

**CHARACTERIZATION AND SELECTION OF KODO MILLET
(*Paspalum scrobiculatum* L.) GENOTYPES WITH HIGH CULM
STRENGTH TO SUIT MECHANICAL HARVESTING**

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TAMIL NADU AGRICULTURAL UNIVERSITY
COIMBATORE – 641 003**

2014

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*Thesis submitted in partial fulfillment of the requirements for the award of the degree of
DOCTOR OF PHILOSOPHY in PLANT BREEDING AND GENETICS
to the Tamil Nadu Agricultural University, Coimbatore.*

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CERTIFICATE

This is to certify that the thesis entitled 'CHARACTERIZATION AND SELECTION OF KODO MILLET (*Paspalum scrobiculatum* L.) GENOTYPES WITH HIGH CULM STRENGTH TO SUIT MECHANICAL HARVESTING' submitted in partial fulfillment of the requirements for the award of the degree of DOCTOR OF PHILOSOPHY in PLANT BREEDING AND GENETICS to the Tamil Nadu Agricultural University, Coimbatore is a record of *bonafide* research work carried out by Mrs. R. SREEJA under my supervision and guidance and that no part of this thesis has been submitted for the award of any other degree, diploma, fellowship or other similar titles. However, part of the thesis work has been published in peer reviewed scientific journal of national/international repute (copy enclosed).

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Acknowledgement

ACKNOWLEDGEMENTS

At the outset I would like to thank God for enabling me to write this thesis.

I owe a deep sense of gratitude to my Chairman, Dr. J. R. Kannan Babu, Professor and Head (Dept. of Pulses), CPBG, TNAU, Coimbatore for him being a nice human being. Thanks for his inspiring guidance and moral support rendered during the course of my study.

My sincere thanks are due to the members of advisory committee, Dr. A. Subramanian, Assistant Professor, Coconut Research Station, Aliyarnagar, Tamil Nadu for his insight on the research problem, technical guidance and critical counseling rendered during various stages of my study and Dr. A. Nirmalakumari, Professor, Centre for excellence in millets, Athiyandal village, Thiruvannamalai district, Tamil Nadu for her constructive comments and kind help. I am very much thankful to Dr. T. Raguchander, Professor (Plant Pathology), CPPS, TNAU for his support and encouragement. I am grateful to Dr. P. Govindaraju, Professor, AC and RI, Killikulam, for his guidance and for providing me all the assistance to carry out the biochemical analysis.

I express my gratitude and thanks to Dr. P. Veerabahiran, Professor, Dr. B. Selvi, Professor, Dr. R. Ravikesavan, Professor and Head, Dr. S. Sivakumar, Professor, Department of Millets, CPBG, for their support and encouragement throughout the study and Dr. A. Yuvaraj, Assistant Professor for providing all the facilities needed for the raising the crops.

My sincere thanks are due to Dr. N. Manivannan, Professor (Oilseeds) for helping me in my publication and Dr. K. Ayyanar, Assistant Professor (Forages) for providing offering help in data analysis.

My thanks are due to Dr. Sivakumar, Professor (Microbiology), Dr. S. Karthikeyan, Professor, (Bioenergy), Dr. D. Uma, Professor (Biochem), Dr. Uma, Research Associate (Bioenergy) for enabling me to carry out the biochemical analysis at Dept. of Bioenergy.

I wish to place on record my heartfelt thanks to Professor and Head and Dr. N. Natarajan, Professor, and Mrs. Pavadharani, JRF, at Dept. of Nanotechnology for helping me carry out the investigation using SEM.

I am very much thankful to Dr. C. R. Anandakumar, Director i/c, Center for Plant Breeding and Genetics and Dr. M. Maheswaran, Director of Research i/c, TNAU, for their kind support at times of need. My special thanks to our PG coordinator Dr. N. Meenakshi Ganesan, and Dr. P. L. Viswanathan, Professor and Head, Department of Oilseeds and our ex-coordinator for their guidance and constant encouragement during the course of study.

I feel happy to express my immense thanks to Dr. Pushpam, Assistant Professor, Dr. Manonmani, Professor, Dr. kalaiarasi, Assistant Professor, Dr. Kalaimagal, Professor, Trichy, Dr. Malarvizhi, Assistant Professor, Dr. Geethanjali, Assistant Professor, from Dept of Plant breeding and Dr. Kumudha, Professor (Microbiology) for their kind words and moral support.

I would like to express my gratitude to Dr. D. Sudhakar, Professor and Head (Biotech), Dr. J. Ramalingam, Professor and Head (Bioinformatics) and Dr. R. Gnanam,

professor, *Dr. E. Kokiladevi*, Assistant Professor and *Dr. N. Balakrishnan* at Dept. of Biotechnology for their support and help during the course of my study and always.

I wish to express my sincere thanks to all the teachers, who inspired, guided and encouraged to carry out my research work.

My special thanks are to *Mr. Balaji* (Biotech), presently a Ph. D. Scholar at Bharathiyar University for helping me to carry out the gene expression studies and to *Ms. Gayathri*, Research Associate (Biochemistry) for supporting me in doing the biochemical studies. *Dr. Ganesan* (Post harvest technology), a Ph. D scholar for enabling me to do the dehulling in Varagu.

It is my pleasure to thank all the research fellows, the Assistant Agricultural Officers, *Mrs. Mythili*, *Mr. Krishnamoorthy* and *Mr. Marudhachalm*, non-teaching staffs and labourers, *Mrs. Kribavathi*, *Mrs. Devi*, *Mrs. Kanimozhi*, *Mrs. Dhanalakshmi*, *Mrs. Bhuvana* and *Mani anna* and *Mrs. Gayathri* at Dept. of genetics and all the workers in Millets Department for helping me in every possible way to make my efforts worthwhile.

I profoundly thank my classmate and loving sister *Ms. Sathya* for standing by me through thick and thin.

My thanks are to all my class mates *Abirami*, *Anusheela*, *Dhasarathan*, *Jegadeeswaran*, *Karpagam*, *Maavimani*, *Mahendran*, *Malathi*, *Manimozhi Selvi*, *Manivannan*, *Mohan*, *Anand*, *Prabhu*, *Ameena*, *Bharathi raja*, *Rajarajan*, *Usha rani*, *Savitha*, *Sivamurugan*, *Surender*, *Sureshkumar*, *Tamilarasi*, *Vairam*, *Vishnuvarthini* and *Mr. Suryakumar*, for bringing back the enjoyment and memories of my college days. I take it a pleasure to thank all my beloved seniors and juniors for their support and help in times of need.

The financial assistance given by the *University Grants Commission, India* to carry out this research is gratefully acknowledged.

I would like to thank my friends and well wishers *Mrs. Rajeswari, Mrs. Padma and Mr. Ramesh Babu* for their constant support and encouragement. My thanks are to *Mrs. Sasikala*, who took care of my kids, when I was busy with my work.

I would like to dedicate my thesis to my parents and in-laws, husband, brother, kids, *Riju and Arya*, and all my relatives, for being my strength, all through. No words can express my gratitude to my husband, *Mr. Arul*, for without his support and encouragement, this wouldn't be possible.

