)	2µl
Primer (10 pM)	2.5 µl
Template DNA (5 ng/µl)	3 µl
Taq DNA polymerase (3U /µl)	0.33 µl

Total reaction volume 20.00µl

3.3.2.4.3 Optimization of PCR programming for RAPD markers for amplification of turmeric DNA

Different temperature profiles and cycle repeats were tested for optimizing the PCR. For RAPD-PCR amplification, the following temperature profile was found to be optimum.

Step 1. Denaturation at 94^0C for 4 minutes

Step 2. Denaturation at 94 ^{0}C for 1 minute

Step 3. Annealing at 37^0C for 1 minute

Step 4. Polymerisation at 72^0C for 1 minute

Step 5. The steps 2 to 4 were repeated for 40 times

Step 6. Extended polymerization at 72^oC for 6 minutes

Step 7. Hold at 4^0C

3.3.2.4.5 SSR MARKER ANALYSIS

After quantification, the DNA was diluted to a concentration of 50 ng/µl for SSR analysis. Nineteen selected primers synthesized by Siju *et al.* (2010 and 2013) were used to analyze the inter-accession diversity in 83 accessions of turmeric for the analysis. (Appendix IIIB).

3.3.2.4.5.1 PCR amplification for SSR marker

A 15 µl reaction was set up in sterile 0.2 ml microfuge tubes. The volume of reaction mixture per one reaction is as follows

DNA(5 ng) 1.0 µl

Forward Primer (5pmoles) 0.5 µl

Reverse Primer(5pmoles)	0.5 µl
dNTP mix (2.5 mM)	0.5 µl
10X Taq buffer	2.5 µl
Taq DNA polymerase (3U /µl)	0.33 µl
Sterile water	9.67 µl
Total reaction volume	**15.00µl**

3.3.2.4.5.2 Optimization of PCR programming for SSR markers for amplification of turmeric DNA

Different temperature profiles and cycle repeats were tested for optimizing the PCR. For SSR amplification, the following temperature profile was found to be optimum.

Step 1. Denaturation at 94^{0}C for 5minutes
Step 2. Denaturation at 94 ^{0}C for 30 seconds
Step 3. Annealing at 55-59°C for 45 seconds
Step 4. Extension at 72^{0}C for 1 minute } 30 cycles
Step 5. Final Extension at 72^{0}C for 7 minutes
Step 6. Hold at 4^{0}C

3.3.2.4.5.3 Agarose gel electrophoresis

Amplified products of SSR primers and RAPD primers were separated on 3 per cent and 1.5 per cent agarose gel respectively stained with ethidium bromide (1 µg/ml of gel). The microscopic pores in the agarose gel act as a molecular sieve.TAE buffer 0.50 X was used as running buffer for electrophoresis. 3 µl of gel loading dye (Bromophenol blue) was added to 20 µl of RAPD and 15 µl of SSR PCR product and mixed well before loading into wells. 1 kb and100 bp (Genie) ladders were loaded as markers. Electrophoresis was carried out at 100 volts for 3 hours and the gel was documented under UV light using Syngene gel documentation system.

3.3.2.4.5.2 Scoring and statistical analysis for molecular analysis

Data was entered using a matrix in which all observed bands or characters were listed. The RAPD and SSR pattern of each accession was evaluated; assigning character state '1' to all the bands that could be reproducible and detected in the gel and '0' for the absence of band for each primer and used to calculate a genetic similarity matrix using the Jaccard (J) coefficient, which is more appropriate for dominant markers as it does not count 0/0 matches in the calculation. The genetic distance between each pair of accessions was calculated by SIMQUAL analysis of the NTSYS-pc software package version 2.1 (Rohlf, 2000).

Jaccard's Co-efficient (S_J) was computed as;

$$(S_J) = n_{AB}/n_A + n_B$$

n_{AB} is the number of bands common for samples A and B, n_A is the total number of bands in sample A and n_B is the total number of bands in sample B.

To measure the informativeness of the markers, the polymorphism information content (PIC) for each SSR marker was calculated according to the formula (Botstein *et al.*, 1980):

$$PIC=1-\Sigma Pi^2 - \Sigma\Sigma Pi^2\ Pj^2$$

where 'i' is the total number of alleles detected for SSR marker and 'Pi' is the frequency of the ith allele in the set of total genotypes investigated and j = i+1. This formula gives us an indicator of how many alleles a certain marker has, and how much these alleles divide evenly.

3.3.2.4.6 CLUSTER ANALYSIS

Cluster analysis was performed for both RAPD and SSR similarity matrices using the Unweighted Pair Group Method using Arithmetic means (UPGMA) algorithm, from which dendrograms depicting similarity among accessions were drawn and plotted using NTSYS-pc. 2.1 Software (Rohlf 2000). This calculates the congruence between assays of values typically densitometric assays. As it compares curves as a whole, it is

independent of band definitions and is thus ideally suited for a quick comparison of pattern without first having to edit the bands. It is largely insensitive to relative concentrations, but is sensitive to differences in background. Jaccard's (1908) Co-efficient considers only the presence of band as similarity and hence, is more conservative in declaring genetic diversity.

CHAPTER - IV

RESULTS AND DISCUSSION

The present study was focused on the genetic variability and divergence in turmeric germplasm by critically evaluating the extent of variability, association of characters and degree of divergence among them, at morphological and molecular levels using morphological and molecular markers.

Efficient use of conserved biodiversity, its characterization and quantification requires information about the degree and distribution of genetic diversity. Genetic diversity is the basis for genetic improvement. Knowledge of germplasm diversity has a significant impact on genetic improvement of crop plants. The variation in the genetic make-up, interaction with environment, dictates the observable pattern of diversity being shown by most of the living organisms. The genetic variation within and between species, generated by the process of mutation, sexual reproduction and selection, ensures its capacity in evolutionary change and ecological adaption.

Development of varieties with high yield levels coupled with desired quality attributes requires information on the nature and extent of variability available in the germplasm collections. Germplasm characterization is an important link between conservation and utilization of the plant genetic resources. For any breeding programme, genetic diversity is the raw material to a breeder, since genetic variation determines the potential for making gains from selections and resolving their phylogenetic relationships. In this regard, studies providing information about the diversity in germplasm would play an important role in crop improvement. For years, farmers have been the major contributors to crop diversity. Locally cultivated varieties well adapted to environmental conditions were the major criteria for long term biodiversity conservation.

Genetic variation among the yield attributes is the major concern. Yield is a complex trait influenced by genetic and environmental factors. Complexity of yield and

its components can be understood by employing various techniques available to plant breeders.

Considerable genetic variation in turmeric for various traits of economic importance has been reported by various workers Babu *et al.*1993, Lynrah *et al*. 1998, Hazra *et al*.2000, Shanmugasundaram *et al*.2000, Sinker *et al.*2005, Roy *et al.*2011, Jan *et al.*(2012), Jilani *et al.*(2012), Singh *et al*.(2012), Rajyalakshmi *et al.(*2013), Prajapati *et al.* (2014), Singh and Ramakrishna. (2014) and Verma *et al.* (2015).

A variety of tools and techniques have been developed based on the principles of genetics and statistics through which, the environmental effects can be largely nullified and real genetic effects are defined. Biometrical genetics like path coefficient analysis has the power to trace the causation and interpretation of results to plan the strategies for plant breeding programmes for necessary genetic improvements. In addition, the D^2 statistics and principal component analysis which give quantitative measurement of divergence among the genotypes and define the relative contribution of characters for differentiation of genotypes.

Turmeric breeding is coupled with numerous obstacles, due to its poor seed set. Normally high yielding varieties possess low curcumin content and vice versa. Hence development of varieties with high curcumin content is the prime concern as curcumin is an important factor for promotion turmeric export.

The present study contemplated to understand and explore the existing variability and genetic diversity in turmeric germplasm with eighty three (83) accessions including three checks. The findings pertaining to genetic variations and inheritance pattern would be helpful to the plant breeders in selecting the breeding approaches, genotypes and traits, more appropriately and efficiently.

The results of the experiments conducted are presented here under.

4.1 DIVERSITY STUDIES IN TURMERIC USING MORPHOLOGICAL MARKERS.

For a successful breeding program, the presence of genetic diversity and variability play a vital role. Genetic diversity is essential to meet the diversified goals of plant breeding such as breeding for increasing yield, wider adaptation, desirable quality, pest and disease resistance. Genetic divergence analysis estimates the extent of diversity existed among selected genotypes.

4.1.1 ANALYSIS OF VARIANCE

The results of the analysis of variance (ANOVA) revealed the presence of significant differences among the tested genotypes for all the characters studied *viz.,* plant height, basal stem diameter, number of leaves, leaf length, leaf width, leaf area, number of tillers, length of leaf petiole, number of primary rhizomes, length of primary rhizomes, number of secondary rhizomes, yield per plant, yield per hectare, curing percentage and days to maturity and this justified the need to estimate genetic distance values for the genotypes. In turmeric, the genotypes perform well not only in plant morphological characters but also in rhizome characters to gain higher yield. The yield should be in compensation with good quality to meet the export standards. As the crop is vegetatively propagated, any new variability can be fixed immediately. Systematic screening of genotypes assist in developing the lines with desired quality and agronomic traits.

Analysis of variance indicated that there was a significant difference among the accessions for plant height, basal stem diameter, number of leaves, leaf length, leaf width, leaf area, number of tillers, length of leaf petiole, number of primary rhizomes, length of primary rhizomes, number of secondary rhizomes, yield per plant, yield per hectare, curing percentage and days to maturity (Table 2).

4.1.2 MEAN PERFORMANCE OF TURMERIC ACCESSIONS

Eighty three accessions of turmeric including three checks *viz.,* CLS 269, KTS-3 and IISR Prathibha were studied for their morphological diversity. Observations were recorded for different quantitative and qualitative characters, as they are important for plant description and mainly influenced by natural selection, socio economic scenario and consumer preference (Jan *et al.* 2012). The overall appearance of most of the *Curcuma* species found to be very similar as they differ only in small morphological details.

4.1.2.1 QUANTITATIVE CHARACTERS

The results of the twenty one quantitative characters are explained here under and presented in Table 3a and 3b.

4.1.2.1 .1 Growth Parameters

4.1.2.1 .1 .1 Plant height (cm)

The plant height measured at 180 days after planting varied significantly among the genotypes ranging from 92.26 cm (IC - 420474) to 190.53cm (Ernad chand) with a mean value of 146.8 cm. Among the three checks, IISR Prathibha recorded more plant height (184.2 cm) followed by CLS- 269 (180.9 cm) and KTS-3 (175.74 cm). Out of eighty accessions, Ernad Chand (190.53 cm), T Sundar (188.33 cm), KTS-7 (188.03 cm) and CLL-335 (186.33 cm) were found to record significantly taller plants (Table 3a) than IISR Prathibha, whereas Salem (183.00 cm) and KTS-6 (183.63 cm) recorded more plant height than CLS - 269 and the accessions Duggirala (180.23 cm) Dindigam (178.4 cm) Cuddapah local (176.03 cm) and Amritpani (176.2 cm) were found to be superior in plant height than KTS-3. Remaining all the accessions recorded lower plant height than the three checks. Similar range of plant height in turmeric was reported by Indiresh *et al.* (1992), Lynrah *et al.* (1998),Singh *et al.* (2003), Singh *et al.* (2012), Jan *et al.* (2012), Jilani *et al.* (2012), Rajyalakshmi *et al.* (2013) and Singh *and Ramakrishna* (2014).

4.1.2.1 .1 .2 Basal stem diameter (cm)

The basal stem diameter of the eighty accessions of turmeric varied significantly with a mean value of 10.7 cm (Table 3a). Among all the accessions, T Sundar recorded significantly highest basal stem diameter (16.89 cm) followed by Vontimitta (13.57 cm) and the lowest stem diameter was recorded in IC-033007 (6.84 cm). Among the three checks, IISR Prathibha recorded more basal stem diameter (12.71 cm) followed by KTS-3 (12.41 cm) and CLS- 269 (10.88 cm). Out of 80 accessions, T Sundar (16.9 cm), Vontimitta (13.58 cm) and CL -7 (13.99 cm) were found to record significantly thickest stem compared to IISR Prathibha, whereas, GL Puram (13.06 cm) and CC- 9401 (13.1cm) recorded more basal stem diameter than KTS -3 and eleven accessions have recorded more basal stem diameter than CLS- 269. Similar trend in basal stem diameter was reported by Singh *et al.* (2012).

4.1.2.1.1.3 Number of leaves

The number of leaves at 180 days after planting recorded significant variation among the genotypes. The difference in number of leaves ranged from 4.48(CLL-335) to 10.31(BSR-2) with a mean of 7.89 (Table 3a). Among the checks IISR Prathibha recorded more number of leaves (9.53). KTS-8 (10.11), Ernad Chand (10.11), Badipadar (9.91) recorded significantly more number of leaves than IISR Prathibha. Similarly sixteen accessions were found to record more number of leaves than CLS-269 (8.32) and less than IISR Prathibha. Remaining all the accessions recorded less number of leaves than the three checks. The trend was in agreement with Singh *et al.* (2003), Roy *et al.* (2011), Singh *et al.*(2012), Jan *et al.*(2012), Jilani *et al.*(2012) and Rajyalakshmi *et al.*(2013), who reported a similar range of number of leaves in turmeric.

4.1.2.1 .1 .4 Leaf length (cm)

Considerable genetic variation was observed in the length of leaf measured at 180 days after planting (Table 3a). The range of leaf length varied from 38.80 to 85.87 cm with a mean of 62.09 cm. The longest leaf was recorded in T Sundar (85.87cm), whereas shortest leaf was observed in IC- 332957 (38.80cm). Among the checks, IISR

Prathibha recorded the longest leaves (75.81 cm) followed by KTS-3 (72.22 cm) and CLS – 269 (68.38 cm). However, among all the eighty accessions, T Sundar (85.87 cm) recorded significantly longest leaves followed by KTS-7 (79.27 cm). Remaining all the accessions recorded smaller leaves than the checks. The results were in accordance with the earlier findings of Roy *et al.*(2011), Jan *et al.*(2012) and Sinker *et al.*(2005) with regard to range in the leaf length in turmeric.

4.1.2.1.1.5 Leaf width (cm)

Significant variation was observed in leaf width among the accessions recorded at 180 days after planting which was ranged from 10.34 (Flourescent) to 19.78 cm (CLL - 335) with a mean of 15.17 cm (Table 3a). Among the three check varieties, IISR Prathibha recorded wider leaves (17.13 cm) followed by KTS-3 (15.86 cm) and CLS -269 (15.28 cm). However, among the accessions CLL-335, T-Sundar, Chintapalli local-2, Chintapalli local-1, Avidi and CL -17 were found to record significantly wider leaves (Table 3a) than all the three checks. However, forty five accessions were having narrow leaves than the three checks. Similar range of leaf width in turmeric was reported by Roy *et al.* (2011), Jan *et al.* (2012) and Sinker *et al.* (2005)

4.1.2.1 .1 .6 Number of tillers per plant:

The number of tillers observed among the genotypes ranged significantly from 0.89 (IC-420606) to 4.39 (IC-394396) with a mean of 1.95. Out of eighty accessions, fifteen accessions recorded more number of tillers and thirty seven accessions have recorded less number of tillers than the three check varieties (Table 3a). A range of 0-7.27 number of tillers was reported by several workers *viz*., Babu *et al.* (1993), Lynrah *et al.* (1998), Singh *et al.* (2003), Singh *et al.* (2012) and Rajyalakshmi *et al.* (2013).

4.1.2.1 .1 .7 Leaf area (cm^2)

The leaf area recorded at 180 days after planting varied from 325.48 cm^2 (IC-332957) to 1205.18 cm^2 (T Sundar). Among the eighty accessions, fifty accessions have recorded less leaf area than the three checks. The range of leaf area was in accordance with the findings of Sinker *et al.* (2005), Lynrah *et al.* (1998) (Table 3a).

4.1.2.1 .1 .8 Length of leaf petiole (cm)

Among the eighty accessions, CL-12 has recorded significantly longest leaf petiole (24.66 cm) followed by CL-16 (21.66 cm). The shortest leaf petiole was recorded in Amritpani (4.33cm). Among the three checks, IISR Prathibha (11.48cm) has recorded longer leaf petioles followed by CLS-269 (11.11 cm) and KTS-3 (11.00cm). However, among the eighty accessions, twenty accessions have recorded longer leaf petioles and fifty nine accessions have recorded shorter leaf petioles than the three checks (Table 3a).

4.1.2.1 .2 YIELD PARAMETERS

4.1.2.1 .2 .1 Number of mother rhizomes

The number of mother rhizomes was found to have significant variations among the genotypes, ranging from 1.02 to 4.59 (Table 3b). Significantly highest number of mother rhizomes was recorded in IC-545139 (4.59), which was followed by BSR-2 (4.35). Least number of mother rhizomes was recorded in the accession in T Sundar (1.02). Among the three check varieties, more number of mother rhizomes were recorded in CLS-269 (2.83), followed by IISR Prathibha (2.7) and KTS-3 (2.64). However, the accessions, IC-545139 (4.59), BSR-2 (4.35), Movatupuzha (3.59), IC-033007 (3.49), IC-394903 (3.49), CL -16 (3.55), IC-330113 (3.79), IC- 416941 (4.45) and IC-212267 (4.45) recorded maximum number of mother rhizomes than the three checks.

4.1.2.1 .2 .2 Length of mother rhizome (cm)

The length of mother rhizome among the accessions ranged between 2.10 cm (Tenali) to 11.17 cm (Rajapuri) (Table 3b) with a mean 6.64cm. Among the three checks, KTS-3 has recorded longer mother rhizomes (6.81 cm) followed by IISR Prathibha (6.42 cm) and CLS-269 (6.07 cm). Out of eighty accessions nineteen have recorded relatively longer mother rhizomes than all the three check varieties.

4.1.2.1 .2 .3 Girth of mother rhizome (cm)

The girth of mother rhizome varied significantly from 5.20 to 15.40. The widest rhizome was registered in IC- 545131 and the rhizome which registered the least was Tenali (5.20cm). Among the three check varieties, IISR Prathibha has recorded plumpy mother rhizomes (10.71 cm) than KTS-3 (10.37cm) and CLS-269 (9.7cm). Among the eighty accession, significantly more girth of mother rhizomes was recorded in 28 accessions than the check varieties (Table 3b).

4.1.2.1 .2 .4 Number of Primary rhizomes

The number of primary rhizomes per plant varied from 0.31 (IC-211401 and IC -211642) to 9.98 (CLL 335) across the accessions. Among the three check varieties, IISR Prathibha was found to produce more number of primary rhizomes per plant (9.95) and none of the eighty accessions have recorded more number of primary rhizomes than any one of the checks.

4.1.2.1 .2 .5 Length of primary rhizome (cm)

Among the eighty accessions, the longest rhizome was recorded in the accession Avidi (12.27cm) followed by Chintapalli local-1 (11.47 cm) while the shortest primary rhizomes was recorded in IC- 212605 (4.69 cm) (Table 3b). The mean length of the primary rhizome was recorded as 7.97 cm. Among the three check varieties,

KTS-3 has produced longer rhizomes (7.5 cm) than IISR Prathibha (6.6cm) and CLS-269 (7.37 cm). However, thirty one accessions among the eighty accessions have produced longer primary rhizomes than the three check varieties.

4.1.2.1 .2 .6 Girth of primary rhizome (cm)

There was considerable variation among the turmeric accession with regard to girth of primary rhizome which ranged from 3.26 cm (IC-212605) to 11.32 cm (KTS-7) (Table 3b) with a mean value of 7.25 cm. Among eighty accessions, eleven accessions have recorded more circumference of the primary rhizome than the three checks.

4.1.2.1 .2 .7 Number of Secondary rhizomes

The variation for number of secondary rhizomes among the accessions varied between 0.63 (IC-211641) to 15.81 (Vontimitta) with a mean value of 6.39. The accession, Vontimitta was on par with Badipadar (13.70), Flourescent (13.65) and GS (12.65) (Table 3b). Among the three check varieties, maximum number of secondary rhizomes was recorded in IISR Prathibha (8.2), followed by CLS-269 (7.03) and KTS-3 (6.69) and among the eighty accessions of turmeric, eleven accessions have recorded more number of secondary rhizomes than IISR Prathibha, eight have recorded more number of secondary rhizomes than CLS-269 and none of the accessions was found to produce more number of secondary rhizomes than KTS-3.

4.1.2.1 .2 .8 Length of Secondary rhizome (cm)

There was a considerable variation among the eighty turmeric accessions for length of secondary rhizomes which ranged from 0.80cm to 10.44 cm with a mean of 4.75 cm**.** The longest being recorded in IC- 420606 (10.44cm) followed by IC-420474 (10.23cm), while the shortest was noticed in Movatupuzha (0.80 cm). The check variety, IISR Prathibha has recorded longest secondary rhizomes (6.84cm) than CLS 269 (6.55cm) and KTS-3 (5.91cm).

Among all the accessions, IC-212585 (8.45cm), IC-420474(10.23cm), IC-420606 (10.44cm), IC- 394903 (9.04cm) and Imphal local (9.85cm), recorded significantly longer rhizomes than IISR Prathibha (Table 3b) and remaining all the accessions have produced significantly shorter secondary rhizomes than the three checks.

4.1.2.1 .2 .9 Girth of secondary rhizome (cm)

The girth of the secondary rhizome among the eighty turmeric accessions studied, ranged between 1.85cm (Tekuripeta) to 10.65 cm (CLI-317) with a mean of 5.38 cm. The accession, CLI-317 was found to be on par with KTS-7 (9.57cm) Avidi (9.25cm) and Penakenametta (9.05 cm). CLI-317 (10.65cm) and KTS-7 (9.57cm) have produced plumpy secondary rhizomes than the check variety, IISR Prathibha (9.29cm) which has recorded more circumference of secondary rhizomes than KTS-3 (8.23cm) and CLS-269 (8.68cm). The accession, Penakenametta also recorded more girth (9.05 cm) of secondary rhizomes than KTS-3 and CLS-269. Remaining all the accessions has recorded thinner secondary rhizomes than all the three check varieties (Table 3b).

Considerable variation with respect to yield and yield attributing characters like number of mother rhizomes, length of mother rhizome, girth of mother rhizome, number of primary rhizomes, length of primary rhizome, girth of primary rhizome, number of secondary rhizomes, length of secondary rhizome, girth of secondary rhizome was reported and acknowledged with earlier studies by Babu *et al.* (1993), Lynrah *et al.* (1998), Singh *et al.* (2003), Cintra *et al.* (2005), Roy *et al.* (2011), Singh *et al.* (2012) Jan *et al.* (2012), Jilani *et al.* (2012), Rajyalakshmi *et al.* (2013) Singh and Ramakrishna (2014) and Verma *et al*. (2015).

Higher production of mother, primary and secondary rhizomes may be due to better growth and vigour in some genotypes, as a result they were highly correlated with yield. The rhizome growth was also attributed for better absorption of nutrients from the soil.

4.1.2.1 .2 .10 Yield/Plant (g)

There was considerable variability among the turmeric accessions with regard to yield per plant which varied from 162.75g to 755.42g (Table 3b) with a mean value of 352.9g per plant. However, the accession, CLL- 335 recorded significantly higher yield per plant (755.42g) followed by IC- 211641 and IC- 212606 (568.75g each). Among the three check varieties, IISR Prathibha recorded higher yield per plant (500.00g) followed by KTS-3 (433.75g) and CLS-269 (382.5g) (Table 3b). However, CLL- 335 (755.42g), IC- 211641 and IC- 212606, (568.75g each), T Sundar (560.42g) and GL Puram (555.42g) were found to record significantly superior yields when compared to all the check varieties, justifying that tall plants with longer and broader leaves will be an ideal plant type for high yield in turmeric. Similar findings were reported by Roy *et al.*(2011).

4.1.2.1 .2 .11 Yield (tha^{-1})

The eighty turmeric accession showed considerable genetic variation for yield per hectare ranging from 12.87 t ha^{-1} (IC-545139) to 59.72 t ha^{-1} (CLL-335) with a mean yield of 27.87 t ha^{-1} (Table 3b). CLL- 335 (59.72 t ha^{-1}) IC- 211641 and IC-212606, (44.96 t ha^{-1}) T Sundar (44.32 t ha^{-1}) and GL Puram (43.91t ha^{-1}) were found to record significantly higher yields per hectare when compared to all the check varieties.

The yield is governed by genetic and environmental factors and varies with the genotypes which is in corroboration with the findings of Rajesh kumar and Jain (1998), Mutyalanaidu and Murthy (2013) and Rao *et al.* (2006). Yield of any crop by and large depends on the vigour of the plant as indicated by various growth parameters like plant height, number of leaves and rhizome characters. Best growth normally results in high yield and is influenced by genetic and environmental factors too, under which the crop in grown. The highest yield (59.72 tha^{-1}) recorded in CLL- 335 may be attributed for

the active photosynthesis favouring accumulation and assimilation of carbohydrates, as this accession has recorded higher values for plant height, number of leaves and leaf area. This is in concordance with Rao *et al*. (2006), Indiresh *et al*. (1992) and Kurian and Nair (1996) in turmeric with regard to the relationship between yield and growth parameters.

4.1.2.1 .2 .12 Curing Percentage

The accessions differed significantly with respect to curing percentage that was shown to vary from 9.96% to 31.46 % with a mean of 21.92%. The accession IC-319621 (31.46%) exhibited significantly highest curing percentage, whereas the lowest was recorded in the accession CL-16 (9.96%). However, forty-eight accessions among the eighty accessions studied recorded more curing percentage than all the three check varieties and among the checks, maximum curing percentage was recorded in the variety, KTS-3 (20.33%) followed by IISR Prathibha (19.7%), and CLS -269 (18.58%) (Table 3b).

The large variation in curing percent among the genotypes was mainly due to genetic factors than the environmental. High curing percent was due to the production of slender rhizomes and retention of low moisture at harvest. Most of the short and medium duration accessions were having more curing percent than long duration types. This is in conformity with the findings of Reddy *et al*. (1989) Vijayalatha *et al*.(2005) and Shanmugasundaram *et al*.(2001). On contrary, Rao *et al.* (2004) reported high curing percent in Armoor, a long duration type and opined that curing percentage increased with increase in age and attains its peak at maturity. However the curing percentage depends on the genotype, duration, soil and nutrient management practices and agro climatic conditions.

4.1.2.1 .2 .13 Curcumin content (%)

Curcumin, chemically dicinnamoyl methane is an odourless yellow crystalline powder. The accessions in the present study indicated significant variation for curcumin content that varied from 1.36% to 6.73% with a mean of 3.09% (Table 3b). The accession, T Sundar possessed high curcumin (6.73%) content followed by Ochira (6.58%). The accession, IC-319621 recorded the least curcumin content (1.36%). Among the three check varieties, CLS-269 has recorded more curcumin content (3.58%) followed by IISR Prathibha (3.47%) and KTS-3 (3.2%). Among eighty accessions of turmeric T Sundar (6.73%), Ochira (6.58%), CLL-335(5.48%), Aleppysupreme (4.78%), Salem (4.58%), Ethamukkala (4.48%), Rajapuri (4.38%), Rajendra Sonia (4.38%), Flourescent (4.18%), Sugandham (3.88%), IC-394396 (3.78%), Amritpani (3.68%), KTS-6 (3.65%), CLI-317 (3.85%) CL-17(3.78 %) and Vontimitta (3.68%) recorded significantly higher curcumin content than CLS-269.

Imphal local (3.55%) Thodupuzha (3.58%), Chintapalli local-2 (3.55%) and IC-540387 (3.55%) were found to record significantly more curcumin content than IISR Prathibha (3.47%).

Similarly, Ernad Chand (3.48%), Movatupuzha (3.48%), Dindigam (3.38), IC-211360 (3.45%), IC-319341(3.37%), IC-212578 (3.25%), GLpuram (3.48%) and Bataguda (3.38%) recorded significantly higher curcumin percentage than KTS-3. The curcumin content varied in different genotypes and it appears to be more dependent on ontogeny of the crop and genotypes. Apart from that this quality trait is highly sensitive to environmental micro and macro changes.

Several reports on Curcumin content showed contradictory results and variations in curcumin content in different agro climatic conditions have been reported (Pujari *et al*.(1987), Rakhunde *et al*.(1988), Ratnambal *et al.* (1986), Vijayakumar *et al*. (1992), Kurian and Valsala (1995), Kurian and Nair (1996), Lynrah *et al.* (1998), Pathania *et al.* (1988), Shanmugasundaram *et al*.(2001), Rao *et al.* (2004), Kumar *et al*.(2015) and Geethanjali *et al.(*2016).

4.1.2.1 .2 .14 Days to maturity

A significant variation was noticed for days taken for maturity of different accessions. The duration of the accessions extended from 186.17days to 247.5 days, Among the eighty accessions, the accession, Mydukur has taken more number of days (247.5 days) where as the accession, IC-420474 has taken less number of days (186.17days) for maturity (Table 3b).

Crop duration determines the cropping sequence to be adopted in a region. Among the eighty accessions, thirty nine were short duration types (180-210 days), thirty two were of medium duration (210-240 days) and nine accessions belonged to long duration types (>240 days). Among the check varieties, CLS-269 was found to be of short duration type, and both KTS-3 and IISR Prathibha were long duration varieties. However KTS-3 has taken significantly more number of days than the remaining two checks and none of the eighty accessions was found to take less number of days than KTS-3 for maturity.

The short duration genotypes exhibited moderate growth and yield while the medium and long duration excelled in plant and rhizome characters which can be ascribed due to the accumulation of more dry matter, maintenance of leaf area index and chlorophyll content for longer period, which was reflected in the final yield. However in Salem variety even though the duration was long (241.5 days), the yield was very low (13.74 t ha^{-1}) and the reason could be attributed that the growth and development of the genotypes highly depend on the climate conditions and soil factors under which they were grown and also the region specificity of the accession. These results are in line with the findings of Rao *et al.* (2006), Shanmugasundaram (2000), Reddy *et al.* (1989), Sasikumar *et al.* (1994) and Cholke (1993).

4.1.2.2 QUALITATIVE CHARACTERS

All the eighty three accessions including checks were classified according to the descriptors given by NBPGR and presented in the Table 4.

4.1.2.2.1 Leaf disposition pattern

Out of eighty three accessions the leaf disposing pattern was erect in forty five accessions (54.22%), semi erect in thirty eight accessions (45.78%) and none of the accessions showed horizontal leaf disposition pattern (Table 4).

4.1.2.2.2 Colour of dorsal surface of leaf

The colour of the dorsal surface of leaves among the eighty three accessions varied from green to dark green. Sixty accessions (72.29%) were having light green coloured leaves whereas, twenty three accessions (27.71%) produced dark green leaves (Table 4).

Breeding for green colour of leaf has been suggested as a method of increasing yield in cereal crops. Increasing light penetration into canopy has been suggested as one way of obtaining higher yield. Duncan (1971) also suggested that increased penetration of light into canopy would increase photosynthetic rate and increases the yield.

Syamkumar and Sasikumar (2007) and Jan *et al.* (2012) also pointed out dark green and light green colour of the leaves in turmeric.

4.1.2.2.3 Spatial arrangement of veins on leaves

In fifty accessions (60.24%), the veins on the leaves were close and in thirty three accessions (39.76%) the veins were distant (Table 4).

4.1.2.2.4 Prominence of leaf venation

The leaf venation was less prominent in forty two accessions (50.6%) and more prominent in forty one (49.4%) accessions (Table 4).

4.1.2.2.5 Rhizome core colour

Among the eighty three accessions studied, it was observed that seventeen accessions (20.48%) possessed light yellow coloured rhizomes, nineteen accessions (22.89%) were having lemon yellow coloured rhizomes, thirty three accessions (39.76)% exhibited light orange yellow coloured rhizomes and fourteen accessions (6.87%) have produced dark orange yellow coloured rhizomes (Table 4). In most of the short duration accessions the rhizome core colour was lemon yellow whereas, in medium and long duration types the rhizome core colour varied from light yellow to dark orange yellow.

There was a large variation in the rhizome core cololur in different *Curcuma* species studied by Cintra *et al*.(2005). Syamkumar and Sasikumar (2007) and Jan *et al.* (2012). Syamkumar and Sasikumar (2007) reported creamy white, pale blue, blue black, pale yellow, pale yellow with bluish green outer ring, lemon yellow and orange yellow coloured rhizomes in different *Curcuma* species.

4.1.2.2.6 Flowering

Among the eighty three accessions, twenty nine accessions (34.94%) have produced flowers and in the remaining fifty four accessions (65.06%) the flowering was absent. Flowering behavior of the *Curcuma* species also varies with the environmental conditions. Syamkumar and Sasikumar (2007), Jan *et al.* (2012) and Kandiannan *et al.* (2015) opined that the flowering behavior of the *Curcuma* species varies with the environmental conditions (Table 4).

4.1.3 GENETIC VARIABILITY STUDIES

4.1.3.1 VARIABILITY

Genetic variability is the prime concern for the breeder for selection and isolation of superior types. A very good response to selection is possible only when divergent genotypes are pooled in the breeding stock.

For the improvement of any crop particularly of its yield, it is essential to have the knowledge of genetic variability for the characters that are associated with the yield. The extent of genetic variability available in the crop could be of immense value to the breeders to effectively design the breeding programmes for characters under improvement (Singh and Chaudhary, 1977). The genotypic coefficient of variation, measures the range of variability available in crop species and also enables to compare the amount of variability present in different characters. However, the phenotypic expression of a character is the result of interaction between the genotype and environment. Hence, the total variance needs to be partitioned into heritable and non-heritable components so as to assess the true breeding nature of a particular trait (Falconer, 1964).

Besides genetic variability, the knowledge on heritability and genetic advance, measures the relative degree to which a character is transmitted to its progeny, thereby helps the breeder to employ a suitable breeding strategy to achieve the objective quickly. Therefore, for successful improvement of any crop, it is necessary to have a thorough knowledge on the variability together with the heritability which would give a better idea on the amount of genetic advance expected out of selection.

Burton (1952), Swarup and Chaugle (1962) indicated that genetic variability with heritability estimates would give a better idea on the amount of genetic advance expected out of selection. Panse (1957) suggested that magnitude of heritable variability is the most important aspect of the breeding material which has close relationship on its response to selection.

The genetic parameters, *viz.,* phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability in broad sense and genetic advance as per cent mean were estimated for all the characters under study (Table 5 and Fig 1). The germplasm lines exhibited a wide range of variability for all the characters.

Analysis of variance revealed highly significant variability for all the characters under study. The PCV estimates were higher than the GCV estimates indicating the influence of environment on the expression of these characters and possibility of obtaining very high selection response in respect of these characters.

The highest percentage of phenotypic and genotypic coefficient of variation was observed for number of secondary rhizomes (51.41 and 47.55 respectively) followed by length of leaf petiole (34.69 and 30.845 respectively) and yield per plant (33.027 and 29.828 respectively). The lowest percentage of PCV and GCV was observed for days to maturity (5.141 and 2.88 respectively).

The magnitude of difference between PCV and GCV was less for curcumin content (0.01) followed by leaf width (0.15) and curing percentage (0.15), while the highest difference was observed for number of primary rhizomes (18.02) followed by girth of primary rhizomes (12.76). The highest PCV and GCV was observed for leaf area (24.38 and 23.55), number of tillers per plant (31.71 and 30.59), number of mother rhizomes (26.9 and 20.29), and curcumin content (30.2 and 30.19) indicating the large influence of environment on the expression of these characters, whereas, highest PCV and moderate GCV was noticed for girth of mother rhizomes (22.42 and 19.09), number of primary rhizomes (29.71 and 11.71), length and girth of secondary rhizome (25.89 and 18.57), yield (25.92 and 17.3), curing percentage (20.03 and 19.88). Moderate PCV and GCV estimates were observed for plant height (16.21&15.83), basal stem diameter (14.03&13.43), leaf length (17.00&16.34), leaf width 12.24&12.09), length of mother rhizome (19.24&10.33), and length of primary rhizome (15.95&13.26). Whereas moderate PCV and low GCV was recorded for number of leaves (10.18&9.5), The PCV and GCV estimates were very low for days to maturity (5.141 and 2.878 respectively).

The PCV estimates were high for number of secondary rhizomes than GCV indicating that the influence of environment on the expression of this character.

The variability pattern was similar to those reported by earlier workers (Pathania *et al.*(1988), Indiresh *et al.*(1992), Babu *et al.(*1993), Lynrah *et al.*(1998), Singh *et al.*(2003), Sinker *et al.*(2005), Rao *et al.*(2004), Jan *et al*.(2011), Jan *et al.* (2012), Rajyalakshmi *et al*.(2013), Singh *et al.*(2012), Singh and Ramakrishna (2014), Prajapati *et al.*(2014) and Verma *et al.* (2015) and Jayasree *et al.* (2014).

4.1.3.2 HERITABILITY

The heritability estimates help the breeders in selection based on the phenotypic performance. Although PCV and GCV reveal the extent of genetic variability present, they do not indicate the extent of heritable variation. So that the selection can be more effective. The practical implications in breeding programmes are based on magnitude of heritable variation of the character concerned.

Out of 21 characters studied, all the characters have exhibited high heritability except number of mother rhizomes, length of mother rhizomes, number of primary rhizomes, girth of primary rhizomes, length and girth of secondary rhizomes and days to maturity. However, Philip &Nair (1985) reported high heritability for number of primary rhizomes and Singh *et al.* (2012) reported low heritability for number of mother rhizomes.

High heritability was observed for curcumin content (99.9%), curing percentage (98.5%), yield (81.6%), plant height (95.4%), basal stem diameter (91.2%), number of leaves (87.2%), leaf length (92.4%), leaf width (97.6%) leaf area (93.3%), length of leaf petiole (79.1%), girth of mother rhizome (72.5%) length of primary rhizome (69.1%) and number of secondary rhizomes (85.5%) indicating that, these characters are important in crop improvement as major part of the phenotypic variability in these characters was contributed by additive gene effects and hence improvement, can be made by simple selection. Similar findings were reported by Pathania *et al.* (1998), Babu *et al.*(1993),

Lynrah *et al*. (1998) Singh et al.(2003), Singh *et al*.(2012), Rajyalakshmi *et al*. (2013), Singh and Ramakrishna (2014), Jayasree *et al.* (2014) and Verma *et al.* (2015). Moderate estimates of heritability was recorded for the traits, number of mother rhizomes (56.9%), length of secondary rhizomes (51.4%) and days to maturity (31.3%) indicating the considerable influence of environment. Low estimates of heritability were recorded for length of mother rhizomes (28.8%), number of primary rhizomes (15.5%) and girth of primary rhizomes (12.2)%.

Highest estimates of heritability observed for curcumin content (99.09) was in conformity with the study reported by Lynarh *et al*.(1998),Pathania *et al.*(1998) declaring the possibility of improvement of fresh turmeric yield through clonal selection for rhizome yield and its component characters and suggesting that clonal selection for curcumin content would be more effective.

High heritability for plant height and leaf width as well as moderate to low heritability for yield have been reported by Babu *et al.*(1993), while, Prajapati *et al*. (2014) registered low heritability for plant height, leaf length, leaf width, number of leaves, number of tillers and curcumin content.

4.1.3.3 GENETIC ADVANCE AS PER CENT MEAN

Heritability along with genetic advance is more helpful in predicting the gains under selection than heritability estimated alone (Johnson *et al*.1955).In the present study, genetic advance as percentage mean ranged from 3.320 (days to maturity) to 90.575 (number of secondary rhizomes).

High heritability along with high genetic advance was observed for plant height, stem diameter, leaf length, leaf width, leaf area, number of tillers, length of leaf petiole, number of mother rhizomes, girth of mother rhizomes, length of primary rhizomes, number of secondary rhizomes, length of secondary rhizomes, yield, curing percentage and curcumin content, indicating the predominance of additive genes and pave the way

for improvement of those characters in individual plant selection. Similar findings were reported by Pathania *et al*.(1988), Lynrah *et al*.(1998) and Singh and Ramakrishna (2014). Heritability and genetic advance were observed to be high for rhizome yield and number of tillers per plant (Rajyalakshmi *et al*. (2013). According to Manohar *et al*. (2004), there was moderate to high heritability and genetic advance for cured yield, fresh weight of mother rhizomes and number of secondary rhizomes.

High variability with appreciable genetic advance was reported for rhizome yield, days taken for maturity, number of leaves, number of fingers, yield of secondary rhizomes and plant height and highest percentage of genetic advance as percent mean was recorded for number of secondary rhizomes by Jan *et al*. (2012).

This study was in conformity with Philip & Nair (1985), Sinker *et al.* (2005) and Singh (2013).

Low heritability coupled with low genetic advance was noticed for days to maturity indicating the large influence of environment for the expression of this character. Similar findings were reported by Singh (2013).

4.1.4 CORRELATION STUDIES

The ultimate goal of crop improvement in turmeric is to achieve a higher level of rhizome yield. Being a complex trait, the rhizome yield is largely influenced by many component characters. So information on strength and direction of correlation of these component characters on rhizome yield and association among the characters would be useful in designing breeding programmes for the improvement of yield.

The relationship between yield and its component characters is likely to vary according the genetic material used, environment under which the material is evaluated as well as due to interaction of the factors. Therefore it is worthwhile to study the association between the variables for identification of important yield components, so

that the weightage can be given to the characters of importance in further breeding programmes (Johnson *et al.*(1955).

In the present study, the interrelationships between twenty-one characters were established through their correlations both at phenotypic and genotypic levels and observed that, for all the characters studied, the genotypic correlation coefficients were higher than phenotypic correlation coefficients thus revealing a strong association among the characters (Tables 6 and 7).

4.1.4.1 Association of plant and rhizome characters on yield per plant

The rhizome yield per plant exerted a positive correlation both at phenotypic and genotypic levels for all the characters studied and is highly and significantly correlated with number of primary rhizomes (r_p=0.534, r_g=0.688), plant height (r_p=0.477, r_g =0.509), basal stem diameter (r_p=0.491, r_g =0.540), number of leaves (r_p=0.209, r_g=0.204), leaf length (r_p=0.321, r_g=0.379), leaf width (r_p=0.119, r_g =0.145), leaf area (r_p=0.290, r_g= 0.345), number of tillers per plant only at genotypic level (0.081), length of mother rhizome (r_p=0.298, r_g=0.569), length of primary rhizome (r_p=0.195, r_g=0.288), girth of primary rhizome (r_p=0.391, r_g=0.739), number of secondary rhizomes (r_p=0.249, r_g =0.276), length of secondary rhizome (r_p=0.045, r_g=0.092), girth of secondary rhizome (r_p=0.282, r_g=0.169), curcumin content (r_p=0.325, r_g=0.365) and days to maturity (r_p=0.138, r_g =0.483). The accessions with more height, longer and broader leaves with more number of primary and secondary rhizomes would be an ideal plant type for high rhizome yield. These correlations were in support of the studies conducted by Hazra *et al.* (2000), Tomar *et al.*2005), Shoba *et al.* (2011), Roy *et al.* (2011) Singh (2013) and Verma *et al.*(2015) that tall plants, having with higher number of suckers per plant (Tables 6 and 7).

The negative correlation was expressed with number of tillers per plant, (r_p=-0.013) only at phenotypic level, length of leaf petiole (r_p=-0.037,r_g=-0.196), number of

mother rhizome (r_p=-0.185 ,r_g =-0.069), girth of mother rhizomes (r_p= -0.012 , r_g = -0.054) and curing percentage. (r_p=-0.263 , r_g =-0.287).

4.1.4.2 Plant height

Both at phenotypic and genotypic levels, plant height exhibited significantly positive correlation with basal diameter (r_p=0.593, r_g=0.611), number of leaves, (r_p=0.509, r_g =0.579), leaf length (r_p=0.744, r_g=0.768), leaf width (r_p=0.253, r_g =0.268), number of tillers (r_p=0.136, r_g =0.154), leaf area (r_p= 0.638 , r_g =0.664), length of mother rhizome (r_p= 0.565 ,r_g =0.747),girth of mother rhizome (r_p=0.616, r_g=0.745), number of primary rhizomes (r_p=0.114, r_g=0.082), length of primary rhizome (r_p=0.242, r_g=0.311), girth of primary rhizome (r_p=0.548, r_g=0.611), number of secondary rhizomes (r_p=0.075, r_g =0.084), girth of secondary rhizome (r_p=0.347, r_g=0.441), curcumin content (r_p= 0.326, r_g =0.337) and days to maturity (r_p=0.108, r_g =0.333).

A negative correlation was observed for length of leaf petiole (r_p=-0.126, r_g=-0.181), number of mother rhizomes (r_p=-0.135, r_g=-0.202), length of secondary rhizome (r_p=-0.008) at phenotypic level and curing percentage at phenotypic and genotypic levels (r_p=-0.228, r_g=-0.232). Plant height exhibited positive correlation with length of secondary rhizome at genotypic level (r_g=0.009) (Tables 6 and 7).

4.1.4.3 Basal stem diameter

The phenotypic and genotypic correlations between basal stem diameter and leaf area was highly significant and positively correlated with leaf area (r_p=0.68, r_g=0.739). It was also positively correlated with number of leaves (r_p=0.521, r_g=0.598) leaf length (r_p=0.596, r_g=0.648) leaf width (r_p=0.488, r_g =0.524), length of leaf petiole (r_p= 0.067, r_g =0.106), length of mother rhizome (r_p= 0.399, r_g =0.698), girth of mother rhizome (r_p=0.479, r_g=0.497), length of primary rhizomes (r_p=0.336, r_g=0.384), girth of primary rhizome (r_p=0.0.369, r_g =0.392), number of secondary rhizomes (r_p=0.249, r_g=0.247), length of secondary rhizome (r_p=0.129, r_g =0.146), girth of secondary rhizome (r_p=0.300 ,

r_g =0.436), curcumin content (r_p=0.067, r_g=0.072) and days to maturity (r_p=0.131 , r_g =0.205), whereas it was negatively correlated with number of tillers (-0.091), number of mother rhizomes (-0.116), and number of primary rhizomes (-0.12) (Tables 6 and 7).

4.1.4.4 Number of leaves

The genotypic and phenotypic correlation coefficients indicated that there was positive correlation between number of leaves and leaf length (r_p=0.445, r_g =0.471), leaf width (r_p=0.480, r_g=0.513), number of tillers (r=0.062 , r_g =0.074), leaf area (r_p= 0.554, r_g=0.592), length of mother rhizome (r_p=0.306, r_g =0.529), girth of mother rhizome (r_p=0.346, r_g=0.416), number of primary rhizomes (r_p=0.137, r_g=0.348), length of primary rhizome (r_p=0.255, r_g=0.382), girth of primary rhizome (r_p=0.325, r_g =0.881), number of secondary rhizomes**,** (r_p=0.076, r_g=0.046), length of secondary rhizome (r_p=0.019, r_g =0.146), girth of secondary rhizome (r_p=0.211, r_g =0.475), curcumin content (r_p=0.212, r_g =0.225) and days to maturity (r_p=0.070 ,r_g=0.164). Curing percentage exhibited a positive correlation at phenotypic level and negative correlation at genotypic level (r_p=0.004, r_g = -0.016) (Tables 6 and 7).

The negative association was observed for length of leaf petiole (r_p=-0.100, r_g =-0.149) and number of mother rhizomes (r_p=- 0.294, r_g =-0.296).

4.1.4.5 Leaf length

The phenotypic and genotypic levels indicated highly significant positive correlation with leaf area (r_p=0.872, r_g=0.866) and it was also positively correlated with all the characters except length of leaf petiole (r_p=-0.098, r_g 0.152) and number of mother rhizomes (r_p=-0.107, r_g=-0.120)(Tables 6 and 7).

4.1.4.6 Leaf width

A positive relationship was observed between leaf width and leaf area (r_p=0.761, r_g =0.767), length of leaf petiole (r_p=0.185, r_g=0.206) length of mother rhizome (r_p=0.214, r_g=0.438), girth of mother rhizome(r_p=0.342, r_g=0.407), number of primary rhizomes (r_p=0.093, r_g=0.256), length of primary rhizome, (r_p=0.277, r_g =0.363) girth of primary rhizome (r_p=0.209, r_g =0.627) number of secondary rhizomes (r_p=0.130, r_g=0.132), length of secondary rhizome, (r_p=0.157, r_g =0.225),girth of secondary rhizome, (r_p=0.151, r_g =0.289) and curing percent, (r_p=0.145, r_g =0.140) .

The negative association was linked with number of tillers, (r_p= -0.109, r_g=-0.112) number of mother rhizomes (r_p=-0.129, r_g=-0.101) curcumin content (r_p=-0.153, r_g=-0.155) and days to maturity (r_p=-0.131, r_g=-0.253) (Tables 6 and 7).

4.1.4.7 Number of tillers

Both at phenotypic and genotypic levels number of tillers indicated positive correlation only with length of mother rhizome (r_p=0.079, r_g=0.226), and curcumin content (r_p=0.156, r_g=0.158) and at genotypic level for number of primary rhizomes (r_g =0.003). The parameter showed negative correlation with all the other characters at both genotypic and phenotypic levels (Tables 6 and 7).

4.1.4.8 Leaf area

Both at phenotypic and genotypic levels, leaf area indicated significant positive association with length of leaf petiole (r_p=0.028, r_g=0.002), length of mother rhizome (r_p=0.402, r_g=0.823), girth of mother rhizome (r_p= 0.557, r_g=0.668), number of primary rhizomes (r_p=0.026, r_g=0.184), length of primary rhizome (r_p= 0.297, r_g =0.461), girth of primary rhizome (r_p= 0.394, r_g=0.388),number of secondary rhizomes (r_p=0.134, r_g=0.141),length of secondary rhizome (r_p=0.117, r_g=0.228) and girth of secondary rhizome (r_p=0.281, r_g =0.478).

The negative association was linked with number of mother rhizomes (r_p=-0.02, r_g =-0.142) curing percentage (r_p=-0.003, r_g=-0.015), curcumin content (r_p=-0.015, r_g =-0.016) and days taken for maturity (r_p=- 0.027, r_g =- 0.063)

4.1.4.9 Length of leaf petiole

Length of leaf petiole exhibited a negative and non significant correlation at both phenotypic and genotypic levels with girth of primary rhizome (r_p=-0.011, r_g=-0.695) number of secondary rhizomes (r_p=-0.021, r_g=-0.024) and curcumin content (r_p=-0.164, r_g=-0.181), whereas positive significant correlations with number of mother rhizomes (r_p=0.086, r_g=0.209), length of primary rhizome (r_p=0.325, r_g =0.417), length of secondary rhizome (r_p=0.142, r_g=0.173), girth of secondary rhizome (r_p=0.114, r_g=-0.436) and curing percentage (r_p=0.064, r_g=0.067).

However, it exerted negative correlation at phenotypic level and positive correlation at genotypic level for length of mother rhizome (r_p=-0.024, r_g=0.19), girth of mother rhizome (r_p=-0.024, r_g=0.089) and days to maturity (r_p=-0.071, r_g=0.223), whereas, for number of primary rhizomes positive correlation at phenotypic level and negative correlation at genotypic level was observed (r_p=0.008, r_g=-0.083) (Tables 6 and 7).

4.1.4.10 Number of mother rhizomes

A significant and positive correlation was recorded between number of mother rhizomes and number of primary rhizomes (r_p=0.195, r_g=0.871) and curing percentage (r_p=0.008, r_g =0.056) at both phenotypic and genotypic levels, whereas, negative correlation was observed with all the other parameters (Tables 6 and 7).

However, it has shown positive correlation at genotypic level for girth of mother rhizome (r_g=0.056) and for length of secondary rhizome (r_g =0.046), negative correlation at phenotypic level for girth of mother rhizome (r_p=-0.046) and girth of secondary rhizome (rp =-0.101).

4.1.4.11 Length of mother rhizome

Length of mother rhizome was significantly and positively correlated with girth of mother rhizome (r_p=0.523, r_g=0.533), length of primary rhizome (r_p=0.102, r_g=0.895), girth of primary rhizome (r_p=0.219, r_g=0.289),number of secondary rhizomes (r_p=0.043, r_g=0.014), length of secondary rhizomes (r_p=0.016, r_g=0.766), girth of secondary rhizomes (r_p=0.141, r_g=0.310), curcumin content (r_p=0.296, r_g =0.552) and days to maturity (r_p=0.255, r_g=-0.956) (Tables 6 and 7).

The negative correlation was related with number of primary rhizomes (r_p=-0.039, r_g =- 0.873) and curing percentage (r_p=-0.242 , r_g=-0.461).

4.1.4.12 Girth of mother rhizome

Length of primary rhizome (r_p=0.409, r_g=0.506), girth of primary rhizomes (r_p=0.504, r_g=0.619), number of secondary rhizomes (r_p=0.207, r_g= 0.251), length of secondary rhizomes at phenotypic level (r_p=0.037) and girth of secondary rhizomes (r_p=0.297, r_g=0.521), curcumin content (r_p=0.319, r_g=0.379) and days to maturity (r_p=0.161, r_g=0.107) recorded significant and positive correlation with girth of mother rhizomes at both genotypic and phenotypic levels (Tables 6 and 7).

However, number of primary rhizomes (r_p=-0.213, r_g=-0.106), length of secondary rhizome at genotypic level (r_g=-0.023) and curing percentage (r_p=-0.313, r_g=-0.355) has shown negative correlation at both the levels.

4.1.4.13 Number of primary rhizomes

Number of primary rhizomes was significantly and positively associated with length of secondary rhizome at phenotypic level (r_p=0.074), curing percentage at both phenotypic and genotypic levels (r_p=0.204, r_g=0.561) and number of secondary rhizomes at genotypic level (0.219), whereas negative association was related with length and girth of primary rhizome, number of secondary rhizomes, girth of secondary rhizome, curcumin content and days to maturity (Tables 6 and 7).

4.1.4.14 Length of primary rhizome

Length of primary rhizome exerted positive correlation with girth of primary rhizome (r_p=0.406, r_g=0.732), number of secondary rhizomes (r_p=0.182, r_g=0.212), length of secondary rhizome (r_p=0.224, r_g=0.115), girth of secondary rhizome (r_p=0.424, r_g =0.313) at both phenotypic and genotypic levels and with curing percentage at genotypic level (0.037) (Tables 6 and 7).

The negatively associated characters were curing percentage at phenotypic level (r_p=-0.004), curcumin content (r_p=-0.132, r_g=-0.156) and days taken for maturity (r_p=-0.172 , r_g =-0.221).

4.1.4.15 Girth of primary rhizome

Both at phenotypic and genotypic levels, girth of primary rhizome registered positive correlation with number of secondary rhizomes (r_p=0.241, r_g=0.956), length of secondary rhizome (r_p=0.068, r_g=0.329), girth of secondary rhizome (r_p=0.517, r_g=-0.069) curcumin content (r_p=0.097, r_g=0.306) and days to maturity (r_p=0.126, r_g=0.174), whereas a negative association was recorded for curing percentage (r_p=- 0.279 , r_g=-0.782) (Tables 6 and 7).

4.1.4.16 Number of secondary rhizomes

Number of secondary rhizomes was positively correlated with length of secondary rhizomes (r_p=0.036, r_g=0.073), girth of secondary rhizomes (r_p=0.199, r_g =0.496), curcumin content (r_p=0.014, r_g=0.017) and days to maturity at phenotypic level (0.046), whereas, a negative association was recorded for curing percentage (r_p=-0.076, r_g=-0.092) and days taken for maturity at genotypic level (-0.149).

4.1.4.17 Length of secondary rhizome

Length of secondary rhizomes expressed a positive correlation with girth of secondary rhizomes (r_p=0.155) at phenotypic level alone and negative correlation with curing percentage (r_p=-0.053, r_g=-0.089) curcumin content (r_p=-0.143 ,r_g=-0.201) and days taken for maturity (r_p=-0.177) at phenotypic level, whereas, with girth of secondary rhizomes (r_g=-0.368) at genotypic level, it was having negative association. A positive correlation with days taken for maturity (r_g =0.117) at genotypic level was recorded ((Tables 6 and 7).

4.1.4.18 Girth of secondary rhizome

Girth of secondary rhizomes attributed a significant and positive correlation with yield (r_p=0.282, r_g =0.169) and curcumin content (r_p=0.091, r_g =0.162) at both phenotypic and genotypic levels and with days to maturity at genotypic level (r_g=0.962). The negative association was expressed with the characters curing percentage (r_p=-0.2528, r_g=-0.349) and days taken to maturity (r_p= -0.012) at phenotypic level.

4.1.4.19 Curing Percentage

Curing percentage exerted non significant correlation at both phenotypic and genotypic levels and was negatively associated with yield.

4.1.4.20 Curcumin content and days to maturity

Curcumin content (r_p=0.3245, r_g=0.3653) and days taken for maturity (r_p=0.1383, r_g=0.4826) were significantly and positively associated with yield at both phenotypic and genotypic levels.

4.1.5 SIMPLE CORRELATION COEFFICIENTS

A similar trend was noticed for simple correlation coefficients (Table 8 & Fig.2) indicating that the rhizome yield is significantly and positively associated with number of secondary rhizomes (0.345) followed by basal stem diameter (0.304), number of primary rhizomes (0.268), plant height (0.242), number of leaves (0.233), girth of primary rhizome (0.122), leaf area (0.083), leaf length (0.068), leaf width (0.059), number of tillers (0.033), length of mother rhizome (0.122), girth of mother rhizome (0.023), curcumin content (0.251) and days to maturity (0.129). The negative association was found with length of leaf petiole (-0.032), number of mother rhizomes (-0.136), length of primary rhizomes (-0.159), length of secondary rhizomes (-0.003), girth of secondary rhizomes (-0.154) and curing percentage (-0.071). Similar results were reported by Shanmugasundaram *et al.*(2001).