(*Sreeja, R.*)

Abstract

ABSTRACT

CHARACTERIZATION AND SELECTION OF KODO MILLET (*Paspalum scrobiculatum* L.) GENOTYPES WITH HIGH CULM STRENGTH TO SUIT MECHANICAL HARVESTING

By

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This study was undertaken at Department of Millets, Centre for Plant Breeding and Genetics with a view to identify kodo millet genotypes suitable for mechanical harvesting and to discern factors contributing to culm strength in kodo millet at morphological, anatomical and biochemical level.

A collection of 297 germplasm accessions of kodo millet, obtained from Department of Millets were characterized and evaluated for traits with special reference to culm strength, to identify genotypes with better culm strength. Correlation and path analyses in these lines indicated that culm diameter, plant height and recovery angle after bending could be the candidate traits, based on which selection could be imposed to identify genotypes with good culm strength in kodo millet. Based on the results of correlation and path analyses, selection was exercised and a total of 30 lines were identified as having better culm strength and were forwarded for further evaluation in replicated trial. Two checks, CO3 (for yield) and *Adari* (for culm strength) and nine weak culm lines were also included for evaluation and comparison.

Genetic variability and diversity analyses were also carried out among the 41 genotypes. Variability studies suggested high level of genotypic and phenotypic variation

for traits such as pulling force, culm weight per unit length, and grain yield per plant indicating more scope for improving the culm strength as well as yield. High heritability for traits such as, days to 50 per cent flowering, culm diameter, culm weight per unit length, grain yield per plant, pulling force and days to maturity showed that these traits were least affected by environment and hence more reliable for selection and further genetic improvement of the respective traits. Culm weight per unit length, grain yield per plant, pulling force and culm diameter exhibited high heritability along with high genetic advance, indicating preponderance of additive gene action for the expression of these four traits.

Cluster and principal component analyses resulted in grouping of genotypes into five clusters with almost similar pattern in cluster composition in both the analyses. In PCA, the first principal component, which accounted for 39 per cent of total variability highly correlated with days to 50 per cent flowering, plant height and pulling force.

Thirteen strong culm lines which exhibited consistency in performance with respect to culm strength were forwarded for yield evaluation trials by raising them in individual plots with replications along with the two checks. Seven genotypes, which were extreme in their phenotype with respect to culm strength, were also included. Correlation and path analyses showed that only lignin per unit length of the culm had very high negative direct as well as indirect effects on degree of lodging, followed by culm diameter. Hence any improvement in lodging can be achieved through improving the lignin content and culm diameter. Among the 13 test entries, Aamo 5, Aamo 210, Aamo 258, Sel 21, TNPsc 176 and TNPsc 183 were superior, with respect to culm and yield related traits and hence these could be suggested as suitable to machine harvesting.

For characterization of culm strength at molecular and anatomical level, three elite genotypes, TNPsc 176, TNPsc 183 and Sel 21 which had better culm strength and yield, were chosen. They were raised in replicated trial, along with CO3 and *Adari*, the checks and Aamo10, a weak culm line. The anatomical study showed that the strong culm lines had high culm thickness, thicker mechanical tissues, more lignin deposition, and more number of vascular bundle per cross section, when compared with their weaker counterparts. Silicon relative signal (%) using SEM-EDX proved that silicon content

contributed for culm strength. Potassium relative signal (%) did not differentiate the lodging and non lodging lines and hence not associated with lodging behavior at least in these four genotypes studied. Moreover, potassium content was higher in non-lignified parenchyma cell walls compared to sclerenchyma cell walls.

Expression studies by qRT-PCR for *FLEXIBLE CULM1 (FC1)* gene, involved in lignin biosynthetic pathway confirmed the role of *FC1* gene in lignin accumulation as the highest expression recorded in strong culm check, *Adari* and the lowest expression in Aamo 10. The test entries TNPsc 183 and TNPsc 176 had optimal level of expression.

Among the three superior genotypes, TNPsc 183 was identified as the ideal line as it had sufficient culm strength, low incidence of lodging, non shattering of inflorescence and uniformity in maturity. The other two genotypes *viz.*, TNPsc 176 and Sel 21 were found to be better yielders with moderate lodging during grain filling, owing to their higher per plant productivity and test weight.

Abbreviations

LIST OF SYMBOLS AND ABBREVIATIONS

°	Degrees
%	Per cent
°C	Degree Celsius
ABD	Augmented Block Design
ADF	Acid Detergent Fibre
ADL	Acid Detergent Lignin
ANOVA	Analysis of Variance
BLAST	Basic Local Alignment Search Tool
PSI-BLAST	Position Specific Iterated-BLAST
CD	Critical Difference
cDNA	Complementary DNA
cm	Centimeter
ct	Cycle threshold
CTAB	Cetyl Trimethyl Ammonium Bromide
DEPC	Diethylpyrocarbonate
df	Degrees of Freedom
DMRT	Duncan's Multiple Range Test
EDTA	Ethylene Diamine Tetra Acetic acid
EDX	Electron Dispersive X-ray
<i>et al.</i>	and others
<i>FCI</i>	<i>FLEXIBLE CULM1</i>
Fig	Figure
GA	Genetic Advance
GAM	Genetic Advance as per cent of Mean
GCV	Genotypic Coefficient of Variance
g	Gram
ha	Hectare
h^2	Heritability
HCl	Hydrochloric acid
HCA	Hierarchical Cluster Analysis
<i>i.e.</i>	Which is to say, in other words
Kg	Kilogram
mm	Millimetre
mg	Milligram
ml	Millilitre

ng	Nanogram
MSA	Multiple Sequence Alignment
N	Newton
NDF	Neutral Detergent Fibre
No	Number
PCA	Principal Component Analysis
PCV	Phenotypic Coefficient of variation
PCR	Polymerase Chain Reaction
qRT-PCR	Quantitative Reverse Transcription-PCR
QTL	Quantitative Trait Locus
RBD	Randomized Block Design
RNA	Ribo Nucleic Acid
SCW	Secondary Cell Wall
SE	Standard Error
SEM	Scanning Electron Microscope
TAE	Tris Acetic acid EDTA
TB O	Toluidine Blue O
μg	Microgram
μl	Microlitre
μm	Micrometer
<i>viz.</i> ,	Namely
vs	versus

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Introduction

CHAPTER I

INTRODUCTION

India is endowed with a great wealth of agro-biodiversity and a large number of species are in cultivation in India. For strengthening food and nutrition security in India, in a climate changing scenario, production of small millets needs to be better appreciated, enhanced and promoted (Padulosi *et al.*, 2009). Small millets in India are represented by six species, namely, finger millet [*Eleusine coracana* (L.) Gaertner], kodo millet (*Paspalum scrobiculatum* L.), foxtail millet [*Setaria italica* (L.) Pal], little millet (*Panicum sumatrense* Roth ex Roemer & Schultes), proso millet (*Panicum miliaceum* L.) and barnyard millet [(represented by two species, *viz.*, *Echinochloa crusgalli* and *E. colona* (L.)]. The small millets are crops of antiquity and are known for their suitability to dry lands, hill and tribal agriculture and contribute to food security at farm and regional levels. They require less water for cultivation, mature early and are well suited for cultivation under water scarcity conditions. The resilience exhibited by these crops makes them ideal for climate change and contingency planting. Among small millets cultivated in India, kodo millet (*P. scrobiculatum* L.) is the second important crop, next only to finger millet.