4.1.6 PATH COEFFICIENT ANALYSIS

The correlation coefficient between yield and a particular yield component is the net result of direct effect of that attribute and indirect effect through other yield contributing traits. Information on the direct and indirect effects on yield is important which is explicable by path analysis proposed by Wright (1921) and illustrated by Dewey and Lu (1959). The inter relationships of the component characters on yield provides likely consequences of their selection for simultaneous improvement of desirable characters with yield.

If the correlation coefficient between causal factor and the effect is almost equal to its direct effect, this correlation explains the true relationship and direct selection through this trait will be useful. If the correlation coefficient is positive, but its direct effect is negative or negligible, the indirect effect appears to be the cause of that positive correlation. In such situation, the other factors are to be considered simultaneously for selection. Sometimes, correlation coefficient may be negative, but the direct effect may be positive and high, under such circumstances, a restricted simultaneous selection has to be followed *i.e.,* restriction has to be imposed to nullify the undesirable indirect effect in order to make use of the direct effect.

In turmeric, the yield of rhizome per plant is the result of direct and indirect effects of several yield contributing characters. To know the contribution of various characters towards wet rhizome yield per plant, the significant correlation of different traits with rhizome yield is partitioned into their direct and indirect effects (Table 9 and Fig. 3).

4.1.6 .1 Direct effects

In the present study positive and direct effects on rhizome yield were high for basal stem diameter (0.370), number of primary rhizomes (0.278), followed by girth of mother rhizome (0.264), leaf width (0.220) and number of secondary rhizomes per plant (0.189), while it was low for leaf length (0.179), number of leaves (0.137) and negligible for plant height (0.097), length of leaf petiole (0.049) and girth of primary rhizome (0.072) (Table 9 and Fig. 3). Hence, direct selection based on basal stem diameter, number of primary and secondary rhizomes and leaf width would be most effective and reliable tool to identify productive genotypes of turmeric. Singh and Ramakrishna (2014) opined that positive direct effects on rhizome yield were high for leaf length and curcumin content, whereas, Shoba *et al*. (2011) reported that positive and direct effect on rhizome yield was maximum for weight of primary rhizomes followed by leaf length, number of leaves and weight of mother rhizomes.

The characters, leaf area exhibited direct and high negative correlation (-0.734) whereas length of primary rhizome(-0.307), girth of secondary rhizome (0.271) and length of mother rhizomes (-0.152) exhibited moderate to low direct negative path coefficients on yield, while, number of tillers per plant (-0.01), number of mother rhizomes (-0.004), length of secondary rhizomes (-0.018), curing percentage(-0.009), curcumin content (-0.01) and days to maturity (-0.082) exhibited negligible direct negative effect on yield. Though, leaf area and length of primary rhizomes exhibited negative correlation they have direct contribution towards yield.

4.1.6 .2 Indirect effects

4.1.6 .2.1 Plant height

The highest positive indirect effect was observed for leaf length (0.067) followed by leaf area (0.061), basal diameter (0.059), number of leaves (0.057), girth of primary rhizome (0.049), number of primary rhizomes (0.043), leaf width (0.034), curcumin content (0.031), length of mother rhizome (0.029), number of secondary rhizomes (0.025), girth of secondary rhizome (0.025), duration (0.022) and girth of mother rhizome (0.018). Other indirect negligible positive effects were observed for number of tillers (0.009), length of secondary rhizome (0.008), length of primary rhizome (0.007), number of tillers (0009) and length of leaf petiole (0.001) (Table 9 and Fig.3).

The indirect effect through number of mother rhizomes, and curing percentage was negative (-0.028 and -0.024 respectively).

4.1.6 .2.2 Basal stem diameter

The highest positive indirect effect for basal stem diameter was through leaf area (0.259) followed by plant height (0.225), leaf length (0.219), number of leaves (0.215), leaf width (0.207). The other indirect positive effects were through girth of

primary rhizome (0.144), number of primary rhizomes (0.142), number of secondary rhizomes (0.138), girth of secondary rhizome (0.099), length of mother rhizome (0.092), length of secondary rhizome (0.088), girth of mother rhizome (0.084), days to maturity (0.080) and curcumin content (0.021).

The negative indirect effect was observed through number of mother rhizomes (-0.128), number of tillers (-0.041) and curing percentage (-0.015) (Table 9 and Fig. 3).

4.1.6 .2.3 Number of leaves

The highest positive indirect effect for number of leaves was through plant height (0.08), leaf area (0.08), basal stem diameter (0.079), leaf width (0.070), leaf length (0.064), curcumin content (0.045), girth of primary rhizome (0.044), number of primary rhizomes (0.034), girth of secondary rhizome (0.031), length of secondary rhizome (0.03) and number of secondary rhizomes (0.029). The other positive indirect effects were observed through length of mother rhizome (0.011), girth of mother rhizome (0.015), days to maturity (0.017) and number of tillers (0.006). Curing percentage (-0.009) and length of leaf petiole (-0.004) have recorded indirect negative effect for number of leaves.

4.1.6 .2.4 Leaf length

This trait registered the highest positive indirect effect through leaf area (0.156), plant height (0.125), basal diameter (0.106), number of leaves (0.084), leaf width (0.074), girth of primary rhizome (0.062), girth of mother rhizome (0.052), number of primary rhizomes (0.052), girth of secondary rhizome (0.046), number of secondary rhizomes (0.045), length of mother rhizome (0.041), length of secondary rhizome (0.042) and days to maturity (0.022).

There was a negligible negative indirect effect through curcumin content (-0.007), number of tillers (-0.009), curing percentage (-0.014), length of leaf petiole (-0.018) and number of mother rhizomes (-0.031).

4.1.6 .2.5 Leaf width

Leaf width showed the highest indirect positive effect through leaf area (0.177), basal stem diameter (0.123), number of leaves (0.113), leaf length (0.091), girth of mother rhizome (0.083), number of primary rhizomes (0.081), girth of primary rhizome (0.081), plant height (0.076), length of primary rhizome (0.067), length of secondary rhizome (0.057), girth of secondary rhizome (0.072),length of mother rhizome (0.047), curing percentage (0.017) and number of secondary rhizomes (0.016).

The indirect negative effect was through number of mother rhizomes (-0.052), number of tillers (-0.022), curcumin content (0.016) and duration (0.022).

4.1.6 .2.6 Number of tillers

This trait registered negligible positive indirect effect through basal diameter, leaf length, leaf width, leaf area, length of leaf petiole, number of mother rhizomes, girth of mother rhizomes, length of primary rhizomes, girth of primary rhizomes, number of secondary rhizomes, length of secondary rhizomes, girth of secondary rhizomes and days to maturity.

Negligible negative indirect effect was associated through plant height, number of leaves, length of mother rhizome, number of primary rhizomes, curing percentage and curcumin content.

4.1.6 .2.7 Leaf Area

The highest indirect positive effect was associated with number of mother rhizomes (0.185) followed by number of tillers (0.058), curcumin content (0.044) and curing percentage (0.007)

The greatest negative indirect effect was found for leaf length (-0.637) followed by leaf width (-0.589) basal stem diameter (-0.514), plant height (-0.462), number of leaves (-0.428), length of leaf petiole (-0.048), length of mother rhizome (-0.188), girth of mother rhizome (-0.284), number of primary rhizomes (-0.279),length of primary rhizomes (-0.145),girth of primary rhizomes (-0.285), number of secondary rhizomes (-0.169),length of secondary rhizomes (-0.224),girth of secondary rhizomes (-0.257) and days to maturity (-0.020).

4.1.6 .2.8 Length of leaf petiole

This trait showed indirect positive effect with leaf width (0.012), length of primary rhizomes (0.010) and negligible indirect positive effect with plant height, basal diameter, leaf area, number of mother rhizomes, girth of mother rhizomes, girth of primary rhizomes and girth of secondary rhizomes.

The negative indirect effect was negligible for number of leaves, leaf length, and number of tillers, length of mother rhizomes, number of secondary rhizomes, length of secondary rhizomes, curing percentage, curcumin content and days to maturity.

4.1.6 .2.9 Number of mother rhizomes

The number of mother rhizomes has registered negligible indirect effect with all the traits.

4.1.6 .2.10 Length of mother rhizome

The highest positive indirect effect was observed through curing percentage (0.024), followed by girth of secondary rhizomes (0.021), number of mother rhizomes (0.011) and the least was for length of leaf petiole (0.001).

The indirect effects of plant height, basal stem diameter, number of leaves, leaf length, number of tillers, leaf area, girth of mother rhizomes, number of primary rhizomes, length of primary rhizomes, number of secondary rhizomes, length of secondary rhizomes, curcumin content and days to maturity were negative. The highest indirect negative effect was noticed for girth of mother rhizomes (-0.070) and lowest for curcumin content (-0.011).

4.1.6 .2.11 Girth of mother rhizome

The positive indirect effect for girth of mother rhizome was associated with length of secondary rhizomes (0.128), followed by length of mother rhizome (0.122) and other traits which were having positive indirect associations were leaf length (0.76), length of mother rhizomes (0.122), girth of primary rhizomes (0.103) leaf area (0.1), length of primary rhizomes (0.098), girth of secondary rhizomes (0.097), basal diameter (0.06), number of primary rhizomes (0.051), plant height (0.05), number of leaves (0.03), number of mother rhizomes (0.028) and length of leaf petiole (0.006).

The indirect effects were negative for number of tillers (-0.029), number of secondary rhizomes (-0.006), curing percentage (-0.013) , curcumin content (-0.029) and days to maturity (-0.003).

4.1.6 .2.12 Number of Primary rhizome

The number of primary rhizomes recorded the highest positive effect for plant height (0.124), followed by girth of primary rhizomes (0.120). The lowest was observed for the characters, days taken to maturity, basal stem diameter, number of leaves, leaf length, leaf width, leaf area, length of leaf petiole, length of mother rhizomes,

girth of mother rhizomes, number of primary rhizomes, length of primary rhizomes, number of secondary rhizomes, length of secondary rhizomes, girth of secondary rhizomes, curcumin content and days maturity and were found to be positive. Only curing percentage was having negative indirect effect (-0.041).

4.1.6 .2.13 Length of primary rhizome

The positive indirect effect was associated with curcumin content (0.079) followed by number of secondary rhizomes (0.069). Days to maturity (0.061) and number of tillers also associated with positive indirect effect.

The highest negative indirect effect was registered for girth of primary rhizomes, (-0.145), length of leaf petiole (-0.137), followed by leaf width (-0.926), girth of secondary rhizome (-0.091) and the lowest was in number of mother rhizomes (-0.015).

The other indirect negative effects were attributed for plant height ,basal stem diameter, number of leaves, leaf length, leaf area, length of mother rhizome, girth of mother rhizome, number of primary rhizomes, length of secondary rhizome, girth of secondary rhizome and curing percentage.

4.1.6 .2.14 Girth of Primary rhizome

This trait registered a positive indirect effect through plant height (0.037), followed by length of primary rhizome (0.034), number of primary rhizomes (0.031) and the lowest was observed for curcumin content (0.005)

The other positive effects were basal diameter, number of leaves, leaf length, leaf width, leaf area, length of mother rhizome, girth of mother rhizome, number of primary rhizome, length of primary rhizome, number of secondary rhizomes, length of secondary rhizome, girth of secondary rhizome, and days to maturity whereas the traits,

number of tillers, number of mother rhizomes and curing percentage were negatively accounted.

4.1.6 .2.15 Number of secondary rhizome

The character exhibited the highest indirect positive effect through basal diameter (0.071) followed by leaf length (0.049), plant height (0.049) and the lowest was observed by length of secondary rhizome (0.009).

The effects were also positive for number of leaves, leaf width, leaf area, length of mother rhizome, number of primary rhizomes, girth of primary rhizome, length of secondary rhizome, curcumin content and days to maturity. The negative indirect effect was highest through number of mother rhizomes (-0.059) followed by length of leaf petiole (-0.049) and the lowest was through girth of secondary rhizome (-0.007). The other negative effects were registered for number of tillers (-0.024), length of primary rhizome (-0.043), girth of mother rhizome (-0.005) and curing percentage (-0.031).

4.1.6 .2.16 Length of secondary rhizome

The highest negligible positive indirect effect was registered for number of mother rhizomes (0.003) and number of tillers (0.003). The other characters attributing for positive effect which are negligible were length of leaf petiole, curing percentage curcumin content and days to maturity.

The highest negligible negative effect was noticed for girth of mother rhizome (-0.009) followed by leaf area (-0.006). The other characters associated with negligible negative effect were plant height, basal diameter, number of leaves, leaf length, Leaf width, length of mother rhizome, girth of mother rhizome, number of primary rhizomes, length of primary rhizome, girth of primary rhizome, number of secondary rhizomes, and girth of secondary rhizome.

4.1.6 .2.17 Girth of secondary rhizome

The indirect effect of girth of secondary rhizomes was positive and highest for curing percentage (0.056), followed by number of mother rhizomes (0.046). The least effect was noticed for number of secondary rhizomes (0.010)

The other characters accounting positive effects were number of tillers, length of mother rhizome, curcumin content and days to maturity. The highest negative indirect effect was noticed for length of secondary rhizome (-0.137), followed by girth of mother rhizome (-0.099) and the lowest was observed for length of leaf petiole. The indirect effects were also negative for plant height, basal stem diameter, number of leaves, leaf length, leaf width, leaf area, number of primary rhizome, and length of primary rhizome and girth of primary rhizome.

4.1.6 .2.18 Curing percent

This trait exerted a positive indirect effect through curcumin content (0.003), while the least was positive and indirect effect for plant height (0.002), girth of primary rhizomes (0.002), number of secondary rhizomes (0.002), girth of secondary rhizome (0.002), number of mother rhizomes (0.002), days to maturity (0.002) and number of primary rhizomes (0.001), where as the negligible positive indirect effect was through basal stem diameter, number of leaves, leaf length, leaf area, length of leaf petiole, girth of mother rhizome and length of secondary rhizome.

The indirect effect of curing percentage was negative and negligible for number of mother rhizomes (-0.001) length of primary rhizome (-0.001) leaf width (-0.001) and number of tillers (-0.0002).

4.1.6 .2.19 Curcumin content (%)

The effect was positive and indirect for curing percentage (0.003) and length of primary rhizomes (0.003), followed by number of mother rhizomes (0.002) and the lowest was observed for length of leaf petiole, leaf length, leaf width, leaf area, girth of mother rhizomes, length of secondary rhizomes and girth of secondary rhizomes which were also positively associated. The negative indirect effect was highest for plant height (-0.003) number of leaves (0.003), followed by days to maturity (-0.0025).The lowest effect was found for basal stem diameter (-0.0005). The lowest effect was also recorded for number of tillers, length of mother rhizome, number of primary rhizomes, girth of primary rhizome and number of secondary rhizomes.

4.1.6 .2.20 Days to maturity

Among all the characters, curing percentage has recorded the highest (0.021) positive indirect effect for days to maturity followed by length of primary rhizome (0.016). Lowest was observed for girth of mother rhizome (0.001).The lowest, positive indirect effect was also observed for number of tillers, leaf width, length of leaf petiole, number of mother rhizomes, length of secondary rhizome and girth of secondary rhizome. The highest negative indirect effect was associated with number of secondary rhizomes (-0.022) followed by curcumin content (-0.021), length of mother rhizome (-0.019) and the lowest in leaf area (-0.002). Plant height, basal stem diameter, number of leaves, leaf length, leaf area, number of primary rhizomes, girth of primary rhizome were also negatively associated.

Similar results were confirmed by various workers (Radha Krishnan *et al.* (1995), Hazra *et al*.(2000), Singh *et al*.(2008), Shobha *et al*.(2011), Jan *et al.* (2012), Prajapati *et al*.(2014), Verma *et al*.(2015) and Singh and Ramakrishna (2014).

4.1.7 GENETIC DIVERGENCE STUDIES

The success in obtaining highly variable genotypes and creating greater variability for efficient selection in a breeding programme depends to a larger measure on the degree

of divergence. One such tool is the D^2 statistics that has found favour for estimating genetic divergence. The importance of the choice of the characters has been stressed since they reject the usefulness of D^2 analysis.

The D^2 statistics measures the forces of differentiation at low levels namely intra and inter cluster levels and thus helps in the selection of genetically divergent genotypes for exploitation in breeding programmes.

4.1.7.1 MAHALANOBIS D^2

The D^2 value between any two accessions was calculated as the sum of squares of the differences between the mean values of all the twenty one characters and used for the final grouping of the accessions.

4.1.7.1.1 Grouping of accessions into different clusters (D^2 analysis)

Mahalanobis D2 analysis provides the information on the nature of different clusters accommodating various genotypes. The entire germplasm was grouped into 10 clusters using Wards minimum variance method. The distribution of accessions into the clusters is presented in Table 10 and illustrated in Figure 4. The cluster II was the largest cluster comprising 45 accessions followed by cluster V being the next largest group with 20 accessions. Cluster I accommodated 7 accessions, cluster VI and VIII comprised of 3 genotypes each and cluster III, IV, VII, IX and X were the least consisting of 1 accession per cluster.

4.1.7.1.2 Average Inter and Intra cluster distances

The mean inter and intra cluster D and D^2 values among ten clusters were presented in Table 11 and illustrated in Figure 5. The intra cluster D^2 values ranged from 0 to 33.03. The lowest (0) intra cluster D^2 value was observed in cluster III, IV, VII, IX

and X, which are having only one accession and highest intra cluster D^2 value was observed in cluster VIII (33.03).

The inter cluster D^2 values ranged from 31.17 to 108.02. The genetic divergence (D^2) between the cluster IV and cluster X was highest (108.2), followed by cluster IV and V (107.95). While the lowest inter cluster D^2 value was recorded between the cluster I and III (31.17). The information regarding the nearest and farthest cluster from each other are presented in Table 12 indicated that cluster III (31.17) was closely related to cluster I and cluster IX (69.22) was distantly related to cluster I. Cluster VII exhibited close proximity (41.07) with cluster II and it exhibited wide diversity with cluster IV (59.87). Cluster I (31.17) was closely related to cluster III and cluster V (91.92) was distantly related with cluster III. Cluster III (36.41) showed close proximity with cluster IV while cluster X (108.02) was distantly related with cluster IV. Cluster V and cluster VIII were closely related (44.24) whereas cluster IV (107.95) showed wide diversity with cluster V. Similarly cluster VI and cluster VII (42.36) exhibited close proximity with each other while cluster IV (103) exhibited distant relationship with cluster VI. Likewise cluster IV (98.62) exhibited wide diversity with cluster VII and cluster II was closely related (41.07). Cluster V (44.24) and cluster VIII were closely related with each other whereas cluster IV (98.38) was widely related. Similarly, cluster IV (95.27) exhibited distant relationship with cluster X, while IX was close to cluster VIII(45.73). Finally the nearest cluster for cluster X was cluster IX (49.88) while the farthest cluster was cluster IV (108.02).

Based on the inter cluster distances, it was evident that all the clusters were distantly related indicating the wide genetic divergence with a scope to get good recombination through advanced breeding techniques like protoplast fusion or somatic hybridization. The highest inter cluster value between cluster IV and X (108.2) and cluster IV and V (107.95) can be expected to exert high heterotic effect in the hybrids when crossed and consequently may generate desirable segregates. Thus broad spectrum of variability can be created in the ensuing generation. This variability may be helpful in future selections and crop improvement programmes. Based on cluster mean values,

cross combinations between the clusters can be made either through advanced breeding techniques or through crossing programmes which may give good recombinants for high yield coupled with high curcumin content in turmeric. This findings are in conformity with the study reported by Rao *et al* . (2005).

4.1.7.1.3 Mean performance of the characters in clusters.

The cluster mean values for different characters are furnished in Table 13.

Genotypes of cluster VI exhibited the highest mean value for plant height (168.124), followed by cluster III (164.54) while the cluster X has accommodated short statured plants (109.44). Basal stem diameter, number of leaves and leaf length was found to be high in the cluster VI (12.63, 9.16 and 70.85 respectively).

Cluster I recorded the highest leaf width (16.94), followed by cluster VI (16.85), whereas the lowest was noticed in cluster VII (12.33). More number of tillers were recorded in cluster V (3.99) followed by cluster IV (12.33), whereas minimum number was in cluster X (1.36). The leaf area was maximum in cluster VI (861.64) followed by cluster II (832.132) and the minimum leaf area was noticed in cluster VIII (484.24). Cluster III recorded the longest leaf petiole (17.17) whereas, the shortest was observed in cluster VII (7.34).

The number of mother rhizomes and girth of mother rhizome was higher in cluster IX (3.59 and 13.48 respectively) where as the least was recorded in cluster IV (1.53) and cluster VII (4.68) respectively. Cluster IV recorded the longest mother rhizomes (9.56), where as the shortest one in the Cluster VII (4.68).

Cluster I has recorded the highest value for number, length and girth of primary rhizome (8.07, 10.15 and 9.16 respectively) where as the lowest was recorded in cluster X, cluster VII and cluster VIII (3.2, 6.62 and 5.99 respectively).

Cluster VI registered the highest number of secondary rhizomes (10.93), whereas minimum number (1.95) was recorded in cluster V.

The length of secondary rhizome was highest in cluster X (7.16), whereas, the girth was highest in cluster I (8.66), while minimum length and girth of secondary rhizome (2.09 and 3.34 respectively) were observed in cluster VIII.

The accessions in cluster VI recorded highest yield/plant (468.64) followed by cluster III (383.87) while the accessions in cluster I recorded minimum yield (276.71). Curing percentage was found to be higher in the accessions of cluster VIII (26.56), while the accessions in the cluster VII recorded low curing percentage (17.06).

The curcumin content was found to be high in the accession (T Sundar) under cluster IV (3.72) whereas it was low in the cluster IX (2.06). The accession under cluster III has taken more number of days (223.5) to maturity, followed by cluster VI (220.75), whereas the accessions under cluster I (192.625) were found to be of short duration types.

The cluster means for 21 characters revealed that cluster VI registered higher mean values for plant height, basal stem diameter, number of leaves, leaf length, leaf area, and yield per plant. Cluster IV has projected for high curcumin content.

The clustering pattern revealed that the accessions collected from same source were distributed in different clusters. Differences in genetic constitution and the influence of environmental factors may be responsible for this type of clustering. Though geographical diversity is considered as one of the criteria for selection, it may not necessarily be the only factor that determines the genetic diversity in the genotypes. Cultivars from different geographical regions were grouped in same cluster. This may be due to the free exchange of propagating material from one location to another. (Murthy & Arunachalam, 1966).

The D^2 statistics identified ten clusters of which, the accessions in cluster I were of short duration types with low curumin content and from the state of Andhra Pradesh except Imphal local.

Cluster IV was having only one accession, ie., T.Sundar which reported high curcumin content, whereas, only CLL-335 which was recorded as better performing accession was present in cluster IX.

Even though KTS, 7 CLL 335 and Duggirala are from Andhra Pradesh, they are grouped into three different clusters, which might be due to the differential genetic makeup.

The clustering pattern in cluster II (45 accessions) and cluster V (20 accessions) included genotypes from different locations pointing out that geographical diversity did not have significant impact on genetic diversity, which might be due to genetic drift and natural selection forces within a region which could have a considerable effect on genetic diversity (Murthy and Arunachalam, 1996). This may be because turmeric is inherently propagated through rhizomes and the chances for genetic variability are restricted unless and otherwise if there is any natural mutation. Though genetic makeup of each genotype was different in a cluster, the expression of these genotypes to rhizome characters and yield potential made them cluster together within.

Genotypes selected from different clusters based on genetic difference will help for further evaluation of the genetic architecture of the quantitative traits in view of the greater spectrum of variability encountered in subsequent populations.

The present results are in agreement with the findings of Sigrist *et al.* (2011) and Cintra *et al*. (2005) who revealed that the genetic divergence by dendrogram is not related to the origin of the accessions and thus the variability is not spatially structured .Similar results were reported by Pinheiro *et al.* (2003) using molecular markers where most of the genetic variation was found to be within the states of Brazil. Jan *et al.* (2012) has placed 20 genotypes of turmeric from different regions of Pakistan into two main clusters with three sub groups in 2nd cluster and the groups were primarily associated with morphological differences and secondly with horticultural use.

Vijayalatha & Cheziyan (2008) also reported the similar results stating that accessions from diverse geographical origins like PTS-2 and Roma of Orissa and VK5 and Suvarna of Kerala got clustered together with accessions from Bhavanisagar (Tamil Nadu) while Prathiba and Suvarna of Kerala were distinct from other Kerala accessions.

4.1.7.1.4 Relative Contribution of different characters towards divergence

The D^2 values have been ranked characters wise for each character indicating their contribution to total genetic divergence are furnished in Table 14 and Figure 6. Accordingly, yield per plant showed the highest contribution towards divergence by ranking first with a contribution of 62.53 percent followed by leaf area with 36.37 percent.

The characters *viz*., plant height, days to maturity and number of secondary rhizomes contributed 0.65, 0.32 and 0.11 percent respectively towards divergence, while basal diameter, number of leaves, leaf length, leaf width, number of tillers, length of leaf petiole, number of mother rhizomes, length of mother rhizomes, girth of mother rhizomes, number of primary rhizomes, length of primary rhizomes, girth of primary rhizomes, length of secondary rhizomes, girth of secondary rhizomes, curing percentage and curcumin content did not contribute to the diversity.

Among the characters that contributed towards genetic divergence, yield recorded highest rank emphasizing the importance of that character towards divergence. This is in corroboration with the findings of Vijayalatha and Cheziyan (2008) and Verma *et al* .(2015).

The next contributing character was leaf area followed by plant height as increase in leaf area and plant height increases the accumulation and assimilation of carbohydrates there by increases the rhizome yield. Similar results for plant height was also reported by Verma *et al.* (2015) and Jan *et al*. (2012).On the other hand Sigrist *et al*. (2011), Cintra *et al*. (2005) and Lynrah *et al.* (1998) suggested that curumin content and number of tillers were most important contributing characters towards genetic divergence.

Quantitative characterization of phenotypic traits has been used in various germplasm descriptions. Such studies are important because they provide information on germplam classification and facilitate the utilization of genetic resources among the genotypes and the indentified traits could be used for germplasm grouping.

The morphological descriptors reflect not only the genetic constitution of the cultivars but also the interaction of the genotypes with the environment (GXE), within which it is expressed (Lin and Bins 1984)

The clustering technique has clearly defined that geographical diversity need not necessarily be related to genetic diversity. The cluster distances that separated the genotypes attributed for the reason that the genotypes were distinct of their genetic background among the clusters and more likely within the cluster.

Hence studies on genetic divergence is an important prerequisite before embarking on genetic improvement of any crop. Since turmeric is vegetatively propagated crop, it offers very little (or) no scope from divergent clusters ensuring genetic variability when subjected to artificial induction to prove for better results (Singh *et al.* 2001)

4.1.7.2 PRINCIPAL COMPONENT ANALYSIS (PCA)

Principal Component Analysis (PCA) is a multivariate statistical tool, which attempts to describe total variation in a sample with fewer variables than in the original data set. It is done by successively selecting linear combination on the principal component of the attributes which retain the highest proportion of unexplained variability between the units and attributes. It differs from the regression analysis that PCA chooses a line with minimum sum of squared perpendicular distances to the data sets.

It was applied to identify patterns in the data. Set containing many correlated variable, into small sets of components of the original variables and axes are accounted for more variation in the original data than any simple variation alone.

The character loading values for Principal Components represented the weights defining the contribution of different characters for the respective principal components. Further, the loadings signs (+ / -) are indicative of the direction of contribution, similar to that of regression coefficients. The mean values of twenty one characters were subjected to PCA.

4.1.7.2 .1 COMPONENT LOADING STUDIES

The PCA of the correlation matrix represented eight components. The PCA showing eigen values less than one was considered as non significant. The component loadings and the per cent variability for each PC are furnished in Tables 15,16 and Figures 7 and 8.

In the present investigation, the first principal components with eigen value >1 has contributed to 71.819 per cent of cumulative variability, among the 83 accessions of turmeric evaluated for 21 characters.

The significance of PC in each variable was determined by comparing the loadings. The PC 1 accounted for highest variability of 26.714 percent and was an index of leaf area (0.366) basal diameter (0.339), plant height (0.326) leaf length (0.315), leaf width (0.296), number of leaves (0.293), girth of primary rhizome (0.266), number of primary rhizomes (0.246), girth of mother rhizome (0.204), girth of secondary rhizome (0.196), length of secondary rhizome (0.178), length of mother rhizome (0.168), number of secondary rhizomes (0.151), length of primary rhizome (0.136), yield per plant (0.107) days to maturity (0.076), curcumin content (0.064) and length of leaf petiole (0.044) in decreasing order of element and explained the variability. Number of mother rhizomes (-0.168), curing percentage (-0.080) and number of tillers (-0.037) contributed negatively towards the genetic divergence.

The second principal component recorded 12.852 per cent of total variability and together with PC I contributed about 39.566 per cent of variance and showed high

loadings for curcumin content (0.369), followed by number of secondary rhizomes (0.329),days to maturity (0.298), yield per plant (0.276), plant height (0.174) number of leaves (0.150), number of tillers (0.123) basal diameter (0.058), length of mother rhizome (0.053) and leaf length (0.026) in decreasing order of the element and explained about variability in this principal component, while length of primary rhizome (-0.411), girth of mother rhizome (-0.266), length of leaf petiole (-0.242), girth of secondary rhizome (0.234), length of secondary rhizome (-0.218) number of mother rhizomes (-0.200), leaf width (-0.178) curing percentage (-0.177), girth of primary rhizome (-0.081), leaf area (-0.072) and number of primary rhizomes (-0.001) contributed negatively towards genetic divergence.

The third principal component PC 3 accorded 7.770 variance and together with PC1 and PC 2 contributed about 47.335 percent of variance and showed high loadings for number of primary rhizomes (0.335) and also showed positive correlation for length of mother rhizome (0.322), length of leaf petiole (0.287), girth of primary rhizome (0.282), length of primary rhizome (0.263), curcumin content (0.252), number of mother rhizomes (0.207), days to maturity (0.206) girth of mother rhizome (0.182), yield per plant (0.111), number of tillers (0.082) and plant height (0.047) in decreasing order of element and explained about the variability in this vector. On contrary, curing percentage (-0.367), leaf area (-0.253), leaf length (-0.229), number of leaves (-0.210), leaf width (-0.177), basal stem diameter (-0.177), number of secondary rhizomes (-0.069), length of secondary rhizome (-0.058) and girth of secondary rhizome (-0.044) contributed negatively towards genetic divergence.

The fourth principal component, PC4 contributed 7.025 percent variability and showed high loadings for length of leaf petiole (0.429) followed by number of tillers (0.356), curing percentage (0.252), leaf width (0.199), number of leaves (0.171), length of primary rhizome (0.152), curcumin content (0.128), yield per plant (0.120), plant height (0.108) basal diameter (0.105) and leaf area (0.066) contributed in decreasing

order for genetic divergence. Length of secondary rhizome (-0.353), girth of mother rhizome (-0.327) girth of secondary rhizome (-0.249), days to maturity (-0.242), number of secondary rhizomes (-0.226) length of mother rhizome (-0.148), girth of primary rhizome (-0.056), leaf length (-0.054) and number of mother rhizomes (-0.030) showed negative correlation towards genetic divergence.

The fifth principal component, PC 5, relatively attributed a total variance of 6.54 with a high load for girth of secondary rhizome (0.478), curcumin content (0.278), number of leaves (0.191), girth of primary rhizome (0.135), length of secondary rhizome (0.131), length of leaf petiole (0.120), length of primary rhizome (0.036) and plant height (0.020) being positive and the negative correlation was noted for curing percentage (-0.320), length of mother rhizome (-0.499), number of mother rhizomes (-0.309), girth of mother rhizome (-0.229) leaf length (-0.169), yield per plant (-0.153), number of secondary rhizomes (-0.135) leaf area (-0.139) basal stem diameter (-0.067), leaf width (0.071), number of tillers (-0.076), number of primary rhizomes (-0.025) and days taken to maturity (-0.037).

The PC 6 accorded a total variance of 5.621 and characterized by number of tillers (0.664), length of secondary rhizome (0.241), girth of mother rhizome (0.218), length of mother rhizome (0.176), curcumin content (0.174), girth of secondary rhizome (0.135), plant height (0.093), number of primary rhizomes (0.088), number of leaves (0.068), leaf length (0.051) and leaf area (0.018) being positive and negative correlation was noted for days to maturity (-0.299), length of leaf petiole (-0.293), number of secondary rhizomes (-0.229), basal stem diameter (-0.225), length of primary rhizome (-0.179), girth of primary rhizome (-0.136), yield per plant (-0131), curing percentage (-0.059), leaf width (0.032) and number of mother rhizomes (-0.011).

The seventh principal component, PC 7, accorded a variance of 5.294 and together with all the other six PCAs contributed about 71.819 of variance, showing high positive loadings for leaf length (0.359) followed by number of mother rhizomes (0.298), plant

height (0.296), days to a maturity (0.271), leaf area (0.173), girth of primary rhizome (0.128), number of tillers (0.088) and girth of secondary rhizome (0.041) in descending order, while negative correlation was recorded highest for yield per plant (-0.464), followed by length of secondary rhizome (-0.421), number of secondary rhizomes (-0.244), curing percentage (-0.210), number of primary rhizomes (-0.197), girth of mother rhizome (-0.085), leaf width (-0.084), length of mother rhizome (-0.073), curcumin content (-0.074), number of leaves (-0.043), basal diameter (-0.033), length of leaf petiole (-0.033) and length of primary rhizome (-0.032).

Among all the principal components, PC 1 projected to score high loadings for plant height, basal stem diameter, number of leaves, leaf width and leaf area while, PC 2 accorded much of its variance through number of secondary rhizomes, yield per plant, curcumin content and days to maturity. PC 3 attributed its variance through length of mother rhizome, number of primary rhizomes, length of primary rhizome and girth of primary rhizome. PC 4 relatively contributed high variance for length of leaf petiole and curing percentage, while for girth of secondary rhizome, PC 5 exhibited highest percent of variance. PC 6 attributed its variance through number of tillers, girth of mother rhizome and length of secondary rhizome whereas PC 7 projected to score high variance for leaf length and number of mother rhizomes (Table 15).

In the current study, the components were dominated by leaf area, curcumin content, and number of primary and secondary rhizomes attributing that these characters were major contributors to total variance with a share towards genetic divergence. The genotypes that exhibited higher mean values for those characters should form the basis for selection.

4.2 DIVERSITY STUDIES IN TURMERIC ACCESSIONS USING MOLECULAR MARKERS

Indexing the variability of germplasm accessions and breeding stocks has become increasingly important for both genetic research and breeding. Such information can be obtained using an array of molecular techniques, even though they differ technically, but possess fewer relative advantages over one another. The information obtained using molecular markers like RAPD and SSR markers offer many benefits for identifying variation and establishing diversity among the accessions. Molecular markers enable the assessment of genetic similarity between accessions in the early stages of development and are advantageous for diversity studies (Nicolosi *et al.* 2000). In the present study, apart from using the morphological traits, RAPD and SSR markers were also used to estimate the extent of molecular diversity among turmeric accessions. The diversity among 83 accessions based on the results of RAPD and SSR data are discussed here under.

All the 83 turmeric accessions were studied for their genetic diversity at molecular level using 21 Random Amplified Polymorphic DNA (RAPD) markers and 19 Simple Sequence Repeat (SSR) markers.

4.2.1 DNA YIELD AND PURITY ASSESSMENT

The DNA was isolated from young healthy leaves of 83 accessions by using the modified CTAB protocol developed by Syamkumar *et al*. 2003. This protocol resulted in white translucent DNA pellets that easily solubilized in Tris EDTA (TE) buffer. The DNA resulted by this method was homogeneous and not degraded. The quality of DNA was verified by spectrophotometer and also by agarose gel electrophoresis. The results indicated that the isolated DNA was of good quality and had high molecular weight (Plate 3).

The genomic DNA was extracted from all the 83 accessions and the DNA yields ranged from 270 to 3270 ηg/µl (Table 17). The purity of DNA was determined by A_{260}:A_{280} ratio which varied from 1.73 to 1.99 indicating that the DNA was relatively of high purity and was suitable for PCR amplification.

The DNA yield (ηg/µl) and the OD ratio (260/280) for all the accessions collected are given in Table 17. The highest content of DNA i.e.3270 ηg/µl was obtained with the accession IC -212605 and Badipadar whereas the lowest amount of DNA (270ηg/µl) was realized with the accession IC-394396.

Higher efficiency of molecular markers over morphological characterization was well demonstrated. The DNA markers used in the present study were easy to use; however, SSR markers are specific and reliable, though there is difficulty in technology, making them amenable for use in any breeding programme. This kind of superior performance of molecular markers over morphological markers was reported in turmeric by Jan *et al.* (2011), Vijayazlatha and Chezhiyan (2008), Sigrist *et al.*(2010) and Siju *et al.*(2010 & 2013).

4.2.2 RAPD ANALYSIS

RAPD analysis has been found to be useful in differentiating closely related species. Their advantage principally lies in detecting simultaneously many randomly distributed loci within the genome, in a simple, cost-effective manner, requiring no previous knowledge of genome sequence.

A total of twenty one RAPD primers were used to amplify the gene loci in the genomes of different turmeric accessions. From the electrophoregrams of the PCR amplicons, the random primers produced the amplification products with sizes ranging from 190 bp to 1500 bp. The amplification and banding pattern of RAPD markers in all the 83 accessions of turmeric were illustrated in plates 4, 5, 6 and 7.

4.2.2.1 Polymorphic information content and Heterozygosity

The polymorphic information content and the heterozygosity were calculated and presented in the Table 18. All the primers showed the PIC value more than 0.5 except the RAPD primer OPC 16 (0.483). The PIC is considered as the character of interest in the selection of informative markers as it reflects the number of detected alleles and the relative distribution of their frequency. Low PIC value (0.483) was observed with the primer OPC 16 which indicates that the marker is unable to discriminate between genotypes and a PIC of >0.5 is indication of a good marker (Elvira *et al.*2015.) Heterozygosity of the primers ranged from 0.576 (OPC 16) to 0.867 (OPA 10).

The number of scored bands per primer ranged from 3 to 9 with a mean value of 4.5 per primer (Table 19). A total of 103 amplified fragments were scored across 83 accessions of turmeric with 21 RAPD primers and were used to estimate genetic relationships among themselves. Amplification of large number of polymorphic bands indicated that the primer sets used in this study could be of significance for the assessment of genetic diversity in turmeric cultivars. Similar results were reported by several authors, *viz.,* Jan *et al.* (2011) reported 92 polymorphic fragments by using 10 RAPD markers, while Thaikert and Yinyong (2009) reported 184 scorable bands of which, 166 were polymorphic by using 19 RAPD primers in turmeric. Similarly Singh *et al*. (2012) could observe 99 amplification fragments using 11 RAPD primers.

The molecular weights of the PCR amplified bands ranged from 190 bp to 1500 bp and the percentage of polymorphism ranged from 75% to 100%. All the random primers scored 100% polymorphism except OPA 4 (75%) and OPA 8 (85.71%). Several workers ((El vira *et al* .(2015) Singh *et al.* (2012) and Jan *et al.* (2011)) have reported different ranges of PCR amplified bands ranging from 130 bp-3000bp.

However, some doubts have been expressed regarding the reproducibility of the RAPD technique and the suitability of the RAPD markers for genetic diversity studies (Rafalski *et al.*1991, Wilkie *et al.*1993 and Hallden *et al.* 1996). However, the reproducibility of RAPDs can be achieved by using optimized PCR conditions and by scoring only reproducible bands as opined by Aliyev *et al.* 2007.

4.2.2.2 Similarity index

Banding profiles obtained with 21 primers for 83 accessions of turmeric were analyzed on the basis of presence or absence of the band. Jaccard's similarity coefficients among these accessions were calculated to establish the genetic relationships and are presented in Table 20. Genetic similarity based on Jaccard's coefficient revealed considerable level of diversity among the accessions under the study. The average genetic similarity among eighty three turmeric accessions ranged from a coefficient of 0.366 to 0.98. Genotypes with low similarity values are more divergent. Among all the accessions CLS 269 was found to show maximum similarity with Penekanametta with a similarity coefficient of 0.98 where as Rajapuri has shown least similarity with (0.3662) with CLL 335. However CLL 335 was found to be distant from all the other 82 accessions.

4.2.2.3 Cluster analysis

The genetic relationships of turmeric accessions were further evaluated by UPGMA cluster analysis, on RAPD data set, using a minimum variance algorithm (Fig 9 and Table 21). In cluster analysis 83 accessions were grouped into three main clusters at 45% similarity level.

Cluster I comprised of 41 genotypes which was further subdivided into 4 sub-clusters. Sub-cluster IA had 26 genotypes while sub-cluster IB had 7 genotypes. Sub-cluster I C had 3 genotypes and sub-cluster ID formed with 5 genotypes.

Similarly, cluster II was having 41 accessions and was further subdivided into 8 sub-groups. Sub-group IIA consists of 21 accessions, Subgroup IIB was formed with 11 accessions .Whereas sub-groups II C and II E were monogenic with only one accession each (IC-353560 and Vontimitta respectively). In subgroup II D two accessions i.e., IC-540386 and IC -319621 were included while sub- group II F was having three accessions viz., CC- 9401,KTS-7 and Aleppy supreme whereas sub- groups II G and II H were accommodated with one accession each ie., Rajapuri and CL 9 respectively.

Third cluster was found to be very distinct with only one accession, CLL 335.

4.2.2.4 Principal component analysis for RAPD analysis

Principal Component Analysis was done based on the Jaccard's similarity matrix obtained and presented in table 22. The first component accounts for 33.13% of variation and second component accounted for 12.93% of variation. In this manner, totally 44 principal components accounted for 99.89 % of variations. The principal component analysis was based on molecular data of 21 RAPD markers to visualize the genetic relatedness among the turmeric accessions in detail. The description of the data using three dimensional pictorial graph and the same is presented in Fig 10. It was evident that the turmeric accessions were dispersed on the PC plot, which is a reflection of variation among the accessions. The results of PCA showed a clear cut separation. However, as depicted in figure, some of the accessions appear to be overlapping with each other depicting high similarity in these accessions.

It was clear from the analysis that the results obtained from the dendrogram were repeated in the PCA, which strengthened the ability and accuracy of the RAPD analysis applied to turmeric accessions in the present study.

From the foregoing discussion, it can be concluded that RAPD markers could be employed for finger printing and characterization of genotypes, assessment of molecular genetic divergence and relatedness among turmeric genotypes. This information can be used successfully for cultivar identification and for assessing the genetic diversity among turmeric accessions. They are effective, precise and more efficient than morphological markers in identifying closely related species.

4.2.3 SSR ANALYSIS

The accessions were also studied for their genetic diversity at molecular level using Simple Sequence Repeat (SSR) markers. Nineteen selected primers synthesized by Siju *et al.* (2010 and 2013) were used to analyze the inter-accession diversity in 83 accessions of turmeric. All the primers were initially evaluated for their annealing temperature using a gradient PCR technique.

The details of SSR primers used for assessing the molecular diversity among 83 accessions of turmeric are furnished in the Annexure III B. Among the nineteen SSR primers used in the analysis, seventeen primers produced the polymorphic bands. All the SSR primers used in the present study produced discrete, scorable and unambiguous bands. The PCR product size obtained by the amplification of SSR primers ranged from 110bp to 440bp with a total of 38 fragments. The amplification and banding pattern of these SSR markers in all the 83 accessions of turmeric were illustrated in Plates 8 to 19.

4.2.3.1 Allele diversity of SSR marker analysis

All the SSR markers used in the genetic analysis were polymorphic with banding pattern ranging from 1 to a maximum of 3 alleles per individual in all the loci (Table 23), which supports the triploid status of *Curcuma longa.* The results are in conformity with the studies conducted by Islam, 2004, Sasikumar, 2005, Ravindran *et al.*2007, which was also verified by Siju *et al.* (2010) and Sigirist *et al*. (2010).Taheri *et al*.(2014) reported a range of 3-7 alleles with an average of 4.5 per locus in irradiated turmeric plants.

The allele frequency, polymorphic information (PIC) content and percent polymorphism were presented in Table 24. The PIC value provides an estimate of discriminatory power of the marker by taking into account not only the number of alleles at a locus but also the relative frequencies of these alleles. The highest PIC was scored by the SSR marker CuMiSat-03 (0.628), while it was found to be lowest with the SSR marker CuMiSat-36 (0.212). The highest allele frequency was recorded by the marker,

CuMiSat-22 (0.506) where as the lowest was by CuMiSat -11 (0.333). All the markers have shown 100% polymorphism with 83 accessions of turmeric except CuMiSat-22, CuMiSat-26 and CuMiSat -27 which have recorded 50% polymorphism. However the SSR markers CuMiSat-16 and CuMiSat-17 were found to be monomorphic.

4.2.3.2 Similarity index

The banding pattern of SSR markers scored in the form of binary data was used for computing Jaccard's similarity index values obtained for each pair wise comparison among the 83 accessions and clustering was done by the UPGMA method using SAHN programme of NTSYS-pc version 2.1 which is presented in the Table 25. The similarity coefficient based on 19 SSR markers ranged from 0.267 to 1. Among the 83 accessions studied, the accessions BSR 2 and IC 332957 were found to be distant with lowest similarity index (0.267) whereas nine sets of accessions, (Wagon, Penekenametta), (Vontimitta with Ochira, Salem, Dindigam and Duggirala), (IC- 319621 with IC-211402, IC-181919), (IC-420474, IC-319341 and IC-394396), (CL-2 and Tekuripeta,) (KTS-3 with IC- 394903 and IC-319621), (Rajapuri with IC-420606), (KTS -8 with CL-17) and (KTS-7 and GL puram), had a similarity coefficient of 1, indicating identical finger prints probably due to duplication of the accessions. Identical microsatellite profiles in the studied microsatellite loci, suggested that the observed morphological differences between the cultivars may be associated with somatic mutations, which were not detectable with the used SSR markers. Hence, analysis of additional loci is necessary to identify and discriminate further the investigated accessions that differ in their phenotypic characteristics.

The accessions CLL 335 and Chintapalli local 1 were found to record low similarity coefficient values with all the other accessions. The similarity index between these two accessions was also found to be very low (0.2857).

4.2.3.3 Cluster Analysis

The similarity values obtained for each pair wise comparison of SSR markers in all the 83 accessions of turmeric were used to construct dendrogram based on hierarchical clustering and the results are presented in Fig. 11 and Table 26. Clustering of 83 turmeric accessions using the dominant scoring (presence /absence) of bands based on UPGMA separated them into two main clusters at 44% similarity level. Cluster I is again separated into three subgroups. Sub-cluster I A comprised of 11 accessions, IB with 13 accession and IC with 6 accessions. Cluster II formed the major group within the dendrogram by including 52 accessions which was further subdivided into 5 subgroups. Sub-cluster IIA was having 5 accessions while IIB has contained 40 accessions. Sub-clusters IIC and IID were comprising of 4 and 2 accessions each respectively whereas sub-group IIE was monogenic with only one accession (IC-0330113).

The accession Chintapalli local 1 was found to be distinct with low similarity coefficient values with all the other accessions and was accommodated in cluster III. Sub cluster IB contained all the accessions which were collected from NBPGR regional station, Meghalaya, whereas, sub cluster I C contained all the accessions in which most of them were local varieties from hilly areas. However, some of the accessions having the same geographic origin were distributed in different sub clusters, suggesting their genetic diversity. Selections from same geographic region were grouped separately in different sub clusters. The existence of duplicates may be due to multiple donations of the same seed source but from different collections. Therefore, diverse geographic origins among accessions may not always be a reliable indicator for the sampling of genetically diverse materials. From the conservation point of view, the identification and elimination of duplicates in a collection can save time and resources both financial and human in germplasm maintenance, due to reduced number of accessions. Further, it could also be concluded that they may also be different accessions derived from somatic mutations that were not detected by the molecular markers used in this study.

The present study of use of microsatellite markers in characterization of turmeric accessions revealed that 19 polymorphic microsatellite markers were found efficient to distinguish important accessions and will certainly be useful for purposes such as the certification of varieties, and for identification of pest and disease resistant lines and high yielding varieties.

The SSR technique is expensive compared to RAPD marker technique; hence a few primers which were developed by Siju *et al.* (2010 and 2013) were used in this preliminary study. Further a finer molecular analysis of turmeric accessions is required with more number of SSR markers in order to detect and identify unique as well as fine resolution of molecular polymorphism between different identical genotypes. The use of a larger number of SSRs with greater genome coverage could help to reveal genetic diversity more accurately and also help to unambiguously differentiate those accessions with identical allelic patterns as revealed by the set of primers used in this study. Further, by increasing the number of accessions from the different regions along with the use of a higher number of polymorphic SSR markers, a better assessment of genetic diversity could be carried out**.**

4.2. 3.4 Principal component analysis for SSR analysis

Principal Component Analysis was done based on the Jaccard's similarity matrix obtained by molecular data of 19 SSR markers to visualize the genetic relatedness among the turmeric accessions in detail (Table 27). The first component extracted 38.7551% of variation and second component accounted for 16.8564% of principal changes. In this manner, totally 17 principal components accounted for a cumulative variation of 99.879% among the 83 accessions. The description of the data was done using three dimensional pictorial graphs and is represented in Fig.12. From the graph, it was evident that the turmeric accessions were dispersed on the PC plot, which is a reflection of its genetic base. The results of PCA showed a clear cut separation. However, as depicted in

figure some of the accessions appear to be overlapping with each other indicating high similarity in these accessions. It was clear from the analysis that the results obtained from PCA were in agreement with the dendrogram generated by UPGMA cluster analysis. This strengthened the ability and accuracy of the SSR analysis applied to turmeric accessions in the present study.

4.2.4 Comparison of clustering pattern based on morphological diversity and molecular diversity

The comparison of morphological and molecular clustering pattern will give information on the amount of variability found between the accessions and the best method of assessing the diversity.

In this study, the morphological characterization of accessions was mainly grouped according to growth and rhizome characters which are complex and polygenic. Such characters are environmentally affected and therefore liable to subjective evaluation. In this sense, the molecular characterization is more efficient in the generation of an unbiased picture of diversity than a morphological approach. However, the morphological characterization is still important in germplasm management, as it is the predominant method of characterization, because the diversity due to mutation is not identified by means of biochemical and molecular techniques and determination of molecular diversity should not be seen as replacement for traditional characterization but rather as a complement to it.

Although the correlation between the morphological and molecular data was low, both methods allowed the grouping of turmeric accessions. Despite the fact that morphological traits were relatively less efficient for precise discrimination of closely related accessions, the cost and time invested were lower than the molecular analysis. Therefore, based on cost, efficiency and information gained, both techniques appeared complementary to study the characterization and diversity among turmeric accessions.

Genetic distances between 83 turmeric accessions were estimated using both morphological and molecular markers (RAPD and SSRs). These methods were able to distinguish all the accessions used in the study. Both morphological and molecular characterization methods were found efficient in grouping the accessions. However, molecular analysis grouped the accession more accurately. The pattern of sub clustering of turmeric accessions in a cluster varied over different marker systems. But SSRs were able to differentiate effectively. However most of the accessions were grouped together by SSRs and RAPD markers whereas, morphological markers could not do the same because as morphological markers obviously can detect cultivar variation only at phenotypic level.

Eight accessions, viz., Wagon, Penekenametta, CLS-269, KTS-3, CLL-335, Gorakhpur 361, Ethamukkala and Thodupuzha were grouped together in both RAPD and SSR analysis, indicating their common ancestral origin whereas, they were grouped separately by morphological characterization. The KTS-8 from Andhra Pradesh and CL-17 from Tamil Nadu were grouped together in the same cluster in all the three ie, morphological (Cluster II), RAPD (II A) and SSR (II B) indicating that both may be duplicates and genetically similar and it also shows that these accessions may be the selections, made from the same source. Similarly, Tekuripeta from Andhra Pradesh and CL-2 from Tamil Nadu may also be duplicates which were existed in the same cluster, with similarity coefficient value of 1.0 with SSR analysis, which generated identical fingerprints across the markers.

Accessions from same region belonged to different clusters and accessions that were collected from different regions were found in same clusters which can be attributed to the absence of influence of location specificity. Among the collected genotypes chance of migration of rhizome seed material by growers from one region to another may also be the reason for the observed clustering of different turmeric accessions.

Local commercial cultivar, Duggirala which was grouped separately in morphological clustering (Cluster X) but it was clustered together with other accessions

and found to have high similarity with Ochira , Salem and Dindigam in RAPD (sub-cluster IIA) and in SSR (sub-cluster IIB). Similarly, Chintapalli local-1 a local cultivar from high altitude zone from Andhra Pradesh was clustered together with Imphal local, which is a local collection from Assam in morphological clustering (cluster I), whereas, they fell into different clusters in RAPD (IB &II A) and SSR (Cluster III and IIB) indicates that phenotypically similar accessions are not always genotypically similar. The identification of turmeric lines based on morphological characterization in large number of germplasm lines is difficult as it is the outcome of genotype and environmental interactions.

The accession, CLL-335 was positioned in a separate group in morphological (Cluster IX) and RAPD (Cluster III) characterization whereas, in SSR it was grouped together in Sub-cluster IA. Similarly, Chintapalli local- 1 was grouped together with other varieties both in RAPD (IB) and morphological clustering, whereas it was found to be unique as it was grouped in a separate cluster in SSR analysis. This variation observed between RAPD and SSR may be due to the fact that the PCR amplified products in the two marker assays originated from different repetitive and non-repetitive regions of the genomes and the possibility that many co-migrating bands may be non-homologous producing a background disturbance that could influence the result.

Several studies on assessment of genetic diversity of plants using molecular markers have established the correlation between geographical distance and genetic similarity between individuals (Islam, 2004, and Singh *et al* .2012).

Studies on screening germplasm collections using phenotypic traits have revealed large genetic divergence among cultivars from South East Asia, which should be an important source of genetic variation for turmeric improvement worldwide (Singh *et al.* 2003, Sasikumar 2005, Choudary *et al*. 2006, and Sigrist, *et al.* 2011).

RAPD analysis could identify the closely related species in which, the similarity coefficients of 0.8 to 0.9 were observed for some accessions. The use of SSR markers has been demonstrated to be a powerful tool to define how turmeric diversity is structured

leading to a better management of the germplasm bank and also to promote the use of these accessions in further breeding programmes.

However, accurate knowledge of genetic diversity among turmeric accession is important for establishing core collections of germplasm in aiding breeding work (Siju *et al.*2010). On contrary, Jan *et al.* 2011 reported that most of the turmeric populations from different regions of Pakistan were having similarity matrix, ranging from 89% to 100% and hence opined that RAPD markers evaluated in their study were not highly efficient for finger printing of turmeric genotypes as they could not differentiate the turmeric genotypes relevant to their populations. Similarly, Thairkert and Yingyong (2009) also expressed that the cluster analysis of genetic diversity of curcuma species were mixed together and placed into several groups and the results from cluster analysis using RAPD markers did not show any distinct relationship with their region.

The SSR markers have been reported to be more responsible and produce more complex marker pattern than RAPD approach. Singh *et al.* (2012) also opined that RAPD markers were less informative than ISSR markers in studying the cultivar diversity in turmeric.

In the present study, synonymous entities with genetic similarity coefficient value of 1.0 were identified by the SSR markers which is utmost important for precise assessment of cultivar type and genetic diversity in turmeric. Among the 83 accessions studied, 9 sets of synonymous entities were identified. *viz.*, (Wagon and Penekenametta), (Vontimitta with Ochira, Salem, Dindigam and Duggirala), (IC- 319621 and IC-211402, IC 181919), (IC-420474, IC-319341 and IC-394396), (CL-2 and Tekuripeta), (KTS-3, IC- 394903 and IC-319621), (Rajapuri and IC-420606), (KTS -8 and CL-17) (KTS-7 and GL puram). Our results are supported by the studies conducted by various authors revealing that existence of synonymous turmeric accessions from India and Brazil using genomic SSR markers as described recently by Siju *et al.* (2010 and Sigrist *et al.* 2010 respectively. Among all the accessions CLL- 335 and Chintapalli local-1 were found to have wide diversity (Plate 20) with all the other

accessions and further studies are needed to identify any desirable traits which will be useful for the breeders for future breeding programme .

Rapid germplasm movement across the length and breadth of the country by the settlers would have resulted in the spread of the cultivars to different parts of India. It is quite possible that the same genetic material would have been spread to different regions in the olden days and got acclimatized to the place under different vernacular names in course of time. Clonal propagation would have resulted in preserving the genetic fidelity of the original material. Recollection of the same entities based on the vernacular identity would have added to the buildup of duplicates in the genebank. The SSR analysis confirms the fact that collecting turmeric accessions based on vernacular identity could result in adding duplicates into the germplasm collection.

Identification of markers associated with specific traits of great interest, which could be converted into SCAR markers needs attention to speed up the crop improvement programme. The degree of genetic variability evaluated through molecular characterization can form the basis for future breeding programme and a finer molecular analysis with additional SSR loci and use of other molecular markers like AFLP and ISSRs is also required to differentiate point mutations involved in identical accessions and to delineate some geographical associations among the accessions.

Our results signify the presence of great genetic variability among elite genotypes of turmeric. Both RAPD and SSR markers are useful in the assessment of turmeric diversity, detection of duplicates in germplasm and selection of a core collection to enhance the efficiency of genotype management for use in turmeric breeding and conservation. Hence, turmeric being a vegetatively propagated and highly heterozygous crop, hybridization between some divergent genotypes could broaden the genetic base for the development of elite genotypes with required qualitative and quantitative characters.