Among the small millets, kodo millet (*P. scrobiculatum* L.), indigenous to India (de Wet *et al.*, 1983) is highly tolerant to drought and grown in marginal and dry lands, for its grain and fodder. It is a traditional, hardy and drought resistant crop cultivated in about 9 lakh hectares in India with an annual production of 3.11 lakh tonnes (Bondale, 1994; Singh, 1994). It thrives well in arid climates with minimum water requirement as compared to other cereal members and least affected by pest and diseases. Apart from its traditional role as a staple for the poor in the marginal agricultural regions, they are gaining new role as nutritional food for the urban high income people.

Premise of the work

In Tamil Nadu, this crop is cultivated in Trichy, Cuddalore, Villupuram, Perambalur, Vellore, Ramnad, Salem, Dharmapuri, Madurai and Pudukottai districts and the area under kodo millet contributes to about 2.5 per cent of the total area under millets in the state. It is grown predominantly as pure crop and in some places as mixed crop

with red gram, sesame and black gram and yields high gross return as compared to other popular crops owing to the high market price of the produce. But, due to rapid industrialization and large scale migration to nearby urban areas, labour is becoming increasingly scarce and also proving costly, leading to reduced net returns by kodo millet farmers.

The labour problem is more pronounced during harvest of the crop, because of its diversion to other crops like maize and cotton. Further due to the onset of cyclonic weather in the above districts during the last week of November and December, which coincides with crop maturity stage, manual harvesting, which is time consuming, results in loss of produce, in terms of quality and quantity, due to delayed harvest. Completion of harvest early provides the opportunity to cultivate a short duration crop of pulse or sesame, thereby increasing the cropping intensity and annual return from agriculture.

In Tamil Nadu, farmers use paddy combine harvesters to reap kodo millet. However, the spreading plant habit, poor culm strength and grain shattering of most of the local land races, lead to heavy lodging at maturity and therefore, the grain loss is more if harvested mechanically. Hence identification of a genotype in kodo millet which suits harvesting by machine is the need of the hour.

The architecture and physical strength of crop plants determine its ability to withstand mechanical harvesting. Not many literates available in this crop with regard to culm strength related studies. Rice, being a model crop has been fairly investigated for its mechanical strength, and the findings can be largely extended to other members of the grass family including kodo millet. Leaf sheath wrapping, basal internode length and the cross-sectional area of the culm are the major plant traits that determine straw strength (Change and Vergara, 1972). The leaf sheaths contribute to the breaking strength of the shoot by 30 - 60% (Chang, 1964). Pushing resistance has been used primarily as an index of resistance to stem bending in several crops (Terashima *et al.*, 1992; Won *et al.*, 1998). Besides, stem diameter and unit culm weight have been directly correlated with lodging resistance and the breaking strength of the stem (Zuber *et al.*, 1999).

In rice nodes, peripheral vascular tissue cells and sclerenchyma cells under the epidermal layer develop thickened cell walls are presumed to provide mechanical strength to the plant body.

Stem strength could also be attributed to its inorganic and organic constituents. Generally, lignin or cellulose determines physical strength, as lower lignin and cellulose cause the culm to be brittle (Jones *et al.*, 2001, Ma *et al.*, 2002; Tanaka *et al.*, 2003). In rice brittle culm1 (*bc1*) mutant line, the altered biosynthesis of cellulose, hemicellulose and lignin in culms reduced secondary cell wall thickness and mechanical strength (Li *et al.*, 2003). The brittle culm mutant 5 (*bc5*) in rice shows brittleness exclusively in nodes but other mutants (*bc1*, *bc2*, *bc3*, *bc4*, *bc6*, *bc7*) show the brittle phenotype in culms and leaves (Aohara *et al.*, 2009).

Likewise, higher silicon (Si) contents are also related to physical strength (Idris, *et al.*, 1975; Ma and Yamaji, 2006). Recent studies have shown that accumulation of Si in rice, a typical Si-accumulating plant, is attributed to an efficient uptake system mediated by two Si transporters *Lsi1* and *Lsi2* (Ma, 2010a).

With this background, the following objectives were formulated with the aim of identifying kodo millet genotype(s) which is non lodging, erect with good culm strength, non-shattering, together with synchronized maturity to suit mechanization. The objectives are,

- a. Phenotyping of kodo millet germplasm accessions based on morphological features.
- b. Screening and selection for high culm strength, bold seeded and erect genotypes with high yield.
- c. Characterization of culm strength based on anatomical and biochemical features.
- d. Analysis of differential expression of genes related to culm strength.

Review of literature

CHAPTER II

REVIEW OF LITERATURE

Small millets (finger millet, kodo millet, foxtail millet, barnyard millet, proso millet and little millet) are very important group of crops in semi-arid regions of the country. They require less water for cultivation, mature early and are well suited for cultivation under water-scarce conditions. The resilience exhibited by these crops makes them ideal for climate change and contingency planting.

Among the small millets, kodo millet, *Paspalum scrobiculatum* L. ($2n=4x=40$), grown for its grain and fodder, is indigenous cereal of India. The genus *Paspalum* (Poaceae) includes around 400 species and is widely distributed across the warmer regions of the world. It is an aggressive colonizer of disturbed habitats (de Wet, 1983). In West Africa, it is a weed of rice and is harvested as a wild cereal. It is grown as a cereal only in India. This cereal is known as *Kodo* in Hindi, *Khoddi* in Urdu, *Arugu* in Telugu and as *Varagu* in Tamil. The species probably harvested as a weed and later became domesticated across most of its range in India (de Wet, 1983).

Botanically, the plants are slender to stout growing to a height of 90cm and inflorescence, a panicle, consists of 3-7 racemes arranged alternately in the primary axis. The racemes are about 13 cm long with spikelets arranged in rows upon the flattened rachis. It is highly self-pollinated on account of highly cleistogamous flowers (Rangaswami-Ayyangar and Panduranga-Rao, 1934) and occasional apospory (Narayanaswami, 1954). The grains are covered by thick glumes and it has a 1000 grain weight of 6-7 gm. The bran and husk form a large proportion of the grain, about 37 per cent (Malleshi and Hadimani, 1994).

Among the small millets, kodo millet ranks second in area and production next to finger millet in India. Kodo millet was grown in 2.35 lakh hectares during 2005-06 in states, like Madhya Pradesh, Maharashtra, Tamil Nadu and Chattisgarh (http://www.dhan.org/smallmillets/docs/presentations/recent_development_and_changes.pdf).