Table: 1 Particulars of turmeric germplasm studied.

S.no	Accession	S.no	Accession
1	Aleppy supreme	41	IC-319621
2	Amritpani	42	IC-330113
3	Avidi	43	IC-332957
4	Badipadar	44	IC-353560
5	Bataguda	45	IC-394268
6	BSR-2	46	IC-394396
7	Chintapalli local -1	47	IC-394903
8	Chintapalli local-2	48	IC-416941
9	CC 94-01	49	IC-420474
10	CL-2	50	IC-420556
11	CL-7	51	IC-420606
12	CL-9	52	IC-521333
13	CL -12	53	IC-540383
14	CL-16	54	IC-540386
15	CL-17	55	IC-540387
16	CLI- 317	56	IC-545139
17	CLL-335	57	Imphal local
18	Cuddapah local	58	KTS-6
19	Dindigam	59	KTS-7
20	Duggirala	60	KTS-8
21	Ernad Chand	61	Lacadong
22	Ethamukkala	62	Meghaturmeric
23	Flourescent	63	Movatupuzha
24	GL puram	64	Mydukur
25	Gorakhpur-361	65	Ochira
26	GS	66	Paderu local
27	IC-033007	67	Penakanametta
28	IC-181919	68	Rajapuri
29	IC-211360	69	Rajendra sonia
30	IC-211381	70	Salem
31	IC-211401	71	Sugandham
32	IC-211402	72	TCP 70
33	IC-211641	73	Tekuripeta
34	IC-211642	74	Tenali
35	IC-212267	75	Thodupuzha
36	IC-212578	76	T Sundar
37	IC-212585	77	Vikici
38	IC-212605	78	Vontimitta
39	IC-212606	79	Wagon
40	IC-319341	80	Wynad local
Checks			
1. CLS-269			
2. KTS-3			
3. IISR Prathibha			

Table :2 Analysis of variance for quantitative traits in turmeric

		Plant Height(cm)	Basal Diameter (cm)	No of Leaves	Leaf Length(cm)	Leaf Width(cm)
Block (ignoring Treatments)	7.00	2776.35***	7.22***	0.73***	495.43***	3.66***
Treatment (eliminating Blocks)	82.00	607.55***	2.58***	0.96***	116.49***	4.09***
Checks	2.00	145.52*	7.71***	4.01***	110.68***	7.14***
Checks+Var vs. Var.	80.00	619.10***	2.45***	0.88***	116.64***	4.02***
ERROR	14.00	25.81	0.20	0.08	8.20	0.08
Block (eliminating Check+Var.)	7.00	12.75	0.09	0.35**	46.88**	0.38**
Entries (ignoring Blocks)	82.00	843.47***	3.18***	0.99***	154.78***	4.37***
Checks	2.00	145.52*	7.71***	4.01***	110.68***	7.14***
Varieties	79.00	670.85***	2.67***	0.76***	129.34***	4.09***
Checks vs. Varieties	1.00	15876.56***	34.52***	13.29	2252.67***	21.52***
ERROR	14.00	25.81	0.20	0.08	8.20	0.08
Ci – Cj	1.00	5.45	0.47	0.31	3.07	0.31
BiVi – BiVj	1.00	15.41	1.34	0.87	8.69	0.87
BiVi – BjVj	1.00	17.79	1.55	1.00	10.03	1.00
Ci – VI	1.00	13.34	1.16	0.75	7.52	0.75

Contd…

(Table 2 cont…)

	No of Tillers	Leaf Area(cm^2)	Length of Leaf Petiole(cm)	No of Mother Rhizomes	Length of Mother Rhizome (cm)	Girth of Mother Rhizome (cm)
Block (ignoring Treatments)	0.25***	69219.42***	79.97***	0.29	6.35*	53.63***
Treatment (eliminating Blocks)	0.54***	33338.00***	10.31**	0.48*	1.68	3.76*
Checks	3.45***	67727.71***	0.49	0.07	1.10	2.12
Checks+Var vs. Var.	0.47***	32478.26***	10.55**	0.49*	1.70	3.80*
ERROR	0.03	1778.88	3.07	0.18	2.11	1.60
Block (eliminating Check+Var.)	0.05	5710.27*	6.85	0.36	8.34*	20.33***
Entries (ignoring Blocks)	0.56***	38759.52***	16.55***	0.47*	1.51	6.60**
Checks	3.45***	67727.71***	0.49	0.07	1.10	2.12
Varieties	0.45***	31672.18***	17.16***	0.46	1.53	6.73**
Checks vs. Varieties	3.18***	540723.13***	0.45	2.10	0.84	4.76
ERROR	0.03	1778.88	3.07	0.18	2.11	1.60
Ci - Cj	0.18	45.23	1.88	0.45	1.56	1.36
BiVi - BiVj	0.50	127.93	5.32	1.28	4.40	3.84
BiVi - BjVj	0.58	147.72	6.14	1.47	5.08	4.43
Ci - VI	0.44	110.79	4.60	1.11	3.81	3.33

contd...

(Table 2 cont…)

	No of Primary Rhizomes	Length of Primary Rhizome	Girth of Primary Rhizomes (cm)	No of secondary rhizomes	Length of Secondary Rhizomes (cm)	Girth of Secondary Rhizomes (cm)	Yield /Plant (g)

		(cm)					
Block (ignoring Treatments)	16.17**	6.60***	4.76	84.23***	0.81	6.91	77948.03***
Treatment (eliminating Blocks)	5.60*	1.53*	1.66	8.11***	2.79**	2.25	12258.55***
Checks	5.72	1.93*	0.05	5.00	1.79	2.26	27762.50**
Checks+Var vs. Var.	5.60*	1.52*	1.70	8.19***	2.81**	2.25	11870.95***
ERROR	2.43	0.50	1.77	1.51	0.71	2.75	2435.12
Block (eliminating Check+Var.)	17.72***	0.94	0.74	35.39***	6.42***	6.25	16635.12**
Entries (ignoring Blocks)	5.47*	2.01**	2.00	12.28***	2.31**	2.31	17492.58***
Checks	5.72	1.93*	0.05	5.00	1.79	2.26	27762.50***
Varieties	2.97	1.87**	2.07	12.38***	1.63*	1.72	15530.23***
Checks vs. Varieties	202.27***	13.15***	0.68	19.08**	56.51* * *	48.94***	151978.33***
ERROR	2.43	0.50	1.77	1.51	0.71	2.75	2435.12
Ci – Cj	1.67	0.76	1.43	1.32	0.91	1.78	52.92
BiVi - BiVj	4.73	2.15	4.03	3.73	2.56	5.03	149.68
BiVi - BjVj	5.46	2.48	4.66	4.31	2.96	5.81	172.83
Ci – VI	4.09	1.86	3.49	3.23	2.22	4.36	129.62

contd...

(Table 2 cont…)

	Rhizome Yield (t/ha)	Curing %	Curcumin %	Days to maturity
Block (ignoring Treatments)	487.12 ***	55.14***	2.53***	159.01
Treatment (eliminating Blocks)	76.60 ***	20.51***	0.78***	158.64
Checks	173.50 **	6.29***	0.30***	1366.17***
Checks+Var vs. Var.	74.18 ***	20.86***	0.79***	128.45
ERROR	15.21	0.29	0.00	83.12
Block (eliminating Check+Var.)	103.95 **	0.04	0.00	46.19
Entries (ignoring Blocks)	109.31 ***	25.21***	1.00***	168.27
Checks	173.50 **	6.29***	0.30***	1366.17***
Varieties	97.05 ***	23.71***	0.98***	129.24
Checks vs. Varieties	949.73 ***	181.06***	3.60***	855.76**
ERROR	15.21	0.29	0.00	83.12
Ci - Cj	4.18	0.58	0.03	9.78
BiVi - BiVj	11.83	1.64	0.08	27.65
BiVi - BjVj	13.66	1.89	0.09	31.93
Ci - VI	10.24	1.42	0.07	23.95

Table: 3 a Growth parameters of turmeric accessions (Means)

S.no	Accession	Plant height(cm)	Basal stem diameter (cm)	No of leaves	Leaf length (cm)	Leaf width (cm)	No of tillers	Leaf area (cm^2)	Length of leaf petiole (cm)
		1	2	3	4	5	6	7	8
1	Aleppy supreme	167.60	11.96	8.78	70.37	16.50	2.32	836.55	15.463
2	Amritpani	176.20	10.32	9.18	68.60	15.08	2.72	743.98	4.329
3	Avidi	154.43	11.45	9.78	64.47	17.73	1.62	826.32	13.996
4	Badipadar	167.93	11.42	9.91	71.47	16.22	1.95	834.28	7.563
5	Bataguda	130.40	7.96	7.18	56.57	13.26	2.32	535.55	8.763
6	BSR-2	163.43	9.55	4.47	66.14	11.02	2.22	511.45	11.163
7	Chintapalli local palli 1	153.83	12.25	7.78	68.67	17.79	1.82	881.02	19.796
8	Chintapalli local -2	138.63	11.77	8.57	61.67	18.73	2.52	836.62	15.496
9	CC 94-01	157.00	13.10	8.78	72.97	15.68	1.67	822.85	9.763
10	CL-2	158.03	11.81	7.47	67.54	14.62	1.42	706.75	12.563
11	CL-7	164.63	13.99	8.68	70.74	15.62	2.02	794.35	18.863
12	CL-9	152.43	10.31	7.68	53.34	16.72	1.82	641.15	12.363
13	CL-12	173.23	10.93	7.28	67.74	16.42	2.52	801.15	24.663
14	CL-16	155.83	10.23	7.88	59.14	13.92	1.42	585.55	21.663
15	CL-17	156.03	12.05	8.27	65.14	17.58	2.42	827.35	20.663
16	CLI 317	118.23	10.45	7.38	54.27	17.21	1.82	680.22	17.996
17	CLL-335	186.33	12.04	10.31	59.07	19.78	2.35	841.18	5.963
18	Cuddapah local	176.03	12.21	8.27	65.14	16.32	1.42	765.15	9.963
19	Dindigam	178.40	12.70	9.57	69.00	18.66	2.42	930.18	6.629
20	Duggirala	180.23	12.19	8.57	66.47	15.21	2.02	727.98	18.296
21	Ernad Chand	190.53	10.80	10.11	67.07	11.42	2.75	547.98	5.862
22	Ethamukkala	174.00	11.44	7.77	66.80	18.48	2.12	892.18	5.929
23	Flourescent	120.33	7.70	6.91	47.47	10.34	1.20	352.68	6.762
24	GL puram	163.13	13.06	9.51	73.27	14.69	1.20	772.98	6.963
25	Gorakgpur-361	113.20	10.38	6.97	52.20	14.40	1.92	542.18	5.729
26	GS	157.03	11.95	7.07	66.94	17.08	1.42	825.05	10.163
27	IC-033007	104.30	6.84	7.04	46.80	12.81	2.10	419.58	11.429
28	IC-181919	151.86	11.68	7.01	71.80	13.46	1.89	710.55	6.696
29	IC-211360	107.70	9.32	6.44	41.80	13.05	2.30	380.18	9.529
30	IC-211381	108.06	10.74	7.44	51.14	14.52	1.39	531.93	10.163
31	IC-211401	113.46	9.32	7.01	53.20	14.88	2.09	573.65	7.896
32	IC-211402	147.86	10.06	7.41	56.80	15.14	1.89	622.05	8.596
33	IC-211641	104.30	8.66	7.84	46.20	16.55	1.30	546.38	12.129
34	IC-211642	122.66	10.54	7.21	56.60	14.70	1.09	604.35	9.996
35	IC-212267	120.86	8.22	7.04	52.34	15.34	2.19	573.08	4.762
36	IC-212578	114.10	8.86	7.84	45.20	15.03	3.80	481.58	14.929
37	IC-212585	106.06	10.74	7.44	51.14	14.52	1.39	531.93	6.762
38	IC-212605	151.66	9.36	7.41	69.80	17.22	2.09	859.15	11.596
39	IC-212606	145.70	9.88	8.04	58.40	15.25	2.10	636.88	11.429
40	IC-319341	99.50	8.74	6.24	41.00	12.57	2.10	357.28	12.529
41	IC-319621	129.86	10.58	7.64	61.54	14.82	1.74	656.26	12.762
42	IC-330113	150.10	9.58	7.64	60.40	16.07	1.55	696.68	15.829
43	IC- 332957	95.50	9.56	7.24	38.80	12.19	1.55	325.48	11.429
44	IC-353560	120.26	9.94	6.81	62.40	13.12	1.14	604.35	10.796
45	IC-394268	146.66	9.82	7.84	63.54	12.70	3.79	588.18	12.762

Contd…..

(Table 3a Cont …)

S.no	Accession	Plant height(cm)	Basal stem diameter (cm)	No of leaves	Leaf length(cm)	Leaf width(cm)	No of tillers	Leaf area(cm²)	Length of leaf petiole(cm)
		1	2	3	4	5	6	7	8
46	IC-394396	115.26	7.90	7.64	51.34	11.62	4.39	436.36	7.162
47	IC-394903	136.26	10.12	7.41	58.80	15.26	2.09	648.65	5.996
48	IC-416941	147.06	11.58	6.24	71.94	13.80	1.39	721.03	10.163
49	IC-420474	92.26	9.02	5.84	51.14	13.16	0.99	486.30	13.563
50	IC-420556	154.86	11.12	7.44	70.74	16.96	1.19	859.34	9.862
51	IC-420606	94.06	9.86	6.61	51.60	13.10	0.89	499.85	11.396
52	IC-521333	157.06	10.64	7.44	71.34	16.00	1.99	820.69	9.462
53	IC-540383	118.66	9.20	7.81	57.60	16.08	1.89	664.75	10.996
54	IC-540386	111.10	8.36	7.04	46.80	13.87	2.30	457.58	10.229
55	IC-540387	112.10	8.96	7.64	46.00	14.89	1.80	485.48	14.029
56	IC-545139	142.86	9.68	8.41	66.00	14.14	2.09	681.95	4.896
57	Imphal local	164.83	12.35	8.78	74.07	15.77	1.02	839.22	18.996
58	KTS-6	183.63	12.77	8.18	70.67	16.33	1.42	830.82	14.096
59	KTS-7	188.03	12.15	8.38	79.27	18.91	2.22	1076.32	14.096
60	KTS-8	166.13	12.80	10.11	72.27	16.26	1.15	845.58	9.862
61	Lacadong	151.40	10.60	6.97	63.17	12.94	2.32	583.15	7.023
62	Meghaturmeric	173.73	12.14	8.71	74.07	15.86	1.35	844.58	11.862
63	Movatupuzha	134.60	11.18	6.97	52.97	11.94	1.52	448.75	12.263
64	Mydukur	161.40	12.86	9.18	72.80	16.94	1.32	888.58	5.729
65	Ochira	160.33	10.52	9.51	65.07	17.02	1.20	798.78	10.262
66	Paderu local	131.63	10.41	7.78	59.27	16.17	2.27	695.12	15.996
67	Penakanametta	164.23	9.45	8.57	66.27	15.59	1.22	745.52	14.896
68	Rajapuri	152.20	8.98	7.77	63.80	15.94	2.32	733.08	6.629
69	Rajendra sonia	137.53	10.55	7.28	51.34	12.60	3.02	454.15	16.962
70	Salem	183.00	9.56	8.18	70.77	11.56	1.52	580.95	6.983
71	Sugandham	164.20	12.00	8.38	68.20	15.58	2.72	764.78	5.729
72	TCP 70	141.53	11.70	8.71	62.47	18.34	1.95	827.88	14.062
73	Tekuripeta	150.20	9.06	8.38	64.40	12.36	1.67	569.78	12.329
74	Tenali	113.00	10.62	6.97	53.97	13.90	1.92	536.85	11.363

75	Thodupuzha	164.00	8.98	8.18	64.17	15.72	1.12	725.75	6.363
76	T Sundar	188.33	16.90	7.71	85.87	19.50	2.95	1205.18	9.862
77	Vikici	155.20	10.64	8.18	68.17	15.94	1.59	782.05	8.763
78	Vontimitta	160.00	13.58	9.18	67.40	16.64	1.17	808.58	7.229
79	Wagon	172.20	9.22	6.78	61.17	11.96	2.92	519.75	7.663
80	Wynad local	150.00	10.48	7.77	61.10	13.86	3.32	608.68	7.009
Check 1	CLS 269	175.45	10.88	8.32	68.38	15.28	2.37	751.61	11.113
Check 2	KTS-3	170.29	12.41	8.28	72.22	15.86	1.69	825.53	11.000
Check 3	IISR Prathibha	178.75	12.71	9.53	75.81	17.13	3.00	934.51	11.475
	Mean	146.80	10.70	7.89	62.09	15.17	1.95	683.73	11.045
	Std. Error	2.84	0.18	0.11	1.18	0.23	0.07	19.65	0.470
	CD at5%	5.45	0.48	0.31	3.07	0.31	0.18	45.23	1.879

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Table :3 b Yield parameters of turmeric accessions (Means)

S.no	Accession	No of mother rhizomes	Length of mother rhizome (cm)	Girth of mother rhizome (cm)	No of primary rhizomes	Length of primary rhizome (cm)	Girth of primary rhizome (cm)	No of secondary rhizomes	Length of secondary rhizome (cm)
		1	2	3	4	5	6	7	8
1	Aleppy supreme	2.99	5.46	8.06	6.18	7.22	7.69	5.10	2.07
2	Amritpani	1.45	9.80	12.57	8.85	8.31	10.01	8.68	5.64
3	Avidi	1.95	4.70	11.71	7.08	12.27	8.25	5.43	6.15
4	Badipadar	1.12	6.44	9.84	6.65	6.76	6.74	13.70	6.75
5	Bataguda	2.99	3.03	8.72	4.38	6.36	7.02	11.04	1.47
6	BSR-2	4.35	9.61	14.71	8.31	7.43	7.33	5.25	3.46
7	Chintapalli local palli 1	1.95	4.13	12.09	8.48	11.47	8.38	5.30	6.21
8	Chintapalli local -2	2.15	4.97	9.76	9.28	8.87	5.92	1.20	7.21
9	CC 94-01	1.99	5.98	8.15	4.58	7.96	7.82	7.37	1.40
10	CL-2	1.65	9.70	9.79	6.31	10.26	7.86	4.39	2.60

11	CL-7	2.35	9.56	9.64	7.91	10.00	8.53	12.05	3.40
12	CL-9	2.95	9.10	10.44	8.11	9.66	7.90	4.92	3.80
13	CL-12	2.75	9.16	9.88	6.11	8.53	7.46	5.05	3.06
14	CL-16	3.55	8.40	10.34	6.31	8.66	7.20	4.85	3.86
15	CL-17	1.95	10.56	13.57	7.31	9.83	8.56	5.52	3.62
16	CLI 317	2.05	5.06	11.01	8.68	9.47	6.98	10.43	5.55
17	CLL-335	2.32	5.14	10.51	9.98	6.96	7.28	6.85	4.65
18	Cuddapah local	2.15	8.80	13.34	7.31	9.80	9.66	4.59	2.26
19	Dindigam	1.05	10.77	12.93	6.45	6.37	7.08	11.68	5.64
20	Duggirala	1.75	7.56	11.68	7.88	8.80	8.65	5.05	6.15
21	Ernad Chand	1.32	5.81	5.30	5.65	6.49	8.94	8.35	5.88
22	Ethamukkala	1.15	9.87	12.53	7.85	7.64	9.15	9.75	5.30
23	Flourescent	1.92	4.87	5.54	5.05	5.82	6.61	13.65	5.28
24	GL puram	1.72	5.54	10.21	5.65	8.02	7.34	7.31	5.81
25	Gorakgpur-361	2.05	7.47	10.53	5.05	6.64	8.25	9.75	4.04
26	GS	2.75	8.92	10.78	6.91	8.13	9.26	12.65	3.40
27	IC-033007	3.49	5.87	7.09	3.05	7.80	6.81	2.63	2.90
28	IC-181919	3.19	6.77	15.36	5.01	6.39	6.16	9.34	6.14
29	IC-211360	2.89	5.97	10.19	5.85	6.97	6.01	7.31	0.90
30	IC-211381	2.75	6.68	12.08	6.58	7.67	8.01	5.94	5.73
31	IC-211401	2.19	5.87	11.94	0.31	7.52	5.12	3.10	5.04
32	IC-211402	2.19	7.32	14.01	1.91	9.25	6.86	3.84	5.70
33	IC-211641	3.19	5.27	10.69	7.65	6.80	5.27	0.63	2.63
34	IC-211642	1.89	6.14	12.74	0.31	7.05	6.92	4.30	5.17
35	IC-212267	4.45	5.84	11.94	6.98	8.77	7.41	2.87	4.19
36	IC-212578	2.59	5.17	9.99	5.25	8.73	6.21	0.90	2.70
37	IC-212585	2.35	4.28	9.08	7.18	9.37	7.61	6.24	8.45
38	IC-212605	2.29	6.07	8.76	5.51	4.69	3.26	3.94	6.04
39	IC-212606	2.99	6.07	8.49	5.95	7.07	6.91	7.11	1.37
40	IC-319341	2.79	5.27	6.89	2.65	7.47	4.41	7.31	2.07
41	IC-319621	2.45	7.13	12.91	5.98	10.43	8.21	4.20	6.26
42	IC-330113	3.79	5.67	9.78	6.05	9.07	8.41	2.16	3.90
43	IC- 332957	2.19	2.97	5.69	3.45	5.87	5.91	4.51	0.87
44	IC-353560	1.89	7.67	11.36	2.81	5.79	6.26	5.44	7.04
45	IC-394268	2.95	6.89	13.18	2.98	8.10	7.47	2.40	4.59

contd...

(Table 3b Cont …)

S.no	Accession	No of mother rhizomes	Length of mother rhizome (cm)	Girth of mother rhizome (cm)	No of primary rhizomes	Length of primary rhizome (cm)	Girth of primary rhizome (cm)	No of secondary rhizomes	Length of secondary rhizome (cm)
		1	2	3	4	5	6	7	8
46	IC-394396	3.15	6.13	12.28	4.38	6.83	6.27	2.54	3.79
47	IC-394903	3.49	6.27	12.06	6.41	7.89	6.16	6.84	9.04
48	IC-416941	4.45	9.58	11.98	9.58	8.87	7.91	8.84	5.43
49	IC-420474	1.55	5.48	11.08	6.18	9.27	7.61	4.34	10.23
50	IC-420556	3.15	6.93	13.28	5.38	10.30	7.96	3.60	6.19
51	IC-420606	3.19	7.37	12.46	1.71	6.99	4.86	5.64	10.44
52	IC-521333	3.55	6.68	13.11	5.78	10.50	8.47	4.20	5.39
53	IC-540383	1.89	6.62	12.33	0.51	7.39	6.99	3.10	5.24
54	IC-540386	3.19	4.59	7.63	0.65	6.00	6.27	5.97	3.07
55	IC-540387	3.19	5.47	10.99	5.05	8.73	6.27	5.97	2.30
56	IC-545139	4.59	8.33	15.40	0.91	7.59	5.99	3.94	4.77
57	Imphal local	2.85	5.43	14.55	6.48	10.00	9.25	4.96	9.85
58	KTS-6	1.75	5.11	11.87	8.48	7.87	10.32	5.90	5.01
59	KTS-7	1.95	6.31	14.34	9.58	8.27	11.32	8.70	6.55
60	KTS-8	1.82	5.31	12.09	4.45	6.09	9.01	10.85	5.48
61	Lacadong	1.79	2.40	6.43	3.98	8.09	6.62	6.04	2.40
62	Meghaturmeric	2.32	5.37	9.37	5.65	7.22	5.34	8.91	5.55
63	Movatupuzha	3.59	4.03	5.81	2.58	7.16	5.42	3.24	0.80
64	Mydukur	1.65	9.59	15.03	5.65	8.84	8.95	12.75	5.10
65	Ochira	1.62	5.87	13.04	5.25	6.89	7.48	6.78	5.68
66	Paderu local	1.55	5.56	12.26	7.88	10.74	7.98	5.96	5.55
67	Penakanametta	2.35	5.98	14.43	7.88	11.14	10.78	1.70	6.68
68	Rajapuri	2.15	11.17	10.43	9.65	6.37	5.28	5.68	4.39
69	Rajendra sonia	1.95	9.46	8.00	6.31	8.06	5.66	7.45	3.46
70	Salem	1.79	4.97	8.72	3.38	6.49	5.36	3.50	1.87
71	Sugandham	1.35	10.35	10.57	6.45	8.31	6.15	5.61	6.44
72	TCP 70	1.92	7.96	12.06	6.05	6.62	4.41	7.11	7.28
73	Tekuripeta	1.15	9.07	11.03	7.05	9.31	6.61	7.15	5.64
74	Tenali	1.99	2.10	5.20	2.18	7.16	3.62	7.37	0.94

75	Thodupuzha	1.69	5.13	9.43	3.58	6.42	7.36	7.37	2.54
76	T Sundar	1.02	6.04	10.87	8.05	7.02	8.21	9.18	6.61
77	Vikici	2.39	4.75	7.94	5.18	8.29	7.62	3.54	2.67
78	Vontimitta	2.35	9.42	12.53	5.65	8.10	7.61	15.81	4.37
79	Wagon	2.29	3.95	7.34	2.38	5.76	8.09	4.70	2.00
80	Wynad local	1.05	9.74	9.74	8.25	6.97	6.68	9.15	5.97
Check 1	CLS 269	2.83	6.07	9.70	8.39	7.37	7.34	7.03	6.55
Check 2	KTS-3	2.64	6.81	10.37	8.68	7.50	7.46	6.69	5.91
Check 3	IISR Prathibha	2.70	6.42	10.71	9.95	6.60	7.49	8.20	6.84
	Mean	2.40	6.64	10.75	5.82	7.97	7.25	6.39	4.75
	Std. Error	0.09	0.22	0.27	0.26	0.16	0.16	0.36	0.24
	CD at5%	0.45	1.56	1.36	1.67	0.76	1.43	1.32	0.91

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(Table 3b Cont …)

S.no	**Accession**	**Girth of secondary rhizomes (cm)**	**Yield per plant (g)**	**Rhizome yield (t ha-1)**	**Curing %**	**Curcumin content (%)**	**Days to maturity**
		9	10	11	12	13	14
1	Aleppy supreme	5.01	19.96	4.78	216.50	388.75	30.73
2	Amritpani	6.80	16.96	3.68	204.50	375.42	29.67
3	Avidi	9.24	19.96	2.00	191.50	222.08	17.56
4	Badipadar	4.41	25.29	3.08	225.17	530.42	41.94
5	Bataguda	4.45	21.96	3.38	225.50	238.75	18.88
6	BSR-2	3.60	16.96	2.38	231.83	488.75	38.64
7	Chintapalli local palli 1	8.05	23.46	1.75	205.50	222.08	17.56
8	Chintapalli local -2	5.91	23.96	3.55	197.50	322.08	25.46
9	CC 94-01	7.41	19.96	2.58	220.50	223.75	17.69
10	CL-2	3.93	22.96	1.88	219.83	316.25	25.00
11	CL-7	3.77	24.96	2.78	221.83	538.75	42.59
12	CL-9	4.86	22.96	3.18	225.83	513.75	40.61
13	CL-12	3.66	23.96	2.78	207.83	316.75	25.04
14	CL-16	4.40	9.96	2.78	235.83	378.75	29.94

15	CL-17	4.60	13.96	3.78	227.83	423.75	33.50
16	CLI 317	10.65	19.96	3.85	216.50	277.08	21.91
17	CLL-335	6.65	17.29	5.48	219.17	755.42	59.72
18	Cuddapah local	3.93	17.96	2.78	240.83	315.75	24.96
19	Dindigam	6.56	16.96	3.38	223.50	325.42	25.72
20	Duggirala	7.25	19.96	4.75	242.00	422.08	33.37
21	Ernad Chand	5.68	15.29	3.48	221.17	350.42	27.71
22	Ethamukkala	4.76	21.96	4.48	242.50	375.42	29.67
23	Flourescent	4.48	12.29	4.18	216.17	445.42	35.22
24	GL puram	4.88	20.29	3.48	228.17	555.42	43.91
25	Gorakgpur-361	3.56	10.96	2.88	208.50	375.42	29.67
26	GS	4.40	14.96	2.18	226.83	488.75	38.64
27	IC-033007	3.81	23.96	2.19	208.50	218.75	17.30
28	IC-181919	6.45	24.96	1.88	202.83	493.75	39.03
29	IC-211360	3.38	24.96	3.45	213.50	308.75	24.41
30	IC-211381	4.92	23.96	2.28	233.17	282.08	22.29
31	IC-211401	6.75	19.96	1.48	208.83	293.75	23.22
32	IC-211402	7.41	28.96	1.58	209.83	468.75	37.06
33	IC-211641	2.68	19.96	1.95	200.50	568.75	44.96
34	IC-211642	8.01	23.96	2.38	211.83	298.75	23.62
35	IC-212267	4.18	23.96	2.78	210.17	197.08	15.57
36	IC-212578	3.48	23.96	3.25	209.50	398.75	31.52
37	IC-212585	7.32	25.96	2.06	222.17	330.08	26.09
38	IC-212605	5.15	19.96	2.78	212.83	223.75	17.69
39	IC-212606	4.68	27.76	2.17	200.50	568.75	44.96
40	IC-319341	3.08	25.96	3.37	208.50	298.75	23.62
41	IC-319621	6.38	31.46	1.36	215.17	365.08	28.85
42	IC-330113	5.94	25.96	3.65	194.50	368.75	29.15
43	IC- 332957	2.08	31.16	2.85	215.50	343.75	27.18
44	IC-353560	5.55	24.96	2.08	226.83	243.75	19.27
45	IC-394268	4.18	21.66	2.68	200.17	242.08	19.13