Madhya Pradesh and Chhattisgarh share about 72 per cent of total area of kodo millet followed by Uttar Pradesh, Tamil Nadu, Gujarat, Maharashtra and Karnataka

(Yadava, 1997). In Tamil Nadu the crop is mainly sown under rain-fed conditions in the districts of Cuddalore, Villupuram and Perambalur and it contributed 2.4 per cent (5871 ha) of the total area under other millets in the state (<http://www.tn.gov.in/crop/AreaProduction.htm>)

Kodo millet production in Tamil Nadu declined from 10,455 tones (0.17% of total food production in Tamil Nadu) during 2004-05 to 6,676 tones (0.11% of total food production in Tamil Nadu) during 2005-06. (<http://www.tn.gov.in/crop/AreaProduction.htm>).

2.1 Importance of mechanization

India is a growing economy and its increasing population can be supported by increasing productivity and multiple cropping. To perform the agricultural operations timely, high capacity machines are required especially during harvest to reap the produce in time and to get maximum return in terms of quality and quantity. According to a status report on “Agricultural mechanization in India 2010”, by the Italian Trade Commission, it is expected that the percentage of population involved in agriculture will come down from the present 64 per cent to 40 per cent by 2020 and this would again increase the need for mechanization in agriculture.

2.2 Relevance of kodo millet in mechanization

Though, crops like rice, wheat form major share in India’s food production, small millets like, kodo millet, still contribute to the regional food security in dry and marginal lands, where major cereal crops fail to yield (Yadava and Jain, 2006). Nowadays, thrust to grow millets is given due to their nutritional superiority as compared to the major cereals. Moreover increasing life style diseases in the recent past, has created an awareness among the consumers to appreciate the nutritive values like presence of more dietary fibre, anti-oxidant (Chandrasekara and Shahidi, 2010) and anti-diabetic properties (Hegde *et al.*, 2005) of this crop. It has been reported to have higher free radical quenching potential when compared to other millets (Hegde and Chandra, 2005). Thus, apart from being a staple for the poor in the marginal agricultural areas, it is gaining attention as healthy food among the urban people as well. Hence, any technological intervention in this crop is essential to boost the production on a profitable scale.

In kodo millet growing tracts of Tamil Nadu, the crop is harvested with paddy combine harvesters, owing to the labour scarcity. The landraces currently in cultivation are not amenable for mechanized harvesting due to their tendency to lodge at grain filling and maturity stages thus resulting in heavy loss. Hence, identification of a non lodging genotype with good culm strength, which can withstand machine harvest, is the need of the hour.

2.3 Effects of lodging

Lodging of cereals is defined as permanent displacement of the culms from the upright position (Pinthus, 1973). Leaf shading and constriction of conducting tissues caused by lodging decrease both photosynthesis and photo-assimilation causing poor grain filling and reduced yield. Lodging has been shown to reduce yield, the quality of grain, and the efficiency of mechanical harvesting (Weber and Fehr, 1966; Kono 1995). A reduction of 20 to 40 per cent in cereal production has been attributed to lodging (Pinthus, 1973; Easson *et al.*, 1993). Lodging has also been reported to complicate harvesting and cause deterioration in the milling and baking quality of the grains due to increased moisture content of the grains and pre-harvest sprouting (Weber and Fehr, 1966; Kono, 1995). Furthermore, in lodged plants the contamination with mycotoxins produced by *Fusarium* species can be significantly increased (Langseth and Stabbetorp, 1996; Scudamore, 2000)

2.4 Causes of Lodging

Lodging happens in plants by environmental effects such as rain and wind. It can also be enhanced by different pathogens and pests affecting stems or roots (Keller *et al.*, 1999) or by agronomic practices such as excessive fertilization and/or high seeding rates in wheat (Stapper and Fischer, 1990; Easson *et al.*, 1993; Berry *et al.*, 2000). Heavy rain or irrigation may reduce the soil strength and the plant becomes prone to root lodging. Root lodging is more pronounced in plants having lesser root spread and depth and in soils with lesser humus and clay. But lodging is reduced by potassium fertilization (Tinarelli, 1988; Bhiah *et al.*, 2010), addition of silicon (Idris *et al.*, 1975) or 2,4-D spraying (Matsubayashi *et al.*, 1967).

Lodging, though largely influenced by environmental factors, is partly a genetic factor. Lodging may still occur in dwarf cultivars if stem strength is weak (Li, 1998), especially during grain filling and maturity due to the weight increase in the upper part of the stem. Hence, to avoid inherent tendency of the plant to lodge, sufficient culm strength is necessary to keep the plant erect till maturity and harvest.

2.5 Plant height as a target for lodging resistance

Semi-dwarf plants were introduced in rice and wheat to reduce lodging during green revolution period (Khush, 1999). Kelbert *et al.* (2004) reported after analyzing the morphological and anatomical features, that the plant height remains the most practical and easily selectable trait for lodging resistance in wheat. Moreover, Ookawa and Ishihara (1992) and Easson *et al.* (1993) had shown that susceptibility to lodging differed even among cultivars with similar heights in rice and wheat respectively and hence plant height is not necessarily the most important factor in determining lodging resistance in crop plants. Flintham *et al.* (1997) reported that there is an optimum plant height for maximum photosynthetic capacity and reducing plant height below this level may reduce crop yields. Cultivars with reduced sturdiness of the lower part are more easily lodged in the field, even in semi-dwarf lines of rice (Terashima *et al.*, 1992).

2.6 The reason behind targeting culm strength for suitability to machine harvest and non-lodging plant type

The architecture and physical strength of crop plants determine its ability to withstand mechanical harvesting. Strength of a plant always lies in its lower part, as it has to support the heavier upper part including the ear, leaves and upper stem (Kashiwagi *et al.*, 2005). To be efficiently cut by mechanical harvester, the plant type must be a compact one (erect or semi erect), with uniform maturity, sufficient height and culm strength and of non lodging type. Hence, a strong basal culm is required to keep the plant erect, while cutting by harvester's blade and also to prevent the plants from lodging.

Since, literatures are not available in kodo millet, with respect to traits relating to culm strength, supporting evidences have been taken from rice and other cereal members for this study. Mechanical rigidity or strength of culm has been studied so far in many crops like, rice (Li *et al.*, 2003; Aohara *et al.*, 2009; Torro *et al.*, 2011), wheat (Zuber *et al.*, 1999,

Kong *et al.*, 2013), barley (Cenci 1984), oat (Jellum 1962). Rice, being a model crop, has been fairly investigated for its mechanical strength, especially to study the lodging behavior and these findings can be largely extended to other members of the grass family including kodo millet to study its suitability to mechanical harvesting as well.

Phenotyping for compactness, uniform maturity and plant height are comparatively easier and require only direct methods to measure. But, the traits culm strength and lodging are complex, difficult to phenotype and highly influenced by environment. Hence, there are several studies which explain the methods to quantify the culm strength of a plant.

Most of the previous works on lodging resistance concentrated on reducing the plant height (Okada *et al.*, 1967, Tomita, 2009) and thereby improving culm strength and lodging resistance. Later on, investigations were carried which threw light on improving the mechanical strength of the culm and thereby improving lodging resistance. These studies relied on stem characteristics such as morphological traits of the culm, chemical components and their histological distributions in the culm and the genes involved in the synthesis/pathways of these chemical components. Studies on rice (Terashima *et al.*, 1994 and 1995) and wheat (Crook and Ennos, 1993 and 1994) have shown that root characteristics and anchorage also influence lodging resistance.

2.6.1 Stem characteristics

Lodging usually occurs when the stems bend or break at the basal internodes (Pinthus, 1973). Thus, the stem basal inter-node traits seem to be more important for lodging resistance in comparison to other aerial traits of plants (Xiao *et al.*, 2002). Moreover the combine harvester cut the plant at 20 cm height from the base of the plant. Hence strength of the plant at the basal portion is more important for lodging resistance and to suit machine harvest. The stem basal inter-node traits include morphological traits, anatomical traits and chemical components

2.6.1.1 Morphological traits

The frequently used method to evaluate lodging includes visual scoring of naturally or artificially lodged plants. The ranking is based on the fact that the degree and area of occurrence of lodging in the field directly reflect the lodging resistant level of

crops. Indirect evaluation includes, measurement of culm strength, plant height, basal inter-node length, culm diameter, weight per unit length of the culm, weight of the upper part of the plant and ear weight. Other than these some different methods were also used like, manual scoring of elasticity of the culm (Jezowski *et al.*, 1987; Keller *et al.*, 1999), measuring recovery angle after bending (Torro *et al.* 2011), testing the pushing resistance of the culm by specific instruments, like prostrate tester in wheat (Xiao *et al.*, 2002) and rice (Kashiwagi and Ishimaru *et al.*, 2004), digital force tester (Wang *et al.*, 2006) in wheat and other similar instruments in barley (Kokubo *et al.*, 1989) and maize (Fouere *et al.*, 1995).

2.6.1.2 Anatomical traits

Early studies showed that the lodging-resistant types of rice had a thicker band of sclerenchyma at the periphery of the stem compared with lodging-susceptible strains (Ramaiah and Mudaliar, 1934). Chang (1964) reported that lodging-resistant varieties in rice generally had higher number of vascular bundles in the culm and that a high percentage of outer vascular bundles were fused with the sclerenchyma and compact parenchyma cells in the culm and fewer or smaller air space within the culm.

Recent studies showed that lodging-resistant rice varieties had more vascular bundles in both the peripheral and the inner section of the outer layers, as compared to lodging-susceptible varieties (Chaturvedi *et al.*, 1995). Studies on solid and hollow stemmed wheat cultivars suggested high degree of correlations between lodging resistance and width of mechanical tissue (Kong *et al.*, 2013).

However, dependent on the plant materials and crops deployed, different or contradictory results had been reported. For example, some authors did not find a significant correlation between stem diameter and lodging resistances as in wheat (Atkins, 1938; Pinthus, 1967; Al-Qaudhy *et al.*, 1988; Kelbert *et al.*, 2004; Wang *et al.*, 2006). In oats and barley, the negative correlation between the stem diameter and stem strength have been observed (Norden and Frey, 1970; Dunn and Briggs, 1989).

2.6.1.3 Chemical components

Plant cell walls possess a strong fibrillous netted structure that provides mechanical support to cells, tissues, and the entire plant body (Li *et al.*, 2003). Many studies have

indicated that the lodging resistance is not only due to morphological and anatomical stem characters, but well associated with chemical ingredients (Kokubo *et al.*, 1989; Taylor *et al.*, 1999; Jones *et al.*, 2001; Wang *et al.*, 2012). The dry matter accumulated in the stem is mainly comprised of carbohydrates including monosaccharide, disaccharides and polysaccharides. The accumulation of dry matter, especially of polysaccharides, the basis for cellulose and hemi-cellulose production, results in stem wall thickening, which add up to an increase in stem strength (Wang and Li, 1996).

Cellulose, hemi-cellulose and lignin have been reported to be correlated with lodging resistance in wheat (Bernards and Lewis, 1998; Wang and Li, 1996). In *Arabidopsis*, Taylor *et al.* (1999) and Jones *et al.* (2001) reported that lignin and cellulose content is related to stem rigidity. Kokubo *et al.* (1989) found a high correlation between the cellulose content of barley cell wall and maximum bending stress.

According to Taylor *et al.* (1999), cellulose usually constitutes 20 to 30 per cent of the dry weight of the primary cell walls and 40 to 90 per cent of the secondary cell walls, depending on the cell type. According to them, in some cells, lignin may be incorporated into the cell wall, enhancing its mechanical strength. Lower lignin and cellulose have been reported to cause the culm to be brittle in *Arabidopsis* (Jones *et al.*, 2001), wheat (Ma *et al.*, 2002) and rice (Tanaka *et al.*, 2003).

Though, lignin is reported to impart mechanical strength to plants, its role in resistance of crops to lodging is still not clear (Frei, 2013). While lignin could lend mechanical support to the stalks, it might also have the opposite effect of making stalks more brittle and thus susceptible to mechanical damage (Davidson and Phillips, 1930). Some recent works indicated that high lignin levels were associated with lodging resistance in wheat (Ma, 2009 and 2010b) and pea (Banniza *et al.*, 2005), and a quantitative trait locus (QTL) for lodging resistance co-located with a QTL for high lignin content in ryegrass (Inoue *et al.*, 2004). A study on rice concluded that lignin played an important role in lodging tolerance but suggested that its distribution and density were more important than its concentration (Ookawa and Ishihara, 1993). No significant differences in lignin content were seen in wheat cultivars differing in lodging resistance (Travis *et al.*, 1996), while another study concluded that cellulose rather than lignin conferred resistance to lodging in wheat (Wang *et al.*, 2012).

In addition, brown-midrib mutations, which were associated with reduced lignin levels, did not affect lodging resistance in maize, sorghum, and pearl millet (Sattler *et al.*, 2010). A study on a maize mutant with drastically reduced mechanical strength suggested that cellulose rather than lignin deposition in the stalk was associated with susceptibility to lodging (Ching *et al.*, 2006).

Several authors have investigated the relationship between chemical elements and molecules versus lodging resistance. The stem of wheat contains 2.3 - 4.6 per cent silicon, which is mostly present in the epidermis of wheat culms, considered to contribute to lodging resistance (Li, 1979). Comparing the silicon contents between lodging resistant and susceptible wheat varieties, Gartner *et al.* (1984) observed significantly higher silicon content in the epidermis and mechanical tissue of culms in the lodging resistant variety. Moreover, silicon of the cell wall was thought to contribute to mechanical strength in rice stems (Zhang *et al.*, 2010a).

Among organic and inorganic constituents of culm, hemi-cellulose and potassium content are highly correlated with breaking strength, but silica content is not related to culm stiffness (Kono and Takahashi 1961a, 1961b). Before heading, large amount of starch accumulates in the culm and sheath. The starch was believed to contribute to shoot's stiffness (Sato, 1959). A later examination however showed no correlation between starch content and breaking strength (Kono and Takahashi, 1961a). Other chemical elements, calcium and magnesium are also associated with lodging resistance (Takahashi, 1995).