Contd…

(Table 3b Cont …)

S.no	Accession	Girth of secondary rhizomes (cm)	Yield per plant (g)	Rhizome yield (t ha-1)	Curing %	Curcumin content (%)	Days to maturity
		9	10	11	12	13	14
46	IC-394396	5.22	23.96	3.78	221.17	222.08	17.55
47	IC-394903	5.55	24.96	2.18	204.83	273.75	21.64
48	IC-416941	3.92	26.96	2.18	216.17	212.08	16.76
49	IC-420474	5.72	21.96	2.08	186.17	240.08	18.97
50	IC-420556	6.72	23.96	1.58	193.17	235.08	18.58
51	IC-420606	6.85	13.96	2.48	220.83	373.75	29.55
52	IC-521333	4.72	29.96	1.78	200.17	262.08	20.71
53	IC-540383	7.48	23.96	1.78	223.83	193.75	15.32
54	IC-540386	2.81	30.96	1.85	206.50	418.75	33.11
55	IC-540387	2.48	25.96	3.55	206.50	418.75	33.11
56	IC-545139	6.48	19.96	2.88	202.83	162.75	12.87
57	Imphal local	7.65	22.96	3.55	204.50	222.08	17.56
58	KTS-6	7.98	22.96	3.65	222.50	272.08	21.51
59	KTS-7	9.57	16.96	2.25	243.50	472.08	37.32
60	KTS-8	5.95	22.29	4.48	241.17	510.42	40.35
61	Lacadong	6.55	25.96	2.78	210.50	288.75	22.83
62	Meghaturmeric	5.15	25.29	2.48	202.17	330.42	26.13
63	Movatupuzha	5.45	17.96	3.48	243.50	288.75	22.83
64	Mydukur	4.96	18.96	3.23	247.50	445.42	35.20
65	Ochira	4.10	24.29	6.58	213.17	482.42	38.14
66	Paderu local	7.11	19.96	2.80	208.50	372.08	29.42
67	Penakanametta	9.05	14.96	2.85	194.50	254.08	20.30
68	Rajapuri	4.90	26.96	4.38	209.50	225.42	17.81
69	Rajendra sonia	2.86	16.96	4.58	208.83	303.75	24.01
70	Salem	4.41	17.96	4.58	241.50	173.75	13.74
71	Sugandham	5.16	24.96	3.88	209.50	475.42	37.58
72	TCP 70	3.88	25.29	2.08	205.17	355.42	28.10
73	Tekuripeta	1.85	28.46	2.88	214.50	395.42	31.25
74	Tenali	2.41	23.96	1.84	193.50	265.75	21.01
75	Thodupuzha	5.95	14.96	3.58	210.50	263.75	20.85
76	T Sundar	5.68	26.29	6.73	202.17	560.42	44.31
77	Vikici	4.95	28.96	1.78	222.50	188.75	14.92

78	Vontimitta	4.30	23.96	3.68	217.50	435.42	34.41
79	Wagon	4.21	15.96	3.18	205.50	438.75	34.69
80	Wynad local	5.50	19.96	2.88	222.50	207.42	16.39
Check 1	CLS 269	6.90	18.00	3.58	205.75	382.50	30.24
Check 2	KTS-3	6.45	19.75	3.20	248.75	433.75	34.29
Check 3	IISR Prathibha	7.51	19.13	3.47	241.00	500.00	39.53
	Mean	5.38	21.92	3.04	215.90	352.50	27.87
	Std. Error	0.20	0.53	0.11	1.30	13.06	1.03
	CD at5%	1.78	0.58	0.03	9.78	52.92	4.18

Table:4 Qualitative parameters of turmeric accessions

S.no	Accession	Leaf disposition pattern	Colour of dorsal surface of leaf	Spatial arrangement of veins on leaves	Prominence of leaf venation	Rhizome core colour	Flowering
1	Aleppy supreme	Semi errect	Dark green	Close	Less prominent	Orange yellow	Absent
2	Amritpani	Errect	Green	Close	Less prominent	Lemon yellow	Absent
3	Avidi	Errect	Green	Close	Less prominent	Orange yellow	Absent
4	Badipadar	Semi errect	Green	Close	More prominent	Orange yellow	Absent
5	Bataguda	Semi errect	Green	Close	Less prominent	Orange yellow	Absent
6	BSR-2	Errect	Dark green	Close	Less prominent	Light yellow	Absent
7	Chintapalli local- 1	Semi errect	Dark green	Distant	More prominent	Lemon yellow	Present
8	Chintapalli local-2	Semi errect	Dark green	Close	Less prominent	Lemon yellow	Absent
9	CC 94-01	Errect	Green	Close	More prominent	Light yellow	Absent
10	CL-2	Semi errect	Green	Close	Less prominent	Light yellow	Absent
11	CL-7	Errect	Green	Close	More prominent	Orange yellow	Absent
12	CL-9	Errect	Green	Distant	More prominent	Light yellow	Absent

13	CL-12	Errect	Dark green	Distant	More prominent	Orange yellow	Present
14	CL-16	Semi errect	Green	Distant	More prominent	Light yellow	Absent
15	CL-17	Semi errect	Green	Close	Less prominent	Light yellow	Absent
16	CLI 317	Semi errect	Dark green	Close	Less prominent	Light yellow	Present
17	CLL-335	Semi errect	Green	Distant	Less prominent	Dark orange yellow	Absent
18	Cuddapah local	Errect	Green	Distant	Less prominent	Light yellow	Present
19	Dindigam	Semi errect	Dark green	Distant	Less prominent	Orange yellow	Absent
20	Duggirala	Semi errect	Green	Close	Less prominent	Orange yellow	Present
21	Ernad Chand	Semi errect	Green	Close	Less prominent	Orange yellow	Absent

Contd…

(Table 4 cont …)

S.no	**Accession**	**Leaf disposition pattern**	**Colour of dorsal surface of leaf**	**Spatial arrangement of veins on leaves**	**Prominence of leaf venation**	**Rhizome core colour**	**Flowering**
22	Ethamukkala	Semi errect	Green	Diatant	More prominent	Orange yellow	Absent
23	Flourescent	Errect	Green	Close	Less prominent	Orange yellow	Present
24	GL Puram	Semi	Dark green	Close	Less	Orange	Absent

		errect			promiment	yellow	
25	Gorakhpur-361	Errect	Dark green	Close	Lessprominent	Orange yellow	Absent
26	GS	Semi errect	Green	Close	Less prominent	Dark orange yellow	Absent
27	IC-033007	Errect	Green	Close	Less prominent	Dark orange yellow	Absent
28	IC-181919	Errect	Green	Close	More prominent	Orange yellow	Absent
29	IC-211360	Semi errect	Green	Distant	Less prominent	Lemon yellow	Present
30	IC-211381	Errect	Green	Close	Less prominent	Orange yellow	Present
31	IC-211401	Semi errect	Green	Distant	More prominent	Lemon yellow	Present
32	IC-211402	Semi errect	Dark green	close	More prominent	Dark orange yellow	Absent
33	IC-211641	Semi errect	Green	Distant	More prominent	Orange yellow	Present
34	IC-211642	Errect	Green	Distant	More prominent	Orange yellow	Absent
35	IC-212267	Semi errect	Green	close	More prominent	Lemon yellow	Present
36	IC-212578	Errect	Green	Distant	More prominent	Lemon yellow	Present
37	IC-212585	Semi errect	Green	Distant	Less prominent	Lemon yellow	Present

IC-212585	Semi errect	Green	Dista
IC-212605	Semi errect	Green	Dista

38	IC-212605	Semi errect	Green	Distant	More prominent	Dark orange yellow	Absent
39	IC-212606	Errect	Green	Distant	More prominent	Dark orange yellow	Present
40	IC-319341	Semi errect	Green	Distant	More prominent	Lemon yellow	Present
41	IC-319621	Errect	Green	Distant	More prominent	Light yellow	Absent

IC-212606	Errect	Green	Distar
IC-319341	Semi errect	Green	Distar
IC-319621	Errect	Green	Distar
IC-330113	Semi errect	Dark green	close

Contd…

(Table 4 cont …)

S.no	**Accession**	**Leaf disposition pattern**	**Colour of dorsal surface of leaf**	**Spatial arrangement of veins on leaves**	**Prominence of leaf venation**	**Rhizome core colour**	**Flowering**
42	IC-330113	Semi errect	Dark green	close	More prominent	Dark orange yellow	Absent
43	IC-332957	Errect	Green	Distant	less prominent	Lemon yellow	Present
44	IC-353560	Errect	Green	close	More prominent	Dark orange yellow	Absent
45	IC-394268	Errect	Green	Distant	More prominent	Lemon yellow	Absent
46	IC-394396	Errect	Green	Distant	less prominent	Orange yellow	Present

47	IC-394903	Semi errect	Dark green	close	More prominent	Lemon yellow	Present
48	IC-416941	Errect	Green	Distant	More prominent	Orange yellow	Absent
49	IC-420474	Semi errect	Green	Distant	More prominent	Lemon yellow	Absent
50	IC-420556	Errect	Green	close	More prominent	Lemon yellow	Absent
51	IC-420606	Semi errect	Green	Distant	More prominent	Lemon yellow	Present
52	IC-521333	Semi errect	Dark green	Distant	More prominent	Dark orange yellow	Absent
53	IC-540383	Errect	Green	Distant	Less prominent	Lemon yellow	Present
54	IC-540386	Errect	Green	Distant	More prominent	Orange yellow	Present
55	IC-540387	Errect	Green	Distant	More prominent	Lemon yellow	Present
56	IC-545139	Semi errect	Green	Distant	More prominent	Orange yellow	Absent
57	Imphal local	Errect	Green	Close	More prominent	Dark orange yellow	Absent
58	KTS-6	Semi errect	Green	Close	Less prominent	Orange yellow	Absent
59	KTS-7	Errect	Green	Close	Less prominent	Light yellow	Absent
60	KTS-8	Errect	Green	Distant	More prominent	Orange yellow	Absent
61	Lacadong	Semi errect	Dark green	Distant	Less prominent	Dark orange	Present

						yellow	
62	Meghaturmeric	Errect	Green	Distant	Lesspromine nt	Lemon yellow	Absent
63	Movatupuzha	Errect	Green	Close	Less prominent	Light yellow	Absent
64	Mydukur	Errect	Dark green	Distant	More prominent	Light yellow	Absent
65	Ochira	Errect	Green	Close	More prominent	Light yellow	Absent

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(Table 4 cont...)

S.no	**Accession**	**Leaf disposition pattern**	**Colour of dorsal surface of leaf**	**Spatial arrangement of veins on leaves**	**Prominence of leaf venation**	**Rhizome core colour**	**Flowering**
66	Paderu local	Errect	Dark green	Distant	More proinent	Orange yellow	Present
67	Penakanametta	Errect	Green	Distant	More prominent	Lemon yellow	Absent
68	Rajapuri	Errect	Green	Close	Less prominent	Dark orange yellow	Absent
69	Rajendra sonia	Errect	Dark green	Close	Lesspromine nt	Orange yellow	Present
70	Salem	Semi errect	Green	Close	More prominent	Orange yellow	Absent
71	Sugandham	Errect	Green	Close	Less prominent	Light yellow	Absent

72	TCP 70	Errect	Dark green	Distant	Les prominent	Light yellow	Present
73	Tekuripeta	Semi errect	Green	Close	Less prominent	Light yellow	Absent
74	Tenali	Errect	Dark green	Distant	More prominent	Light yellow	Present
75	Thodupuzha	Errect	Green	Close	Less prominent	Orange yellow	Absent
76	T Sundar	Errect	Green	Close	Less prominent	Orange yellow	Present
77	Vikici	Semi errect	Green	Close	Less prominent	Light yellow	Absent
78	Vontimitta	Errect	Green	Close	Less prominent	Orange yellow	Absent
79	Wagon	Errect	Dark green	Close	More prominent	Orange yellow	Present
80	Wynad local	Semi errect	Green	Close	Less prominent	Dark orange yellow	Absent
81	CLS - 269	Errect	Green	Close	More prominent	Dark orange yellow	Absent
82	KTS-3	Errect	Dark green	Close	Less prominent	Orange yellow	Absent
83	IISR Prathibha	Semi errect	Green	Close	Less prominent	Orange yellow	Absent

Table 5: Estimates of variability and genetic parameters for yield and yield attributes in turmeric.

S. No.	Character	Mean	Range	Phenotypic coefficient of variation	Genotypic coefficient of variation	Heritability (broadsence) (%)	Genetic Advance as per cent Mean (%)(GAM)
1	Plant height (cm)	146.56	92.26-190.53	16.21	15.83	95.4	31.85
2	Basal stem diameter(cm)	10.68	6.84-16.89	14.06	13.43	91.2	26.43
3	Number of leaves	7.89	4.48-10.31	10.18	9.50	87.2	18.28
4	Leaf length(cm)	62.09	38.8-85.87	17.00	16.34	92.4	32.36
5	Leaf width(cm)	15.17	10.34-19.49	12.24	12.09	97.6	24.61
5	Leaf area (cm2)	683.73	204.52-1205.18	24.38	23.55	93.3	46.84
6	Number of tillers	1.95	0.89-4.39	31.77	30.59	92.7	60.68
7	Length of leaf petiole(cm)	11.05	4.33-24.66	34.69	30.85	79.1	56.50
8	Number of mother rhizomes	2.40	1.02-4.59	26.90	20.29	56.9	31.52
9	Length of mother rhizome (cm)	6.64	2.10-11.17	19.24	10.33	28.8	11.43
10	Girth of mother rhizome(cm)	10.75	5.20-15.40	22.42	19.09	72.5	33.47
11	Number of primary rhizomes	5.82	0.31-9.98	29.73	11.71	15.5	9.49
12	Length of primary rhizome (cm)	7.97	4.69-12.27	15.95	13.26	69.1	22.71
13	Girth of primary rhizome(cm)	7.25	3.26-11.32	19.60	6.85	12.2	4.93
14	Number of secondary rhizomes	6.39	0.64-15.81	51.41	47.55	85.5	90.58
15	Length of secondary rhizome(cm)	4.75	0.80-10.44	25.89	18.57	51.4	27.44
16	Girth of secondary rhizome(cm)	5.38	1.85-10.65	25.92	17.30	44.6	23.79
17	Yield per plant (g)	352.90	162.75-755.42	33.03	29.83	81.6	55.50
18	Rhizome yield (t ha^{-1})	27.87	12.18-59.72	33.03	29.83	81.6	55.50
19	Curing (%)	21.92	9.96-31.46	20.03	19.88	98.5	40.64
20	Curcumin content (%)	3.09	1.36-6.73	30.20	30.19	99.9	62.16
21	Days to maturity	215.90	186.17-248.75	5.14	2.88	31.3	3.32

PCV, GCV and Genetic Advance as per cent Mean: Low = 0-10%; Medium = 10-20%; High = 20% above
Heritability: Low = Less than 30%; Medium = 30-60%; High = More than 60%

Table 6: Phenotypic correlation co-efficients among yield and its components in turmeric

S.no	Character	Plant height (cm)	Basal stem diameter (cm)	No of leaves	Leaf length (cm)	Leaf width (cm)	No of tillers	Leaf area (cm²)	Length of leaf petiole (cm)	No of mother rhizomes	Length of mother rhizome (cm)	Girth of mother rhizome (cm)	No of primary rhizomes
1	Plant height(cm)	1.000	0.593 *	0.509	0.744 **	0.253	0.136	0.638*	-0.126	-0.135	0.565*	0.616*	0.114
2	Basal stem diameter (cm)		1.000	0.521	0.596 *	0.488	-0.091	0.680 **	0.067	-0.224	0.399	0.479	-0.116
3	No of leaves			1.000	0.445	0.480	0.062	0.554 *	-0.100	-0.294	0.306	0.346	0.137
4	Leaf length(cm)				1.000	0.354	0.009	0.872 ***	-0.098	-0.107	0.423	0.56*	0.097
5	Leaf width(cm)					1.000	-0.109	0.761 **	0.185	-0.129	0.214	0.342	0.093
6	No of tillers						1.000	-0.040	-0.094	-0.070	0.080	-0.027	-0.017
7	Leaf area(cm²)							1.000	0.028	-0.153	0.402	0.557*	0.026
8	Length of leaf petiole(cm)								1.000	0.086	-0.024	-0.025	0.008
9	No of mother rhizomes									1.000	-0.036	-0.046	0.195
10	Length of mother rhizome (cm)										1.000	0.527	-0.039
11	Girth of mother rhizome (cm)											1.000	-0.213
12	No of primary rhizomes												1.000
13	Length of primary rhizome (cm)												
14	Girth of primary rhizome (cm)												
15	No of secondary rhizomes												
16	Length of secondary rhizome(cm)												
17	Girth of secondary rhizome(cm)												
18	Yield per plant (g)												
19	Rhizome yield (tha^{-1})												
20	Curing %												
21	Curcumin content(%)												
22	Days to maturity												

Contd…

(Table 6: Cont...)

Character	Length of primary rhizome (cm)	Girth of primary rhizome (cm)	No of secondary rhizomes	Length of secondary rhizome (cm)	Girth of secondary rhizome (cm)	Yield per plant (g)	Yield (tha-)	Curing %	Curcumin content (%)	Days to maturity
Plant height(cm)	0.242	0.548 *	0.075	-0.008	0.347	0.477	0.477	-0.23	0.326	0.108
Basal stem diameter (cm)	0.336	0.369	0.249	0.129	0.300	0.491	0.491	-0.027	0.067	0.131
No of leaves	0.255	0.325	0.076	0.019	0.211	0.209	0.209	0.004	0.212	0.070
Leaf length(cm)	0.245	0.412	0.074	0.056	0.290	0.321	0.321	-0.109	0.079	0.051
Leaf width(cm)	0.277	0.209	0.130	0.157	0.151	0.119	0.119	0.145	-0.153	-0.131
No of tillers	-0.175	-0.111	-0.204	-0.198	-0.070	-0.013	-0.013	-0.015	0.156	-0.125
Leaf area(cm^2)	0.297	0.394	0.134	0.117	0.281	0.290	0.290	-0.003	-0.015	-0.027
Length of leaf petiole(cm)	0.325	-0.011	-0.021	0.142	0.114	-0.037	-0.037	0.064	-0.164	-0.071
No of mother rhizomes	-0.033	-0.129	-0.089	-0.101	-0.123	-0.185	-0.185	0.008	-0.085	-0.007
Length of mother rhizome (cm)	0.102	0.220	0.043	0.016	0.141	0.298	0.298	-0.242	0.296	0.255
Girth of mother rhizome (cm)	0.409	0.504	0.207	0.037	0.297	-0.012	-0120	-0.314	0.320	0.161
No of primary rhizomes	-0.113	-0.115	-0.063	0.074	-0.025	0.534*	0.534*	0.204	-0.051	-0.097
Length of primary rhizome (cm)	1.000	0.406	0.182	0.224	0.424	0.195	0.195	-0.004	-0.132	-0.172
Girth of primary rhizome(cm)		1.000	0.241	0.068	0.517	0.391	0.391	-0.280	0.097	0.126
No of secondary rhizomes			1.000	0.036	0.199	0.249	0.249	-0.076	0.014	0.046
Length of secondary rhizome (cm)				1.000	0.155	0.045	0.045	-0.053	-0.143	-0.177
Girth of secondary rhizome (cm)					1.000	0.282	0.282	-0.253	0.091	-0.012
Yield per plant (g)						1.000	1 ***	-0.263	0.325	0.138
Rhizome yield (tha^{-1})							1.000	-0.263	0.325	0.138
Curing %								1.000	-0.305	-0.162
Curcumin content(%)									1.000	0.257
Days to maturity										1.000

Table 7: Genotypic correlation coefficients among yield and its components in turmeric

Character	Plant height (cm)	Basal stem diameter (cm)	No of leaves	Leaf length (cm)	Leaf width (cm)	No of tillers	Leaf area (cm²)	Length of leaf petiole (cm)	No of mother rhizomes	Length of mother rhizome (cm)	Girth of mother rhizome (cm)
Plant height(cm)	1.000	0.611	0.579	0.768	0.268	0.154	0.664	-0.181	-0.202	0.747	0.745
Basal stem diameter (cm)		1.000	0.598	0.648	0.524	-0.068	0.739	0.106	-0.310	0.698	0.497
No of leaves			1.000	0.471	0.513	0.074	0.592	-0.149	-0.296	0.530	0.416
Leaf length(cm)				1.000	0.352	0.002	0.866	-0.152	-0.120	0.881	0.671
Leaf width(cm)					1.000	-0.112	0.767	0.206	-0.100	0.438	0.407
No of tillers						1.000	-0.048	-0.059	-0.167	0.226	-0.047
Leaf area(cm²)							1.000	0.001	-0.142	0.823	0.667
Length of leaf petiole(cm)								1.000	0.209	0.190	0.089
No of mother rhizomes									1.000	-0.271	0.056
Length of mother rhizome (cm)										1.000	0.533
Girth of mother rhizome (cm)											1.000
No of primary rhizomes											
Length of primary rhizome (cm)											
Girth of primary rhizome (cm)											
No of secondary rhizomes											
Length of secondary rhizome (cm)											
Girth of secondary rhizome (cm)											
Yield per plant (g)											
Rhizome yield (tha^{-1})											
Curing %											
Curcumin content(%)											
Days to maturity											

Contd…

(Table :7 cont…..)

Character	No of primary rhizomes	Length of primary rhizome (cm)	Girth of primary rhizome (cm)	No of secondary rhizomes	Length of secondary rhizome (cm)	Girth of secondary rhizome (cm)	Yield per plant (g)	Yield (t ha^{-1})	Curing %	Curcumin content (%)	Days to maturity
Plant height(cm)	0.082	0.311	0.611	0.084	0.009	0.441	0.509	0.509	-0.232	0.337	0.333
Basal stem diameter cm)	-0.166	0.384	0.392	0.247	0.146	0.436	0.540	0.540	-0.017	0.072	0.205
No of leaves	0.348	0.382	0.880	0.046	0.146	0.475	0.204	0.204	-0.016	0.225	0.164
Leaf length(cm)	0.082	0.406	0.469	0.080	0.160	0.464	0.380	0.380	-0.125	0.082	0.096
Leaf width(cm)	0.256	0.363	0.627	0.132	0.225	0.289	0.145	0.145	0.140	-0.155	-0.253
No of tillers	0.003	-0.214	-0.191	-0.188	-0.318	-0.024	0.081	0.081	-0.027	0.159	-0.215
Leaf area(cm^2)	0.184	0.461	0.388	0.141	0.228	0.478	0.345	0.345	-0.016	-0.016	-0.063
Length of leaf petiole(cm)	-0.083	0.417	-0.695	-0.024	0.173	-0.436	-0.196	-0.196	0.067	-0.181	0.223
No of mother rhizomes	0.871	-0.234	-0.210	-0.117	0.046	-0.323	-0.070	-0.070	0.056	-0.116	-0.434
Length of mother rhizome (cm)	-0.873	0.895	0.289	0.014	0.766	0.310	0.569	0.569	-0.461	0.552	-0.956
Girth of mother rhizome (cm)	-0.106	0.506	0.619	0.251	-0.023	0.521	-0.054	-0.054	-0.355	0.380	0.107
No of primary rhizomes	1.000	-0.735	-0.802	0.220	-0.353	-0.936	0.688	0.688	0.561	-0.138	0.974
Length of primary rhizome (cm)		1.000	0.732	0.212	0.115	0.313	0.288	0.288	0.037	-0.156	-0.221
Girth of primary rhizome (cm)			1.000	0.956	0.330	-0.070	0.740	0.740	-0.782	0.306	0.174
No of secondary rhizomes				1.000	0.072	0.496	0.276	0.276	-0.092	0.017	-0.149
Length of secondary rhizome (cm)					1.000	-0.368	0.092	0.092	-0.090	-0.201	0.117
Girth of secondary rhizome (cm)						1.000	0.169	0.169	-0.349	0.162	0.962
Yield per plant (g)							1.000	1.000	-0.287	0.365	0.483
Rhizome yield (tha^{-1})								1.000	-0.287	0.365	0.483
Curing %									1.000	-0.308	-0.258
Curcumin content(%)										1.000	0.459
Days to maturity											1.000

Table:8 Simple correlation coefficients among yield and its components in turmeric

Character	Plant height (cm)	Basal stem diameter (cm)	No of leaves	Leaf length(cm)	Leaf width (cm)	No of tillers	Leaf area (cm²)	Length of leaf petiole (cm)	No of mother rhizomes	Length of mother rhizome (cm)
Plant height(cm)	1.0000	0.6072 ***	0.5885 ***	0.6922***	0.3460**	0.0889	0.63***	0.0057	-0.2850**	0.3050**
Basal stem diameter (cm)		1.0000	0.5791 ***	0.5915***	0.5601***	-0.1113	0.70***	0.1476	-0.3445**	0.2474*
No of leaves			1.0000	0.4657***	0.5137***	0.0438	0.58***	-0.0307	-0.4708***	0.0816
Leaf length(cm)				1.0000	0.4123***	-0.0545	0.87***	-0.0977	-0.1736	0.2299**
Leaf width(cm)					1.0000	-0.0979	0.80***	0.2464*	-0.2354*	0.2142
No of tillers						1.0000	-0.08	-0.0418	-0.0165	0.0948
Leaf area(cm²)							1.000	0.0651	-0.2523*	0.2558*
Length of leaf petiole (cm)								1.0000	0.0844	-0.0036
No of mother rhizomes									1.0000	-0.0741
Length of mother rhizome (cm)										1.0000

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(Table:8 cont...)

Character		**No of primary rhizomes**	**Length of primary rhizome (cm)**	**Girth of primary rhizome (cm)**	**No of secondary rhizomes**	**Length of secondary rhizome (cm)**
Plant height(cm)	0.1899	0.4445***	0.0759	0.5059 ***	0.2556*	0.0819
Basal stem diameter (cm)	0.2277*	0.3829***	0.2068	0.3881 ***	0.3731** *	0.2371*
No of leaves	0.1147	0.2509*	0.0613	0.3194 **	0.2483*	0.2096
Leaf length(cm)	0.2887* *	0.2897**	0.0814	0.3452 **	0.2609*	0.2328*
Leaf width(cm)	0.3791* * *	0.3681***	0.3021**	0.3000 **	0.0742	0.2607*
No of tillers	-0.1086	0.1033	-0.1547	-0.1114	-0.1286	-0.1735
Leaf area(cm^2)	0.3865* * *	0.3810***	0.1977	0.3883 ***	0.2302*	0.3050**
Length of leaf petiole(cm)	0.0221	0.2109	0.4468***	0.1125	-0.2595*	-0.0033
No of mother rhizomes	0.1045	-0.1152	0.0499	-0.1475	-0.3155**	-0.1873
Length of mother rhizome (cm)	0.4638* * *	0.3176**	0.1456	0.1874	0.2412*	0.1643
Girth of mother rhizome (cm)	1.000	0.1936	0.3706***	0.3897 ***	-0.0235	0.4858***
No of primary rhizomes		1.0000	0.3359**	0.4319***	0.1744	0.2611*
Length of primary rhizome (cm)			1.0000	0.4737***	-0.2249*	0.1933
Girth of primary rhizome (cm)				1.0000	0.1768	0.1639
No of secondary rhizomes					1.0000	0.0515
Length of secondary rhizome (cm)						1.0000
Girth of secondary rhizome (cm)						
Yield per plant (g)						
Rhizome yield (tha^{-1})						
Curing %						
Curcumin content(%)						
Days to maturity						

Contd...

(Table:8 cont...)

Character	Girth of secondary rhizome(cm)	Curing %	Curcumin content%	Days to maturity	Rhizome yield (t/ha)	Yield per plant (g)
Plant height(cm)	0.2540*	-0.2439*	0.3170**	0.2274*	0.2417*	0.2416*
Basal stem diameter (cm)	0.2667*	-0.0399	0.0554	0.2172*	0.3042**	0.3042**
No of leaves	0.2480*	-0.0674	0.3245**	0.1243	0.2332*	0.2332*
Leaf length(cm)	0.2564*	-0.0754	-0.0404	0.1239	0.0684	0.0684
Leaf width(cm)	0.3255**	0.0791	-0.0741	-0.0996	0.0587	0.0588
No of tillers	-0.1226	0.0184	0.1547	-0.1153	0.0328	0.0328
Leaf area(cm^2)	0.3499**	-0.0089	-0.0598	0.0276	0.0832	0.0832
Length of leaf petiole(cm)	0.0616	-0.0241	-0.0341	-0.0944	-0.0321	-0.0322
No of mother rhizomes	-0.1718	0.1033	-0.2206*	-0.1089	-0.1364	-0.1364
Length of mother rhizome (cm)	-0.1371	-0.1584	0.0696	0.2317*	0.1218	0.1219
Girth of mother rhizome (cm)	0.3684***	-0.0493	-0.1116	-0.0118	0.0231	0.0233
No of primary rhizomes	0.1735	-0.1484	0.2471*	0.0565	0.2676*	0.2677*
Length of primary rhizome (cm)	0.2955**	0.0888	-0.2578*	-0.1980	-0.1599	-0.1598
Girth of primary rhizomes (cm)	0.3893***	-0.2145	0.0709	0.1579	0.1219	0.1220
No of secondary rhizomes	-0.0379	-0.1615	0.1894	0.2698*	0.3452**	0.3451**
Length of secondary rhizomes (cm)	0.5061***	-0.0355	-0.1029	-0.1212	-0.0035	-0.0034
Girth of secondary rhizomes (cm)	1.0000	-0.2065	-0.0579	-0.0470	-0.1538	-0.1538
Yield /plant (g)		1.0000	-0.3008**	-0.2566*	-0.0713	-0.0713
Rhizome yield (tha^{-1})			1.0000	0.2569*	0.2521*	0.2521*
Curing %				1.0000	0.1299	0.1299
Curcumin content(%)					1.0000	1.0000***
Days to maturity						1.0000

Table 9: Path co-efficients among yield and its components in turmeric

No	Character	Plant height(cm)	Basal stem diameter (cm)	No of leaves	Leaf length (cm)	Leaf width (cm)	No of tillers	Leaf area (cm²)	Length of leaf petiole (cm)	No of mother rhizomes
1	Plant height(cm)	**0.097**	0.059	0.057	0.067	0.034	0.009	0.061	0.001	-0.028
2	Basal stem diameter (cm)	0.225	**0.370**	0.214	0.219	0.207	-0.041	0.259	0.055	-0.128
3	No of leaves	0.081	0.079	**0.137**	0.064	0.071	0.006	0.080	-0.004	-0.065
4	Leaf length(cm)	0.125	0.106	0.084	**0.180**	0.074	-0.010	0.156	-0.018	-0.031
5	Leaf width(cm)	0.076	0.123	0.113	0.091	**0.220**	-0.022	0.177	0.054	-0.052
6	No of tillers	-0.001	0.001	0.000	0.001	0.001	**-0.010**	0.001	0.000	0.000
7	Leaf area(cm²)	-0.462	-0.514	-0.428	-0.637	-0.589	0.058	**-0.734**	-0.048	0.185
8	Length of leaf petiole(cm)	0.000	0.007	-0.002	-0.005	0.012	-0.002	0.003	**0.049**	0.004
9	No of mother rhizomes	0.001	0.001	0.002	0.001	0.001	0.000	0.001	0.000	**-0.004**
10	Length of mother rhizome (cm)	-0.046	-0.038	-0.012	-0.035	-0.032	-0.014	-0.039	0.001	0.011
11	Girth of mother rhizome (cm)	0.050	0.060	0.030	0.076	0.100	-0.029	0.102	0.006	0.028
12	No of primary rhizomes	0.124	0.107	0.070	0.081	0.102	0.029	0.106	0.059	-0.032
13	Length of primary rhizome (cm)	-0.023	-0.063	-0.019	-0.025	-0.093	0.047	-0.061	-0.137	-0.015
14	Girth of primary rhizome (cm)	0.037	0.028	0.023	0.025	0.022	-0.008	0.028	0.008	-0.011
15	No of secondary rhizomes	0.048	0.071	0.047	0.049	0.014	-0.024	0.044	-0.049	-0.060
16	Length of secondary rhizome(cm)	-0.001	-0.004	-0.004	-0.004	-0.005	0.003	-0.006	0.000	0.003
17	Girth of secondary rhizome (cm)	-0.069	-0.072	-0.067	-0.069	-0.088	0.033	-0.095	-0.017	0.047
18	Yield per plant (g)	0.002	0.000	0.001	0.001	-0.001	0.000	0.000	0.000	-0.001
19	Rhizome yield (tha^{-1})	-0.003	-0.001	-0.003	0.000	0.001	-0.001	0.001	0.000	0.002
20	Curing %	-0.019	-0.018	-0.010	-0.010	0.008	0.009	-0.002	0.008	0.009
21	**Yield /plant (g)**	**0.242**	**0.304**	**0.233**	**0.068**	**0.059**	**0.033**	**0.083**	**-0.032**	**-0.136**
	Partial R^2	0.023	0.113	0.032	0.012	0.013	0.000	-0.061	-0.002	0.001

Contd…

(Table 9 Cont...)

S.no	Character	Length of mother rhizome (cm)	Girth of mother rhizome (cm)	No of primary rhizomes	Length of primary rhizome (cm)	Girth of primary rhizome (cm)	No of secondary rhizomes	Length of secondary rhizome (cm)	Girth of secondary rhizome (cm)	Curing %	Curcumin content (%)	Days to maturity
1	Plant Height(cm)	0.030	0.018	0.043	0.007	0.049	0.025	0.008	0.025	-0.024	0.031	0.022
2	Basal Stem Diameter (cm)	0.092	0.084	0.142	0.077	0.144	0.138	0.088	0.099	-0.015	0.021	0.080
3	No of Leaves	0.011	0.016	0.034	0.008	0.044	0.034	0.029	0.034	-0.009	0.045	0.017
4	Leaf Length(cm)	0.041	0.052	0.052	0.015	0.062	0.047	0.042	0.046	-0.014	-0.007	0.022
5	Leaf Width(cm)	0.047	0.083	0.081	0.066	0.066	0.016	0.057	0.072	0.017	-0.016	-0.022
6	No of Tillers	-0.001	0.001	-0.001	0.002	0.001	0.001	0.002	0.001	0.000	-0.002	0.001
7	Leaf Area(cm^2)	-0.188	-0.284	-0.279	-0.145	-0.285	-0.169	-0.224	-0.257	0.007	0.044	-0.020
8	Length of Leaf Petiole(cm)	0.000	0.001	0.010	0.022	0.006	-0.013	0.000	0.003	-0.001	-0.002	-0.005
9	No of Mother Rhizomes	0.000	0.000	0.000	0.000	0.001	0.001	0.001	0.001	0.000	0.001	0.000
10	Length of Mother Rhizome (cm)	**-0.152**	-0.070	-0.048	-0.022	-0.028	-0.037	-0.025	0.021	0.024	-0.011	-0.035
11	Girth of Mother Rhizome (cm)	0.122	**0.264**	0.051	0.098	0.103	-0.006	0.128	0.097	-0.013	-0.029	-0.003
12	No of Primary Rhizomes	0.088	0.054	**0.278**	0.093	0.120	0.048	0.073	0.048	-0.041	0.069	0.016
13	Length of Primary Rhizome (cm)	-0.045	-0.114	-0.103	**-0.307**	-0.145	0.069	-0.059	-0.091	-0.027	0.079	0.061
14	Girth of Primary Rhizomes (cm)	0.014	0.028	0.031	0.034	**0.072**	0.013	0.012	0.028	-0.015	0.005	0.011
15	No of Secondary Rhizomes	0.046	-0.004	0.033	-0.043	0.034	**0.190**	0.010	-0.007	-0.031	0.036	0.051
16	Length of Secondary Rhizomes (cm)	-0.003	-0.009	-0.005	-0.004	-0.003	-0.001	**-0.018**	-0.009	0.001	0.002	0.002
17	Girth of Secondary Rhizomes (cm)	0.037	-0.100	-0.047	-0.080	-0.105	0.010	-0.137	**-0.271**	0.056	0.016	0.013
18	Curing %	0.001	0.000	0.001	-0.001	0.002	0.001	0.000	0.002	**-0.009**	0.003	0.002
19	Curcumin Content(%)	-0.001	0.001	-0.002	0.002	-0.001	-0.002	0.001	0.001	0.003	**-0.010**	-0.002
20	Days to maturity	-0.019	0.001	-0.005	0.016	-0.013	-0.022	0.010	0.004	0.021	-0.021	**-0.082**
21	**Yield per Plant (g)**	**0.122**	**0.023**	**0.268**	**-0.160**	**0.122**	**0.345**	**-0.003**	**-0.154**	**-0.071**	**0.252**	**0.130**
	Partial R^2	-0.018	0.006	0.074	0.049	0.009	0.065	0.000	0.042	0.001	-0.002	-0.011

R SQUARE = 0.3455 RESIDUAL EFFECT = 0.8090

Table 10: Distribution of Turmeric accessions in different clusters

Cluster No	Number of accessions	Name of the accessions
I	7	Avidi, Chintapalli local -1, Penekenametta, Paderu local, CLI-317, Imphal local, KTS-6.
II	45	KTS-8, GLpuram, Badipadar, Ochira, Vontimitta, Mydukur local, Dindigam, Ethamukkala, CL-7, GS, CL-12, CL-17, TCP-70, IC-521333, IC-420556, IC-330113, IC-211381, IC-212585, CLS-269, KTS-3,IISRPrathibha, AleppySupreme, Amritpani, Sugandham, Rajapuri, Wynad local, Chintapalli local-2, Vikici, CC-94-01, Lakadong, CL-2, Cuddapah local, CL-9, Tekuripeta, IC-540383, IC-211642, IC-211401, IC-353560, IC-211402, IC-319621.
III	1	KTS-7
IV	1	T.Sundar
V	20	Gorakhpur-361, Thodupuzha, Flourescent, Wagon, IC-420606, IC-420474, IC-319341, IC-211360,IC-332957, IC-033007, IC-540387, Tenali, IC-540386, Bataguda, Rajendra Sonia, IC-211641, IC-212606, IC-394396, IC-394268, IC-212578.
VI	3	BSR-2, IC-416941, CL-16
VII	1	IC-545139
VIII	3	Salem, Movatupuzha, Ernad chand
IX	1	CLL-335
X	1	Duggirala

Table :11 Average inter and intra – cluster (diagonal) distance D^2 and D values (parenthesis) in turmeric

	I Cluster	II Cluster	III Cluster	IV Cluster	V Cluster	VI Cluster	VII Cluster	VIII Cluster	IX Cluster	X Cluster
I Cluster	**20.16 (4.490)**	37.30 (6.108)	31.17 (5.583)	63.26 (7.954)	60.23 (7.761)	52.05 (7.214)	51.23 (7.157)	62.45 (7.903)	69.22 (8.320)	51.89 (7.200)
II Cluster		**31.41 (5.604)**	46.92 (6.850)	59.87 (7.738)	45.38 (6.737)	42.61 (6.530)	41.07 (6.410)	46.52 (6.820)	54.38 (7.374)	57.99 (7.610)
III Cluster			**0**	36.41 (6.034)	91.92 (9.588)	63.96 7.990)	75.35 (8.680)	82.43 (9.080)	74.52 (8.630)	77.81 (8.820)
IV Cluster				**0**	107.90 (10.380)	103.00 (10.150)	98.62 (9.930)	98.38 (9.910)	95.27 (9.760)	108.02 (10.390)
V Cluster					**32.296 (5.680)**	48.319 (6.950)	45.169 (6.720)	44.242 (6.650)	54.038 (7.350)	61.68 (7.850)
VI Cluster						**30.88 (5.560)**	42.357 (6.510)	53.146 (7.290)	55.39 (7.440)	66.38 (8.150)
VII Cluster							**0**	52.21 (7.230)	72.726 (8.530)	79.79 (8.930)
VIII Cluster								**33.03 (5.750)**	45.73 (6.760)	64.79 (8.050)
IX Cluster									**0**	49.88 (7.060)
X Cluster										**0**

Table 12 : The nearest and farthest clusters from each cluster based on D^2 values in turmeric germplasm.

Cluster No	Nearest cluster with D^2 values	Farthest cluster with D^2 values
I	III (31.17)	IX (69.22)
II	VII (37.3)	IV (59.87)
III	I (31.17)	V (91.92)
IV	III (36.41)	X (108.02)
V	VIII (44.24)	IV (107.95)
VI	VII (42.36)	IV (103)
VII	II (41.07)	IV (98.62)
VIII	V (44.24)	IV (98.38)

IX	VIII (45.73)	IV (95.27)
X	IX (49.88)	IV (108.02)

Table 13: Cluster means for yield and its components in turmeric

S.no	Cluster	Plant height (cm)	Basal stem diameter (cm)	No of leaves	Leaf length(cm)	Leaf width(cm)	No of tillers	Leaf area (cm²)	Length of leaf petiole(cm)	No of mother rhizomes	Length of mother rhizome (cm)	Girth of mother rhizome (cm)
1	Cluster I	157.35	11.41	8.33	67.12	16.94	1.68	821.82	16.23	2.05	5.28	12.78
2	Cluster II	160.98	11.67	8.53	69.59	16.65	2.06	832.13	12.06	2.42	5.99	9.49
3	Cluster III	164.54	11.39	7.9	56.36	15.83	1.86	647.37	17.17	2.39	9.04	11.29
4	Cluster IV	150.82	10.21	7.92	61.77	14.07	2.61	626.09	9.73	1.53	9.96	9.95
5	Cluster V	125.34	8.86	7.78	53.36	13.12	3.99	502.04	11.62	2.89	6.06	11.82
6	Cluster VI	168.12	12.63	9.16	70.85	16.85	1.80	861.64	8.62	1.67	8.09	11.84
7	Cluster VII	152.59	9.64	7.59	59.46	12.33	1.99	524.93	7.34	2.17	4.7	7.83
8	Cluster VIII	114.33	9.05	7.22	48.02	14.11	1.90	484.24	11.99	2.97	4.92	8.27
9	Cluster IX	145.20	10.33	7.05	64.74	14.59	1.86	680.50	8.44	3.59	7.44	13.48
10	Cluster X	109.44	9.92	7.02	54.35	14.26	1.36	562.14	10.19	2.21	6.26	11.63

Contd….

Table 13 contd…

S.no		No of primary rhizomes	Length of primary rhizome (cm)	Girth of primary rhizome (cm)	No of secondary rhizomes	Length of secondary rhizome (cm)	Girth of secondary rhizome (cm)	Yield per plant (g)	Curing %	Curcumin content (%)	Days to maturity
1	Cluster I	8.07	10.15	9.16	6.04	6.44	8.66	276.71	20.15	2.84	192.63
2	Cluster II	6.95	7.23	6.44	5.38	5.15	5.83	334.92	22.03	3.03	213.97
3	Cluster III	7.05	9.37	8.18	4.91	3.62	4.66	383.87	18.82	3.14	223.50
4	Cluster IV	7.54	7.81	6.08	7.01	5.18	4.05	321.48	23.46	3.72	212.97
5	Cluster V	4.20	7.89	6.65	1.95	3.69	4.29	287.64	23.19	3.24	210.28
6	Cluster VI	6.61	7.68	8.28	10.93	5.26	5.05	468.64	21.43	3.65	220.75
7	Cluster VII	4.60	6.62	7.10	7.45	3.09	5.14	361.92	17.06	3.70	220.00
8	Cluster VIII	4.25	7.29	5.99	5.10	2.09	3.34	377.95	26.56	2.69	204.80
9	Cluster IX	5.63	8.74	7.25	5.29	5.65	5.54	315.92	25.21	2.06	207.70
10	Cluster X	3.20	7.63	6.68	4.76	7.16	6.58	282.00	22.33	2.08	216.71

Table 14: Contribution of characters towards genetic divergence.

S.no	Source	Number of times ranked 1st	Percent contributi on
1	Plant height(cm)	22	0.65
2	Basal stem diameter (cm)	0	0
3	No of leaves	0	0
4	Leaf length(cm)	0	0
5	Leaf width(cm)	0	0
6	No of tillers	0	0
7	Leaf area(cm^2)	1238	36.37
8	Length of leaf petiole(cm)	0	0
9	No of mother rhizomes	0	0
10	Length of mother rhizome(cm)	0	0
11	Girth of mother rhizome (cm)	0	0
12	No of primary rhizomes	0	0
13	Length of primary rhizome(cm)	0	0
14	Girth of primary rhizome(cm)	0	0
15	No of secondary rhizomes	4	0.11
16	Length of secondary rhizome (cm)	0	0
17	Girth of secondary rhizome (cm)	0	0
18	Yield per plant (g)	2128	62.53
19	Curing %	0	0
20	Curcumin content %	0	0
21	Days to maturity	11	0.32

Table 15: Eigen values, per cent variability and cumulative variability for principal components of morphological characters in turmeric.

S.no	Principal component	Eigen value (Root)	Per cent variation explained	Cumulative variation explained
1	1 Vector	5.61	26.714	26.714
2	2 Vector	2.699	12.852	39.566
3	3 Vector	1.632	7.77	47.335
4	4 Vector	1.475	7.025	54.36
5	5 Vector	1.374	6.544	60.904
6	6 Vector	1.18	5.621	66.525
7	7 Vector	1.112	5.294	71.819
8	8 Vector	0.849	4.043	75.863

Table :16 Character loading of principal components for morphological characters in turmeric

S.no	Character	1 Vector	2 Vector	3 Vector	4 Vector	5 Vector	6 Vector	7 Vector	8 Vector
1	Plant height(cm)	0.326	0.174	0.047	0.108	0.020	0.093	0.296	0.135
2	Basal stem diameter (cm)	0.339	0.058	-0.117	0.105	-0.067	-0.225	-0.033	-0.055
3	No of leaves	0.293	0.150	-0.210	0.171	0.191	0.068	-0.043	-0.036
4	Leaf length(cm)	0.315	0.026	-0.229	-0.054	-0.169	0.051	0.359	0.056
5	Leaf width(cm)	0.296	-0.178	-0.177	0.199	-0.071	-0.032	-0.084	-0.300
6	No of tillers	-0.037	0.123	0.082	0.356	-0.076	0.664	0.088	0.115
7	Leaf area(cm^2)	0.366	-0.072	-0.253	0.066	-0.139	0.018	0.173	-0.125
8	Length of leaf petiole(cm)	0.044	-0.242	0.287	0.429	0.120	-0.293	-0.033	-0.424
9	No of mother rhizomes	-0.168	-0.200	0.207	-0.030	-0.309	-0.011	0.298	0.098
10	Length of mother rhizome(cm)	0.168	0.053	0.322	-0.148	-0.499	0.176	-0.073	-0.275
11	Girth of mother rhizome (cm)	0.204	-0.266	0.182	-0.327	-0.229	0.218	-0.085	-0.037
12	No of primary rhizomes	0.246	-0.001	0.335	0.208	-0.025	0.088	-0.197	0.139
13	Length of primary rhizome(cm)	0.136	-0.411	0.263	0.152	0.036	-0.179	-0.032	0.231
14	Girth of primary rhizome(cm)	0.266	-0.081	0.282	-0.056	0.135	-0.136	0.128	0.526
15	No of secondary rhizomes	0.151	0.329	-0.069	-0.226	-0.135	-0.229	-0.244	0.150
16	Length of secondary rhizome(cm)	0.178	-0.218	-0.058	-0.353	0.131	0.241	-0.421	-0.147
17	Girth of secondary rhizome (cm)	0.196	-0.234	-0.044	-0.249	0.478	0.135	0.041	0.081
18	Yield /plant (g)	0.107	0.276	0.111	0.120	-0.153	-0.131	-0.464	0.224
19	Curing %	-0.080	-0.177	-0.367	0.252	-0.320	-0.059	-0.210	0.293
20	Curcumin content %	0.064	0.369	0.252	0.128	0.278	0.174	-0.074	-0.140
21	Days to maturity	0.076	0.298	0.206	-0.242	-0.037	-0.299	0.271	-0.183

Table : 17 DNA yield, absorbance ratio and quantity of different accessions of turmeric

S.No	Accession No.	260/280 ratio	Quantity (ng/µl)
1	Wagon	1.85	600
2	Amritpani	1.87	330
3	Penakanametta	1.86	540
4	Gorakhpur-361	1.89	1020
5	Mydukur	1.91	1320
6	Sugandham	1.86	630
7	Ochira	1.86	570
8	Vontimitta	1.81	290
9	CLL-335	1.82	870
10	Salem	1.82	690
11	Duggirala	1.87	1380
12	Dindigam	1.84	780
13	TCP-70	1.73	930
14	Cuddapah local	1.94	1140
15	Paderu local	1.98	780
16	Etahmukkala	1.83	1050
17	IC- 212585	1.86	1110
18	IC-540386	1.82	1110
19	IC-319621	1.84	660
20	IC-211402	1.86	1290
21	IC-211360	1.86	1470
22	IC-394396	1.81	270
23	IC-181919	1.80	390

24	IC-571333	1.83	1800
25	IC-332957	1.99	1470
26	IC-212267	1.84	1200
27	IC-353560	1.79	2220
28	IC-420474	1.79	420
29	IC-211641	1.82	1230
30	IC-416941	1.78	1320
31	IC-540383	1.81	720
32	IC_420556	1.83	870
33	IC-319341	1.81	2070
34	Megha turmeric	1.86	2100
35	CL -12	1.85	1980
36	IC -420606	1.85	2100

Contd…

(Table 17 Contd…)

S.No	Accession No.	260/280 ratio	Quantity (ng/µl)
37	IC-330113	1.81	1620
38	IC -212605	1.82	3270
39	IC -394268	1.8	1470
40	IC-540387	1.82	990
41	IC 212606	1.91	1380
42	CL -16	1.81	1350
43	IC 211401	1.85	1470
44	IC-394903	1.81	2010
45	IC- 033007	1.79	2040
46	IC-211381	1.93	1860

47	Tenali	1.79	1350
48	IC-212578	1.85	1740
49	CL-2	1.82	2400
50	IC -545139	1.92	2130
51	IC-211642	1.88	1770
52	GS	1.89	2220
53	Tekuripeta	1.8	2340
54	Imphal local	1.84	2100
55	BSR-2	1.83	2910
56	Chintapalli local 1	1.84	2130
57	Vikici	1.86	1950
58	Florascent	1.8	1620
59	Movatupuzha	1.79	2280
60	Avidi	1.922	1740
61	Thodupuzha	1.81	2190
62	KTS-8	1.993	2490
63	Bataguda	1.879	1590
64	KTS-6	1.811	2790
65	GL puram	1.887	1350
66	CL-17	1.81	2700
67	Ernad chand	1.894	1860
68	Badipadar	1.865	3270
69	CLI-317	1.8	1770
70	Chintapalli local 2	1.974	1950
71	Wynad local	1.808	3030
72	CC-94-01	1.79	3000
73	KTS-7	1.791	2280
74	Aleppy supreme	1.887	1500

Contd…

(Table 17 cont …)

S.No	Accession No.	260/280 ratio	Quantity (ng/µl)
75	Rajendra sonia	1.799	2040
76	T Sundar	1.837	2400
77	Lacadong	1.853	3240
78	Rajapuri	1.828	1800
79	CL-7	1.823	1500
80	Cl-9	1.803	2190
81	CLS 269	1.91	390
82	KTS-3	1.83	1140
83	IISR-Prathibha	1.86	900

Table :18 Allele heterozygosty and polymorphic information content of the RAPD markers used in the analysis of turmeric genotypes.

S.no	Primer	Allele heterozygosity	PIC
1	OPA4	0.828	0.803
2	OPA5	0.842	0.822
3	OPA 7	0.794	0.761
4	OPA 8	0.827	0.805
5	OPA 10	0.867	0.853
6	OPB 9	0.718	0.667
7	OPC 13	0.745	0.704
8	OPC 16	0.576	0.483
9	OPC 20	0.653	0.58
10	OPD 1	0.632	0.56
11	OPD 3	0.671	0.616
12	OPE 9	0.742	0.695
13	OPF 13	0.723	0.671
14	OPG13	0.825	0.8
15	OPG 20	0.666	0.592
16	OPJ10	0.748	0.7
17	OPJ 18	0.665	0.591
18	OPR 7	0.797	0.764
19	OPS 12	0.65	0.575
20	OPT 8	0.715	0.662
21	OPT20	0.667	0.592

Table: 19 Description of primers and PCR amplicons generated from RAPD analysis of the turmeric accessions.

Primer	Range of amplicons (bp)	No of loci	No of polymorphic loci	% Polymorphism
OPA 4	200-1500	4	3	75
OPA 5	200-1200	7	7	100
OPA 7	250-950	7	7	100
OPA 8	2250-1200	7	6	85.71
OPA 10	200-1150	9	9	100
OPB 9	750-1100	4	4	100
OPC 13	200-750	5	5	100
OPC 16	500-850	3	3	100
OPC 20	350-600	3	3	100
OPD 1	270-750	4	4	100
OPD 3	300-800	4	4	100
OPE 9	260-550	4	4	100
OPF 13	270-700	4	4	100
OPG 13	200-800	5	5	100
OPG 20	300-700	4	4	100
OPJ 10	200-650	4	4	100
OPJ 18	270-750	3	3	100
OPR 7	190-920	5	5	100
OPS 12	250-320	3	3	100
OPT 8	270-750	4	4	100
OPT 20	450-1000	3	3	100
	Mean	**4.57**		

Table: 21 Distribution of genotypes into different clusters based on RAPD analysis

S.No	Cluster No.	Number of Genotypes	Genotypes included in cluster
1	IA	26	Wagon, Penekenametta, CLS-269, KTS-3, IC-211360, BSR-2, Thodupuzha, Florescent, Gorakhpur-361, Ethamukkala, CL-16, Ernad Chand, IC-212578, IC-211642, Sugandham, IC-319621, IC--211402, Avidi, IC-571333, IC-420556, IC-211267, IC-033007, Megha turmeric, IC-330113, CL-12 and IC-212605.
2	IB	7	TCP -70, Paderu local, Chintapalli local-1,Tenali Chintapalli local 2,Wynad local and Lacadong.
3	IC	3	Rajendra Sonia, CL-7 and T.Sundar
4	ID	5	IC-211641, IC-540383, IC-540387, IC-211401, and IC-211381
5	IIA	21	Amrutapani, Ochira, Mydukur, Salem, Duggirala,Cuddapah local, Dindigam, CL-2, GS, Tekuripeta, Bataguda, KTS-8, IC-545139, Movatupuzha, KTS-6, CL-17, Imphal local, GL Puram, Badipadar, CLI 317 and Vikici.
6	IIB	11	IISR Prathibha, IC-394903, IC-420474, IC-394396, IC-181919, IC-332957, IC-416941, IC-394268, IC-212606, IC-319341 and IC-420606.