Chemical analysis includes estimation of cellulose content and its crystallinity, hemi-cellulose and lignin content, and other elemental analysis for silicon, potassium, calcium and magnesium in the culm, use of histochemical stains for cellulose (calcoflour) and lignin (phloroglucinol-HCl), FTIR, etc

2.6.2 Case studies relating to culm strength and lodging

Dunn and Briggs (1989) studied five Canadian six-row barley (*Hordeum vulgare* L.) cultivars, including two semi dwarfs ('Duke' and 'Samson'), that differ in lodging resistance and height. Significant cultivar differences were observed for culm length, number of internodes, length of four basal internodes, culm diameter, culm wall thickness, number of

vascular bundles, and thickness of the sclerenchyma ring. No cultivar differences were found for thickness of the sclerenchyma cell walls. Of the characters studied, culm length, basal internode length, culm wall thickness, and sclerenchyma ring thickness were most closely associated with differences in lodging resistance among the cultivars.

Grass culms are known to differ in breaking strength. To explain this, Kokubo *et al.* (1989) studied the fourth inter-node of 4 brittle and 2 non brittle barley (*Hordeum vulgare* L.) strains and reported that inner and outer culm diameters of brittle strains were not significantly different from those of non-brittle strains. But, maximum bending stress, at which the culm was broken, was 192 ± 34 g/mm for brittle and 490 ± 38 g/mm for non-brittle strains. Cell walls of brittle culms had 6 to 64 per cent as much cellulose content as those of non-brittle culms. Maximum bending stress correlated significantly with cellulose content per unit length of the culm ($r = 0.93$), but not with the contents of non-cellulosic compounds. Hence, they concluded that lower cellulose content of the brittle culm was significantly correlated with brittleness.

Ooawa *et al.* (1993) found that the densities of lignin, glucose and xylose in the cell wall materials of the fifth inter-node of different rice varieties grown under different conditions were associated with stem strength. Appenceller *et al.* (2004) recorded that the dry matter in unit length of maize stalk explained approximately half of the variation in mechanical strength of the stem.

Zuber *et al.* (1999) conducted a field study in 15 breeding lines of spring wheat (*Triticum aestivum* L.) to determine the relationship of morphological traits with lodging resistance. They found significant correlations of lodging score with stem diameter and stem weight per cm. They reported that thicker stems and heavier stems were indicative of better lodging resistance and explained 48.5 per cent and 49.7 per cent respectively of the phenotypic variation in lodging resistance. Multiple linear regression equations indicated that 77.2 per cent of the variation in lodging resistance was based on stem weight per cm and on the weight of the ear.

Li *et al.* (2000) studied the structural characteristics of *Triticum aestivum* cv Lankao 906-4, a thicker culm cultivar with Jing 411, a widely planted cultivar using Wiesner reaction (Phloroglucinol-HCl) and CLSM (Confocal Laser Scanning Microscope)

analysis using the samples taken from the second internode from base of the plant. They reported that the lodging resistance of the culm depended on the thickness and cell layers of the mechanical tissue and lignifications of the culm.

Zhu *et al.* (2004) based on their study in wheat hybrids reported that culm diameter, thicker wall, higher thickness-diameter ratio and higher percentage of mechanical tissue of culm and higher lignin content contributed for its better mechanical strength as compared to the hybrids.

Studies on F_1 and F_2 of wheat by Sarker *et al.* (2007) revealed that the second inter-node breaking strength was positively correlated with diameter, wall thickness and unit-stem weight of second inter-node, and main shoot weight but negatively correlated with plant height and second inter-node length at both genotypic and phenotypic levels. It was observed from path coefficient analysis in both F_1 and F_2 generations that the second inter-node unit-stem weight had high positive direct effect on its breaking strength. Main shoot weight, second inter-node diameter and wall thickness positively influenced breaking strength through its unit-stem weight.

Kashiwagi *et al.* (2008) studied two NILs (Near isogenic lines) in rice developed between cultivars Nipponbare and Kasalath, one containing a single stem diameter QTL (*sdm8*; NIL114), and another with four stem diameter QTLs (*sdm1*, *sdm7*, *sdm8*, *sdm12*; NIL28). They reported that the greater culm stiffness of the NILs was not a product of the quantitative or qualitative (cellulose crystallinity) differences in their cellulose, lignin, or silicon. They also suggested the possibility that QTLs for stem diameter concomitantly increased stem weight, culm wall thickness, stem non-structural carbohydrates, and thus result in greater physical strength measured in terms of pushing resistance. In addition, these same QTLs could also make higher plant height, lower tiller number, and greater crown width, which might have negative effects on lodging resistance and grain yield.

Torro *et al.* (2011) compared two methods for indirect evaluation of lodging resistance in rice, especially in field conditions: the recovery ability after bending (measured as the tiller angle difference before and after bending); and the resistance to pulling, to test the sturdiness of the lower part of the plant and concluded that tiller angle

difference, rather than pulling resistance, could be a valuable trait to select for resistance to lodging in early segregating generations of a pedigree breeding program.

Wu *et al.* (2011) compared three large culm rice cultivars with three common rice cultivars and reported that large culm cultivars had greater plant size, culm diameter, flag leaf length and width and lower tillers numbers. Anatomically they had larger leaf vascular bundles and more culm vascular bundles.

Wang *et al.* (2012) used FTIR (Fourier Transform Infrared Resonance) combined with principal component analysis and cluster analysis to detect heterogeneity in stem lodging resistance and found that the main chemical components changing among the developmental internodes of one cultivar were lignin and to a lesser extent cellulose. Hence, they proposed that the main cell wall component responsible for cell wall strength is cellulose. Histo-chemical staining of four types of wheat stems with various abilities to resist lodging revealed that cellulose contributed more than lignin to the ability to resist lodging.

Owing to paucity of literature pertaining to culm strength and traits associated with it in kodo millet, similar reports available in other cereal crops are reviewed hereunder.

Results of experiments on culm strength and lodging

Sl. No.	Crop	Findings	References
1	Wheat	The ratio of stem wall thickness to its outer radius, sclerenchyma tissue proportion, the average number of big vascular bundles per unit and the cellulose content are four important factors affecting the mechanical strength	Wang <i>et al.</i> (2006)
		Cellulose contributed more than lignin to the ability to resist lodging	Wang <i>et al.</i> (2012)
2	Rice	Wider culm diameter with a thinner culm wall maximizes lodging resistance without reducing yield potential	Ookawa <i>et al.</i> (2010)
		Reduction in cellulose content increases lignin content	Li <i>et al.</i> (2003) and Ambavaram <i>et al.</i> (2011)
		Sclerenchyma cells in stems provide the major support for stem integrity	Evans <i>et al.</i> (2007)
		The physical strength was positively and significantly correlated with the total amounts of potassium and silicon in culms during grain filling except that at the heading stage.	Zhang <i>et al.</i> (2010a)
		Culm length and silica content were the highest in lodging resistant cultivars. But acid detergent lignin (ADL) had no significant effect on lodging behavior	Hasan <i>et al.</i> (1993)
		The rice stem's mechanical property is mainly decided by its stem diameter, stem wall thickness, the number of big and small vascular bundles.	Chuanren <i>et al.</i> (2004)
3	Barley	Culm length, basal internode length, culm wall thickness, and sclerenchyma ring thickness were most closely associated with differences in lodging resistance among the cultivars	Dunn and Briggs (1989)
		The lower cellulose content was significantly correlated with brittleness of culm.	Kokubo <i>et al.</i> (1989)