7	IIC	1	IC-353560
8	IID	2	IC-540386, IC-319621
9	IIE	1	Vontimitta
10	IIF	3	CC- 9401, KTS-7 and Aleppy Supreme
11	IIG	1	Rajapuri
12	IIH	1	CL-9
13	III	1	CLL-335

Table: 22 Principal component analysis on the contribution of RAPD markers among the turmeric accessions

S.no	Accession	Eigen value	Per cent variation	Cumulative variation
1	Wagon	27.499	33.131	33.131
2	Amritpani	10.729	12.927	46.058
3	Penakanametta	4.682	5.641	51.698
4	Gorakhpur-361	2.222	2.677	67.154
5	Mydukur	1.971	2.375	69.529
6	Sugandham	1.776	2.140	71.669
7	Ochira	1.717	2.069	73.738
8	Vontimitta	1.571	1.893	75.630
9	CLL-335	1.377	1.659	77.289
10	Salem	1.299	1.565	78.854

11	Duggirala	1.241	1.496	80.349
12	Dindigam	1.192	1.436	81.785
13	TCP-70	1.106	1.333	83.118
14	Cuddapah local	1.081	1.303	84.421
15	Paderu local	0.979	1.180	85.601
16	Etahmukkala	0.954	1.149	86.750
17	IC- 212585	0.840	1.013	87.762
18	IC-540386	0.778	0.937	88.700
19	IC-319621	0.742	0.894	89.593
20	IC-211402	0.692	0.834	90.427
21	IC-211360	0.674	0.812	91.240
22	IC-394396	0.635	0.766	92.005
23	IC-181919	0.598	0.721	92.726
24	IC-571333	0.569	0.686	93.412
25	IC-332957	0.527	0.635	94.048
26	IC-212267	0.514	0.620	94.667
27	IC-353560	0.451	0.544	95.211
28	IC-420474	0.419	0.504	95.715
29	IC-211641	0.399	0.481	96.197
30	IC-416941	0.373	0.449	96.646
31	IC-540383	0.358	0.431	97.077
32	IC_420556	0.304	0.366	97.443

Table 21 contd…

Table 22 contd…

S.no	Accession	Eigen value	Per cent variation	Cumulative variation
33	IC-319341	0.273	0.329	97.773
34	Megha turmeric	0.263	0.317	98.090
35	CL -12	0.257	0.310	98.400
36	IC 420606	0.237	0.286	98.686
37	IC-330113	0.228	0.275	98.960
38	IC -212605	0.206	0.248	99.208
39	IC 394268	0.199	0.240	99.448
40	IC-540387	0.190	0.229	99.677
41	IC 212606	0.181	0.218	99.895
42	CL -16	0.165	0.198	> 100%
43	IC 211401	0.160	0.192	> 100%
44	IC-394903	0.141	0.169	> 100%
45	IC 033007	0.112	0.135	> 100%
46	IC-211381	0.102	0.123	> 100%
47	Tenali	0.088	0.106	> 100%
48	IC-212578	0.081	0.098	> 100%
49	CL-2	0.072	0.086	> 100%
50	IC -545139	0.065	0.078	> 100%
51	IC-211642	0.058	0.070	> 100%
52	GS	0.047	0.056	> 100%
53	Tekuripeta	0.043	0.052	> 100%
54	Imphal local	0.034	0.042	> 100%
55	BSR-2	0.029	0.035	> 100%

56	Chintapalli local 1	0.026	0.031	> 100%
57	Vikici	0.018	0.022	> 100%
58	Florascent	0.011	0.013	> 100%
59	Movatupuzha	0.007	0.008	> 100%
60	Avidi	0.006	0.008	> 100%
61	Thodupuzha	-0.003	-0.004	> 100%
62	KTS-8	-0.005	-0.006	> 100%
63	Bataguda	-0.008	-0.010	> 100%
64	KTS-6	-0.009	-0.011	> 100%

Contd…

Table 22 Contd…

S.no	**Accession**	**Eigen value**	**Per cent variation**	**Cumulative variation**
65	GL Puram	-0.014	-0.017	> 100%
66	CL-17	-0.019	-0.023	> 100%
67	Ernad chand	-0.023	-0.028	> 100%
68	Badipadar	-0.025	-0.030	> 100%
69	CLI-317	-0.030	-0.037	> 100%
70	Chintapalli local -2	-0.040	-0.048	> 100%
71	Wynad local	-0.047	-0.057	> 100%
72	CC-94-01	-0.048	-0.058	> 100%
73	KTS-7	-0.058	-0.070	> 100%
74	Aleppy spreme	-0.065	-0.078	> 100%

75	Rajendra sonia	-0.078	-0.094	> 100%
76	T Sundar	-0.088	-0.106	> 100%
77	Lacadong	-0.103	-0.125	> 100%
78	Rajapuri	-0.134	-0.162	> 100%
79	CL-7	-0.140	-0.169	> 100%
80	Cl-9	-0.238	-0.287	> 100%
71	CLS 269	4.528	5.456	57.154
82	KTS-3	3.479	4.192	61.346
83	IISR-Prathibha	2.600	3.132	64.478

Table: 23 Description of primers and PCR amplicons generated from SSR analysis of the turmeric accessions.

S.no	Primer	Allele size(bp)	Total no of Alleles	No of polymorphic Alleles	% polymorphism
1	CuMiSat-3	180-440	3	2	100
2	CuMiSat-5	160-190	2	2	100
3	CuMiSat-8	150-180	2	2	100
4	CuMiSat-11	150-200	3	2	100
5	CuMiSat-16	160-180	2	2	100
6	CuMiSat-17	220	1	0	0
7	CuMiSat-18	200	1	0	0
8	CuMiSat-19	150-200	2	2	100
9	CuMiSat-20	175-200	2	2	100
10	CuMiSat-21	110-140	2	2	100
11	CuMiSat-22	115-140	2	1	50
12	CuMiSat-26	100-175	2	1	50
13	CuMiSat-27	100-110	2	1	50
14	CuMiSat-28	130 -230	3	2	100
15	CuMiSat-29	130-150	2	2	100
16	CuMiSat-30	160-180	2	2	100
17	CuMiSat-35	120-150	2	2	100
18	CuMiSat-36	110 -150	2	2	100
19	CuMiSat-37	170-200	2	0	100

Table: 24 Allele Frequency and polymorphic information content of the SSR markers used in the analysis of turmeric genotypes

S.no	SSR primer	Major allele frequency	Polymorphic information content
1	CuMiSat-03	0.337	0.628
2	CuMiSat-05	0.494	0.455
3	CuMiSat-08	0.337	0.531
4	CuMiSat-11	0.333	0.498
5	CuMiSat-16	0.500	0.311
6	CuMiSat-17	0.476	0.387
7	CuMiSat-18	1.000	0
8	CuMiSat-19	0.494	0.498
9	CuMiSat-20	0.500	0.474
10	CuMiSat-21	0.476	0.509
11	CuMiSat-22	0.506	0.260
12	CuMiSat-26	0.482	0.441
13	CuMiSat-27	0.494	0.509
14	CuMiSat-28	0.333	0.368
15	CuMiSat-29	0.488	0.492
16	CuMiSat-30	0.452	0.522
17	CuMiSat-35	0.452	0.553
18	CuMiSat-36	0.500	0.212
19	CuMiSat-37	0.500	0.488

Table : 26 Distribution of genotypes into different clusters based on SSR analysis

S.No	Cluster no.	Number of genotypes	Genotypes included in the cluster
1	IA	11	Wagon, Penakenametta, CLS-269, Rajendra sonia, KTS-3, Gorkhpur-361, CLL-335, Ethamukkala, IC -212585, BSR-2 and Thodupuzha.
2	IB	13	IC -211360, IC-211381, IC-211642, IC-212267, IC-540383, IC-211641, IC-540387, IC-211401, IC-212578, CL-12, CL-16, CL-7 and Flourescent
3	IC	6	TCP-70, Paderulocal, Tenali, Chintapalli local 2, Wynad local and Lacadong.
4	II A	5	Amritpani, Mydukur, IISR Prathibha, IC-353560 and IC-212605
5	II B	40	Ocihra, Vontimitta, Dindigam, Salem, Duggirala, IC-349396, IC-319341, IC-420606, Rajapuri, CL-2, Tekuripeta, IC-545139, CL-9, IC-332957, Imphal Local, IC-416941, IC-212606, Bataguda, Vikici, CC-9401, KTS-8, Aleppy Supreme, CL-17, Badipadar, Movatupuzha, GL-puram, KTS-7, IC-181919, IC-420474, IC-394268, GS, IC -394903, KTS-6, CLI-317, Cuddapah local, IC -212585, IC -540386, Megha turmeric, T Sundar and Avidi
6	II C	4	Sugandham, IC-319621, IC-211402 and IC-571333
7	II D	2	IC-420556 and IC-330113
8	II E	1	IC-033007
9	III	1	Chintapalli local 1

Table :27 Principal component analysis on the contribution of SSR markers among turmeric accessions

S.no	Accession	Eigen value	Per cent variation explained	Cumulative variation
1	Wagon	32.16672	38.7551	38.7551
2	Amritpani	13.99085	16.8564	55.6115
3	Penakanametta	6.742336	8.1233	63.7348
4	Gorakhpur-361	3.44251	4.1476	83.9421
5	Mydukur	2.894378	3.4872	87.4293
6	Sugandham	2.409637	2.9032	90.3325
7	Ochira	1.780943	2.1457	92.4782
8	Vontimitta	1.633832	1.9685	94.4467
9	CLL335	1.232581	1.485	95.9317
10	Salem	0.985214	1.187	97.1187
11	Duggirala	0.795521	0.9585	98.0772
12	Dindigam	0.586159	0.7062	98.7834
13	TCP-70	0.486696	0.5864	99.3698
14	Cuddapah local	0.423041	0.5097	99.8795
15	Paderu local	0.290978	0.3506	> 100%
16	Etahmukkala	0.267321	0.3221	> 100%
17	IC- 212585	0.178008	0.2145	> 100%
18	IC-540386	0.114933	0.1385	> 100%
19	IC-319621	0.113928	0.1373	> 100%
20	IC-211402	0.067468	0.0813	> 100%
21	IC-211360	0.038701	0.0466	> 100%
22	IC-394396	0.025314	0.0305	> 100%
23	IC-181919	0.008716	0.0105	> 100%

24	IC-571333	0.003571	0.0043	> 100%
25	IC-332957	0.001393	0.0017	> 100%
26	IC-212267	0.000978	0.0012	> 100%
27	IC-353560	0.000479	0.0006	> 100%
28	IC-420474	0.000313	0.0004	> 100%
29	IC-211641	0.000236	0.0003	> 100%
30	IC-416941	0.000195	0.0002	> 100%
31	IC-540383	0.00011	0.0001	> 100%
32	IC_420556	0	0.0001	> 100%
33	IC-319341	0	0.0001	> 100%
34	Megha turmeric	0	0.0001	> 100%
35	CL 12	0	0	> 100%
36	IC 420606	0	0	> 100%
37	IC-330113	0	0	> 100%

Contd…

(table 27 cont…)

S.no	**Accession**	**Eigen value**	**Per cent variation explained**	**Cumulative variation**
38	IC -212605	0	0	> 100%
39	IC 394268	0	0	> 100%
40	IC-540387	0	0	> 100%
41	IC 212606	0	0	> 100%
42	CL -16	0	0	> 100%
43	IC 211401	0	0	> 100%
44	IC-394903	0	0	> 100%
45	IC 033007	0	0	> 100%
46	IC-211381	0	0	> 100%

47	Tenali	0	0	> 100%
48	IC-212578	0	0	> 100%
49	CL-2	0	0	> 100%
50	IC -545139	0	0	> 100%
51	IC-211642	0	0	> 100%
52	GS	0	0	> 100%
53	Tekuripeta	0	0	> 100%
54	Imphal local	0	0	> 100%
55	BSR-2	0	0	> 100%
56	Chintapalli local 1	0	0	> 100%
57	Vikici	0	0	> 100%
58	Flourascent	0	0	> 100%
59	Movatupuzha	0	0	> 100%
60	Avidi	-0.00008676	-0.0001	> 100%
61	Thodupuzha	-0.00048832	-0.0006	> 100%
62	KTS-8	-0.00377429	-0.0045	> 100%
63	Bataguda	-0.00501424	-0.006	> 100%
64	KTS-6	-0.00518507	-0.0062	> 100%
65	GL Puram	0.00616175	-0.0074	> 100%
66	CL-17	-0.01215094	-0.0146	> 100%
67	Ernad chand	-0.01524635	-0.0184	> 100%
68	Badipadar	-0.01668873	-0.0201	> 100%
69	CLI317	-0.02145958	-0.0259	> 100%
70	Chintapalli local 2	-0.02395133	-0.0289	> 100%
71	Wynad local	-0.03125393	-0.0377	> 100%
72	CC-94-01	-0.03681592	-0.0444	> 100%
73	KTS-7	-0.03999194	-0.0482	> 100%
74	Aleppy spreme	-0.04264099	-0.0514	> 100%

75	Rajendra sonia	-0.05503744	-0.0663	> 100%

Contd…

(table 27 cont ….)

S.no	**Accession**	**Eigen value**	**Per cent variation explained**	**Cumulative variation**
76	T Sundar	-0.06356425	-0.0766	> 100%
77	Lacadong	-0.07980993	-0.0962	> 100%
78	Rajapuri	-0.112329	-0.1353	> 100%
79	CL-7	-0.19605793	-0.2362	> 100%
80	Cl-9	-0.24514039	-0.2953	100%
81	CLS 269	5.301526	6.3874	70.1222
82	KTS-3	30651136	5.1886	75.3108
83	IISR-Prathibha	72148341	4.4837	79.7945

CHAPTER-V

SUMMARY AND CONCLUSIONS

An experiment entitled **"Studies on genetic diversity in turmeric (*Curcuma longa* L.) using morphological and molecular markers"** was undertaken during 2013 -2015 at Horticultural Research Station, Kovvur, West Godavari District, Andhra Pradesh, to document the information on qualitative characters, to assess the extent of variability on growth and yield components and their association, to identify the principal components contributing towards genetic divergence and to assess the extent of genetic diversity of 83 turmeric accessions using D^2 analysis, RAPD and SSR markers.

The observations were recorded on 21quantitative and 6 qualitative characters. Twenty one RAPD and nineteen SSR markers were used to study the genetic diversity in turmeric. The salient findings of the present investigation are summarized here under

5.1 STUDIES ON GENETIC DIVERSITY IN TURMERIC USING MORPHOLOGICAL MARKERS

- The evaluation of turmeric genotypes revealed significant differences with respect to growth and yield related characters.
- Among the 83 accessions of turmeric studied including check varieties, the accession, Ernad Chand recorded tallest plants while the accession, T Sundar recorded the highest basal stem diameter, leaf length and leaf area. The accession, CLL-335 recorded more number of leaves, leaf width, number of primary rhizomes and rhizome yield per hectare while the accession, Ochira recorded more number of secondary rhizomes per plant.
- Among the eighty accessions, thirty nine were short duration types (180-210 days), thirty two were of medium duration (210-240 days) and nine

accessions belong to long duration types. The accession Mydukur has taken more number of days (247.5 days), whereas the accession IC-420474 has taken less number of days (186.17days) for maturity, whereas among the checks CLS-269 was having short duration and IISR Prathibha and KTS-3 were of long duration types.

- Highest curing percentage was obtained with the accession IC- 319621 (31.46%). The accession T Sundar possessed high curcumin (6.73%) content followed by Ochira (6.58%). The inner core colour of the rhizomes observed among the eighty three accessions varied from light yellow to dark orange yellow. Among all the accessions, seventeen accessions (20.48%) possessed light yellow coloured rhizomes, nineteen accessions (22.89%) were having lemon yellow coloured rhizomes , thirty three accessions (39.76)% exhibited light orange yellow coloured rhizomes and fourteen accessions (6.87%) have produced dark orange yellow coloured rhizomes.
- The highest percentage of phenotypic and genotypic coefficient of variation was observed for number of secondary rhizomes, followed by length of leaf petiole and yield per plant. High heritability was observed for curcumin content (99.9%), curing percentage (98.5%), yield (81.6%), plant height (95.4%), basal stem diameter (91.2%), number of leaves (87.2%), leaf length (92.4%), leaf width (97.6%), leaf area (93.3%), length of leaf petiole (79.1%), girth of mother rhizome (72.5%) length of primary rhizome (69.1%) and number of secondary rhizomes (85.5%) indicating that these characters are important in crop improvement. Highest estimates of heritability observed for curcumin content (99.09) could be utilized for the improvement of fresh turmeric yield through clonal selection.
- The rhizome yield per plant exerted a significant and positive correlation both at phenotypic and genotypic levels for all the characters studied and is highly and significantly correlated with number of primary rhizomes, plant height ,basal stem diameter, number of leaves, leaf length, leaf width, leaf area, number of tillers per plant, length of mother rhizome,

length of primary rhizome, girth of primary rhizome, number of secondary rhizomes, length of secondary rhizome, girth of secondary rhizome, curcumin content and days to maturity.

- Path coefficient analysis projected basal diameter, number of primary rhizomes, girth of mother rhizome, leaf width and number of secondary rhizomes per plant as major contributors towards yield due to high positive effects.
- The D^2 statistic for 21 quantitative traits revealed significant contribution of the characters towards variability. The accessions were grouped into 10 clusters and the highest inter cluster value observed between cluster IV and X (108.2) and cluster IV and V (107.95),which can be expected to exert high heterotic effect in the hybrids when crossed and consequently may generate desirable segregates. Thus, broad spectrum of variability can be created in the ensuing generation which may be helpful in future selections and crop improvement programmes.
- Among the ten clusters, cluster VI registered higher mean values for plant height, basal stem diameter, number of leaves, leaf length, leaf area, and yield per plant, whereas, cluster IV projected for high curcumin content. cluster I was representing short duration types with low curcumin content and were from Andhra Pradesh, except Imphal local.

- Genotypes from different locations were grouped in cluster II pointing out that geographical diversity did not have significant impact on genetic diversity.
- Among the characters that contributed towards genetic divergence, yield recorded highest rank, emphasizing the importance of that character towards divergence.
- The PCA executed for 83 accessions has revealed that, among all the principal components, PC I scored high loadings for plant height, basal stem diameter, number of leaves, leaf width and leaf area, PC II accorded much of its variance through number of secondary rhizomes, yield per plant, curcumin content and days to maturity, whereas, PC III attributed

its variance through length of mother rhizome, number of primary rhizomes, length of primary rhizome and girth of primary rhizome. PC IV relatively contributed high variance for length of leaf petiole and curing percentage. PC V exhibited highest per cent of variance for girth of secondary rhizomes, whereas, PC VI attributed its variance through number of tillers, girth of mother rhizome and length of secondary rhizome . PC VII projected to score high variance for leaf length and number of mother rhizomes.

5.2 STUDIES ON GENETIC DIVERSITY IN TURMERIC USING MOLECULAR MARKERS

- The genomic DNA was extracted from all the 83 accessions and the DNA yields ranged from 270 to 3270 ηg/µl. The purity of DNA was determined by A_{260}:A_{280} ratio which varied from 1.73 to 1.99 indicating that the DNA was relatively of high purity and was suitable for PCR amplification.
- A total of twenty one RAPD primers were used to amplify the gene loci in the genomes of different turmeric accessions. From the electrophoregrams of the PCR amplicons, the random primers produced the amplification products with sizes ranging from 190 bp to 1500 bp. All the primers showed the PIC value more than 0.5 except the RAPD primer OPC 16 (0.483).
- RAPD analysis indicated that among all the accessions CLS 269 showed maximum similarity with Penekanametta with a similarity coefficient of 0.98 where as Rajapuri showed least similarity with CLL 335 (0.366). However, CLL 335 was found to be distant from all the other 82 accessions.
- Genetic distances between 83 turmeric accessions were estimated using both morphological and molecular markers (RAPD and SSRs). These methods were able to distinguish all the accessions used in the study. Both morphological and molecular characterization methods were found efficient in grouping the accessions. However, molecular analysis grouped the accession more accurately. The pattern of sub clustering of

turmeric accessions in a cluster varied over different marker systems. But SSR's were able to differentiate effectively. However, most of the accessions were grouped together by SSRs and RAPD markers whereas widely differed from morphological characterization, as morphological markers obviously can detect cultivar variation only at phenotypic level.

- Accessions from same region were grouped into different clusters and accessions that were collected from different regions, were found in same clusters, which was attributed to the absence of influence of location specificity. Among the collected genotypes, chance of migration of rhizome seed material by growers from one region to another may also be the reason for the observed clustering of different turmeric accessions.
- In the present study, synonymous entities with genetic similarity coefficient value of 1.0 were identified by the SSR markers, which is utmost important for precise assessment of cultivar type and genetic diversity in turmeric. Among the 83 accessions studied, nine sets of synonymous entities were identified.(Wagon and Penekenametta,),(Vontimitta with Ochira, Salem, Dindigam and Duggirala), (IC- 319621 and IC-211402,IC 181919), (IC-420474, IC-319341 and IC-394396), (KTS-3, IC-394903 and IC-319621), (Rajapuri and IC-420606), (CL-2 and Tekuripeta), (KTS -8 and CL-17) and (KTS-7 and GL puram)}.
- Both RAPD and SSR markers are useful in the assessment of turmeric diversity, detection of duplicates in germplasm and selection of a core collection to enhance the efficiency of genotype management for use in turmeric breeding and conservation. Even though turmeric is a vegetatively propagated and highly heterozygous crop, hybridization between some divergent genotypes could broaden the genetic base for the development of elite genotypes.
- Identification of markers associated with specific traits of great interest, which could be converted into SCAR markers needs attention to speed up the crop improvement programme. The degree of genetic variability evaluated through molecular characterization can form the basis for future

breeding programme and a finer molecular analysis with additional SSR loci and use of other molecular markers like AFLP and ISSRs is required to differentiate point mutations involved in identical accessions and to delineate some geographical associations among the accessions.

FUTURE LINE OF WORK

- Development of genetic maps for identifying QTLs (Quantitative Trait Loci) in turmeric will greatly facilitate the application of MAS (Marker Assisted Selection) for vigorous and rapid crop improvement programmes.

- The unique polymorphic fragments of RAPD's can be converted as sequence tagged sites (STS) markers that could be exploited for DNA finger printing of the cultivars which are useful in detecting the mixtures in the cultivars especially in clonally propagated crops like turmeric.

- Identification and characterization of the key enzymes involved in the biosynthesis of medicinally important molecules of turmeric.

- Genetic transformation studies using somatic hybridization protoplast fusion and polyploid breeding to get good transgenics, for high yield coupled with high curcumin content in turmeric

Appendix -I Monthly weather data recorded at Horticultural Research Station Kovvur during the period April, 2013 to March, 2014.

Month and Year	Temperature (^{o} C)		Relative Humidity (%)		Rainfall (mm)	Rainy days
	Maximum	Minimum	8.00 hrs	14.00 hrs		
April,2013	35.8	24.8	78.2	50.4	22.2	3
May,2013	39.45	26.87	72.93	41.38	9	2
June,2013	33.83	25.83	74.76	*74.76*	167	12
July,2013	29.77	24.54	75.67	65.29	107.2	18
Aug,2013	31.74	25.93	73.22	58.93	139.9	11
Sept,2013	32.4	25.0	73.0	61.0	219.3	10
Oct, 2013	30.83	24.51	78.77	67.96	341	10
Nov,2013	30.6	20.9	70	57.4	31.7	3
Dec,2013	30.1	18.61	63.03	48.45	0	0
Jan, 2014	29.93	18.70	74.29	51.03	0	0
Feb,2014	31.07	19.64	65.96	52.25	0	0
Mar,2014	33.67	22.0	69.96	48.33	0	0
Total	32.43	23.11	72.48	56.43	**1037.3**	**69**
Mean	**32.43**	**23.11**	**72.48**	**56.43**		

Appendix II. Preparation of stock solutions

1. CTAB 20%	Dissolve 20 g Cetyl trimethyl ammonium bromide in some sterile distilled water the final volume made up to 100 mlwith sterile distilled water.
2. PVP 10%	Dissolve 10g PVP in sterile distilled water and final volume made up to 100 ml
3. NaCl 5 M	Dissolve 29.22g of NaCl in sterile distilled water and final volume made up to 100 ml (Mol Wt of NaCl = 58.44)
4. EDTA 0.5M, 8.0 pH)	Dissolve 18.61g of EDTA in sterile distilled water and final volume made up to 100 ml (Mol Wt of EDTA = 372.2). EDTA dissolves at pH 8.0. Autoclave it.
5. Tris HCl 1M, (p^H 8.0)	Dissolve 12.1g Tris in 80 ml of distilled water, adjust pH to 8.0 with conc. HCl. Make up final volume to 100 ml and autoclave (Mol Wt of Tris = 121.1)
6. Chloroform : isoamyl alcohol (24:1)	Mix 240 ml chloroform and 10 ml isoamyl alcohol. Store in dark at room temperature.
7. Ethenol (70 %)	Absolute alcohol 70 ml + distilled water 30 ml Mix
8. T10E1	Mix 1.0 ml Tris 10 mM (8.0 pH) and 0.2 ml EDTA 1 mM(8.0 pH) and make up volume to 100 ml with sterile distilled water.
9. RNase (10mg/ml)	Dissolve 10 mg RNase in 1 ml sterile distilled water in a tube and place it water bath at 40° C for 10 minutes. Allow this to cool to room temperature and store it at -20^oC.
10. Ethidium bromide (10mg/ml)	Dissolve 10 mg Ethidium bromide in 1.0 ml distilled water in a tube, wrap the tube with aluminium foil and store at 4° C. **Caution : Ethidum bromide is extremely mutagenic**
11. Bromo phenol blue (BPB)	Dissolve 40 g sucrose and 0.25g of BPB in 60 ml distilled water and make up final volume to 100 ml
12. 50 X TAE	Dissolve 121.0g Tris buffer in 300 ml distilled water. Add 50.0 ml EDTA (0.5M) and 28.55 ml Acetic acid and make volume to 500 ml with distilled water. Autoclave it.

Appendix - IIIA. List of RAPD primers with their nucleotide sequences used for amplification of turmeric DNA.

S. No	Primers	Sequence
1	OPA -4	AATCGGGCTG
2	OPA -5	AGGGGTCTTG
3	OPA -7	GAAACGGGTG
4	OPA -8	GTGACGTAGG
5	OPA -10	GTGATCGCAG
6	OPB-9	TGGGGGACTC
7	OPC-13	AAGCCTCGTC
8	OPC – 16	CACACTCCAG
9	OPC - 20	ACTTCGCCAC
10	OPD – 1	ACCGCGAAGG
11	OPD -3	GTCGCCGTCA
12	OPE - 9	CTTCACCCGA
13	OPF -13	GGCTGCAGAA
14	OPG - 13	CTCTCCGCCA
15	OPG - 20	TCTCCCTCAG
16	OPJ -10	AAGCCCGAGG
17	OPJ - 18	TGGTCGCAGA
18	OPR -7	ACTGGCCTGA
19	OPS-12	CTGGGTGAGT
20	OPT -8	AACGGCGACA
21	OPT 20	GACCAATGCC

Appendix -IIIB. List of SSR primers with their nucleotide sequences used for amplification of turmeric DNA.

S. No	Marker		Primer sequence
1	CuMiSat-3	Forward Primer(5'-3')	GCACTACTTCCTTCTCGTTCAA
		Reverse primer(5'-3')	CGTCGTAAAGATTAGCGTGTG
2	CuMiSat-5	Forward Primer(5'-3')	AGCAGTGCGTCTTTCATC
		Reverse primer(5'-3')	CTCTTGTCACGGAACCTC
3	CuMiSat-8	Forward Primer(5'-3')	CATTGCGTGCCCACTTCC
		Reverse primer(5'-3')	CCTCCCTGTCGCTCTCCTC
4	CuMiSat-11	Forward Primer(5'-3')	ACAGTCCCCTTCCCACTC
		Reverse primer(5'-3')	TCTTGTTCCTATGCTCTACGC
5	CuMiSat-16	Forward Primer(5'-3')	CATTTGTTCTGCTCGCTTCTAC
		Reverse primer(5'-3')	CTGCTCCGCTGTCTCTCAC
6	CuMiSat-17	Forward Primer(5'-3')	ATGTGGTTGAGGAATGATGAGAC
		Reverse primer(5'-3')	CTATTTCCCATAGCCCTTGTAGC
7	CuMiSat-18	Forward Primer(5'-3')	GTTCACAGCTTTAGCAGGGACAA
		Reverse primer(5'-3')	CTCCTCTCCATATTCTCCATCTCG
8	CuMiSat-19	Forward Primer(5'-3')	CATGCAAATGGAAATTGACAC
		Reverse primer(5'-3')	TGATAAATTGACACATGGCAGTC
9	CuMiSat-20	Forward Primer(5'-3')	CGATACGAGTCCATCTCTTCG
		Reverse primer(5'-3')	CCTTGCTTTGGTGGCTAGAG
10	CuMiSat-21	Forward Primer(5'-3')	TCATTCAAAGTCCGATGGAA
		Reverse primer(5'-3')	TTCGAGTGCAGAAGGAGAATTA
11	CuMiSat-22	Forward Primer(5'-3')	AATTTATTAGCCCGGACCAC

		Reverse primer(5'-3')	AAGAAAGTGAGTAGAAACCAAAGC
12	CuMiSat-26	Forward Primer(5'-3')	CATTCCGATGAATTGTATG
		Reverse primer(5'-3')	GCAGTTGTTTTGCTTCAG
13	CuMiSat-27	Forward Primer(5'-3')	TATAGATAGCCATGCTGAAG
		Reverse primer(5'-3')	CCATTTTAGTTCATTACGTG
14	CuMiSat-28	Forward Primer(5'-3')	TTCAACTTCTCCTCGCTCAG
		Reverse primer(5'-3')	GCAAGGTCTGCATCTATTTCTC
15	CuMiSat-29	Forward Primer(5'-3')	GTGGTATCCCCATGAAGAGC
		Reverse primer(5'-3')	ATGACCAAGCCCTTTCACC
16	CuMiSat-30	Forward Primer(5'-3')	CTCTAATGTCGCCTCTCACG
		Reverse primer(5'-3')	GCATCTCCCGTTCTTCTCC
17	CuMiSat-35	Forward Primer(5'-3')	GGTTCGTCGCTGGAAAGTAAT
		Reverse primer(5'-3')	GCATCTCAACAGGGGCTG
18	CuMiSat-36	Forward Primer(5'-3')	TGGGCTCAATGGTTGATACG
		Reverse primer(5'-3')	CTCCTCATCGCTATCCGAGG
19	CuMiSat-37	Forward Primer(5'-3')	CCATTGGCGAGGATGAAGC
		Reverse primer(5'-3')	CCTGCCAAGCAAAGCCAAG