Sl. No.	Crop	Findings	References
4	Bamboo	Larger stems had proportionally more sclerenchyma	Li <i>et al.</i> (1994)
5	Grass species	No correlation between stem diameter and the proportion of stem area attributed to sclerenchyma cells	Evans <i>et al.</i> (2007)
6	Maize	Correlation between Potassium content with crushing strength, ring thickness and weight of a 2-inch section were negative and statistically not significant	Zuber and Loesch (1966)

Correlation studies reported in other crops relating to culm strength and lodging

Sl No	Crop	Dependent trait	Component trait	Relationship	References
1	Wheat	Stem strength	Diameter of the basal inter-node	Positive	Wang <i>et al.</i> (2000), Min (2001), Yu <i>et al.</i> (2003), Yao <i>et al.</i> (2011)
			Wall thickness of the basal inter-node	Positive	Wang <i>et al.</i> (2000), Min (2001), Yu <i>et al.</i> (2003), Yao <i>et al.</i> (2011)
			Dry weight per cm of the basal inter-node	Positive	Wang <i>et al.</i> (2000), Min (2001), Yu <i>et al.</i> (2003), Yao <i>et al.</i> (2011)
			Length of the basal inter-node	Negative	Yao <i>et al.</i> (2011)
			Peduncle length	Negative	Yao <i>et al.</i> (2011)
			Plant height	Negative	Yao <i>et al.</i> (2011)

Sl No	Crop	Dependent trait	Component trait	Relationship	References
2	Wheat	Lodging resistance	Lignin content (%)	No correlation	Kong <i>et al.</i> (2013)
			Cellulose content (%)	No correlation	Kong <i>et al.</i> (2013)
			Width of the mechanical tissue layer	Positive	Kong <i>et al.</i> (2013)
			Width of stem wall	Positive	Kong <i>et al.</i> (2013)
			Weight of the three lower internodes	Positive	Kong <i>et al.</i> (2013)
3	Wheat	Width of the mechanical tissue layer	Width of stem wall	Positive	Kong <i>et al.</i> (2013)
			Weight of the three lower internodes	Positive	Kong <i>et al.</i> (2013)
4	Wheat	Area of transverse section	Number of vascular bundles	Positive	Kong <i>et al.</i> (2013)
			Number of small vascular bundles	Positive	Kong <i>et al.</i> (2013)
5	Wheat	Cellulose content	Lignin content	Positive	Kong <i>et al.</i> (2013)
6	Grass species	Stem diameter	Sclerenchyma cell area	No correlation	Evans <i>et al.</i> (2007)
7	Rice	Lodging resistance	Plant height	Positive	Chang and Vergara (1972)

Genes related to culm strength

To understand the mechanisms that regulate the mechanical strength of the plant body and the biosynthesis of plant cell walls, mutants defective in stem strength have been isolated and characterized. For example, the barley brittle culm (*bc*) mutants were first described based on the physical properties of the culms, which had 80 per cent reduction in the amount of cellulose and a twofold decrease in breaking strength compared with those of wild-type plants (Kokubo *et al.*, 1989, 1991), indicating that cellulose content is related to the mechanical strength of the plant body.

The brittle culm (*bc*) mutants of Graminae plants exhibit reduced mechanical strength of the plant body, especially in culms (Kokubo *et al.*, 1989, 1991; Ching *et al.*, 2006; Sindhu *et al.*, 2007). In rice, 13 *bc* mutants (*bc1*, *bc2*, *bc3*, *bc4*, *bc5*, *bc6*, *bc7*, *bc10*, *bc11*, *bc12*, *bc14*, *bc15* and *bc88*) had been found to date, and some of them were used as classic genome markers. These *bc* mutants are valuable materials for understanding the mechanism of secondary cell wall formation.

Rice *BC1* and maize *Brittle stalk 2* encode COBRA-like glycosyl-phosphatidylinositol-anchored proteins (Li *et al.*, 2003; Ching *et al.*, 2006). In rice, brittle culm1 mutant line, with reduced secondary cell wall thickness and mechanical strength, had altered biosynthesis of cellulose and lignin in culms and resulted in, approximately, 70 per cent reduction in cellulose and 30 per cent increase in Klason lignin compared with that of wild-type culms, indicating that plant cells might have a sophisticated mechanism to balance the contents between cellulose and lignin (Li *et al.*, 2003).

Rice *bc7* and *bc11* have been reported to have mutations in *OsCesA4*, a secondary cell wall specific Cesa protein whose sequence is highly similar to *AtCesA8/IRX1* (Yan *et al.*, 2007). Furthermore, on the basis of sequence similarity, *OsCesA4*, *OsCesA7*, and *OsCesA9* have been proposed to correspond to *AtCesA8*, *AtCesA4*, and *AtCesA7*, respectively (Tanaka *et al.*, 2003).

Leaf tension assays and bend tests of the internodes showed that the brittle phenotype [in *brittle stalk2* (*bk2*), a recessive mutant in maize] did not result from loss of tensile strength but from loss in flexibility that caused the tissues to snap instead of bend. (Sindhu *et al.*, 2007). The mature and developing stems of brittle mutant were markedly

enriched in lignin compared to wild type stems. As *Bk2* was highly expressed during early development, well before the onset of the brittle phenotype, they proposed that *Bk2* functions in a patterning of lignin-cellulosic interactions that maintain organ flexibility rather than having a direct role in cellulose biosynthesis. Hence, maize *Brittle stalk2* gene which encodes a COBRA-like protein was expressed in early organ development but required for tissue flexibility at maturity (Sindhu *et al.*, 2007).

Li *et al.* (2009a) identified a flexible culm1 (*fc1*) mutant by screening a rice T-DNA insertion mutant library. This mutant exhibited an abnormal development phenotype, including late heading time, semi-dwarf habit, and flexible culm. They then cloned the *FLEXIBLE CULM1 (FC1)* gene in rice using a T-DNA tagging approach. The *FC1* encodes a cinnamyl-alcohol dehydrogenase and is mainly expressed in the sclerenchyma cells of the secondary cell wall and vascular bundle region. In these types of cells, a deficiency of *FC1* in the *fc1* mutant caused a reduction in cell wall thickness, as well as a decrease in lignin. Extracts from the first internodes and panicles of the *fc1* plants exhibited drastically reduced cinnamyl-alcohol dehydrogenase activity. Further histological and biochemical analyses revealed that the ρ -hydroxyphenyl and guaiacyl monomers in *fc1* cell wall were reduced greatly. Their results indicated that *FC1* plays an important role in the biosynthesis of lignin and the control of culm strength in rice.

The *OsDRP2B* encoding a plant classical dynamin has been identified as the causative gene for the rice *bc3* mutant (Hirano *et al.*, 2010). The brittle culm mutant 5 in rice shows brittleness exclusively in nodes but other mutants (*bc1*, *bc2*, *bc3*, *bc4*, *bc6*, *bc7*) show the brittle phenotype in culms and leaves (Aohara *et al.*, 2009).

Using chromosome segment substitution lines, Ookawa *et al.* (2010) identified an effective quantitative trait loci for culm strength, which was designated *STRONG CULM2 (SCM2)*. Positional cloning of the gene revealed that *SCM2* was identical to *ABERRANT PANICLE ORGANIZATION1 (APO1)*, a gene previously reported to control panicle structure. A near-isogenic line carrying *SCM2* showed enhanced culm strength and increased spikelet number because of the pleiotropic effects of the gene.

Kotake *et al.* (2011) studied rice *Bc6* mutant line along with two cultivars IR 68 and Taichung 65 to represent wild-type control. The *Bc6* is a semi-dominant bc mutant

with easily breakable plant bodies. The *Bc6* encodes a cellulose synthase catalytic subunit, *OsCesA9*, and has a missense mutation in its highly conserved region. In culms of the *Bc6* mutant, the proportion of cellulose was reduced by 38 percent, while that of hemi-cellulose was increased by 34 percent. Introduction of the semi-dominant *Bc6* mutant gene into wild-type rice significantly reduced the percentage of cellulose, causing brittle phenotypes. Transmission electron microscopy analysis revealed that *Bc6* mutation reduced the cell wall thickness of sclerenchyma cells in culms. In rice expressing a reporter construct, *BC6* promoter activity was detected in the culms, nodes, and flowers, and was localized primarily in xylem tissues. This expression pattern was highly similar to that of *BC1*, which encodes a COBRA-like protein involved in cellulose synthesis in secondary cell walls in rice. These results indicated that *BC6* was a secondary cell wall-specific *CesA* that played an important role in proper deposition of cellulose in the secondary cell walls.

Rao *et al.* (2013) characterized a brittle culm 88 mutant of the *japonica* rice variety Wuyunjing 7 obtained by Ethylene Methyl Sulfonate induced chemical mutagenesis. Genetic analysis indicated that *bc88* was controlled by a single recessive nuclear gene. The *bc88* mutant exhibited a diversity of pleiotropic phenotypes, including brittle culm at the whole-plant growth stages, withered leaf tips at the seedling stage, and 18-days delay in heading date at the mature stage. Genetic analysis indicated that the *bc88* mutant was controlled by a single recessive nuclear gene. The mutated *bc88* gene isolated by map-based cloning contains only one point mutation in the 5th exon relative to its wild-type *BC88* (LOC_Os09g25490 and Os09g0422500), leading to an amino acid change from proline to lysine in *bc88* plants. Alignment of the putative protein sequence with its homologs indicated that the mutation was located in the conserved region of the sequence. Detection of the transcription level of *BC88* in rice plants shows that the expression level of *BC88* was higher in spikes and culms than in leaves, roots, and leaf sheaths. These contribute to understanding of the molecular mechanism of cellulose synthesis. The target gene *BC88* can be a useful tool in molecular marker-assisted selection for rice culm trait breeding.

Likewise, higher silicon contents are also related to physical strength (Idris, 1975; Ma and Yamaji, 2006). Recent studies have shown that accumulation of Si in rice, a typical Si-accumulating plant, is attributed to an efficient uptake system mediated by two Si transporters Lsi1 and Lsi2 (Ma, 2010a).

Genes and QTLs related to culm strength and lodging identified in several crops is listed hereunder.

Genes/QTLs, related to culm strength reported in different crops

Sl. No.	Gene/QTL	Associated function	References	Site of expression	Phenotype
1	Rice				
	<i>BC1</i>	Mutations in <i>BC1</i> cause reduction in cell wall thickness, cellulose content and increase in lignin level	Li <i>et al.</i> (2003)	Developing scleranchyma cells and in vascular bundles	Brittle phenotype in culms and nodes
	<i>BC5</i>	Involved in the formation of secondary walls in the node sclerenchyma tissue, especially one week after heading	Aohara <i>et al.</i> (2009)	Sclerenchyma in node	Brittle phenotype in nodes
	<i>BC6</i>	<i>Bc6</i> mutant, the proportion of cellulose was reduced by 38%, while that of hemicellulose was increased by 34% and reduced the cell wall thickness of sclerenchymal cells in culms	Kotake <i>et al.</i> (2011)	Culms, nodes, and flowers, and was localized primarily in xylem tissues	Easily breakable plant bodies
	<i>bc7(t)</i>	Identified from <i>japonica</i> variety Zhonghua 11 by means of ⁶⁰ Co-γ radiation. This mutant displays normal phenotype similar to its wild type plants except for the fragility of all plant body, with ~10% decrease in the cellulose content.	Yan <i>et al.</i> (2007)		

Sl. No.	Gene/ QTL	Associated function	References	Site of expression	Phenotype
	<i>BC12</i>	A gene encoding a kinesin-4 protein. BC12 functions as a dual-targeting kinesin protein and is implicated in cell-cycle progression, cellulose microfibril deposition and wall composition in the monocot plant rice.	Zhang <i>et al.</i> (2010b)	Tissues undergoing cell division and secondary wall thickening.	Dwarfism resulting from a significant reduction in cell number and brittleness.
	<i>BC15</i>	Reduced mechanical strength and cellulose content	Wu <i>et al.</i> (2012)		
	<i>bc 88</i>	<i>bc88</i> mutant, controlled by a single recessive gene, exhibits a diversity of pleiotropic phenotypes, including brittle culm at the whole-plant growth stages, withered leaf tips at the seedling stage, and 18-d delay in heading date at the mature stage.	Rao <i>et al.</i> (2013)	Spikes and culms	Brittle culm , withered leaf tips and delayed maturity
	<i>FLEXI- BLE CULM 1</i>	<i>fc1</i> mutant caused a reduction in cell wall thickness, as well as a decrease in lignin. <i>FCI</i> plays an important role in the biosynthesis of lignin and the control of culm strength in rice.	Li <i>et al.</i> (2009a)	<i>FCI</i> encodes a cinnamyl-alcohol dehydrogenase and is mainly expressed in the sclerenchyma cells of the secondary cell wall and vascular bundle region	Late heading time, semi-dwarf habit, and flexible culm.
	<i>prl5</i>	QTL from Kasalath on chromosome 5. NILs with <i>prl5</i> of Kasalath in the Nipponbare background, had higher dry weight and densities and the contents of accumulated carbohydrates than those of Nipponbare. Pushing resistance and lodging resistance were up to twice as high as in Nipponbare, without affecting yield.	Kashiwagi and Ishimaru (2004)	Lower stem	

Sl. No.	Gene/QTL	Associated function	References	Site of expression	Phenotype
	<i>SCM2</i>	<i>Strong culm 2</i> , a pleiotropic QTL for culm strength and increased spiklet number from chromosome 6 of Habataki, a high yielding Indica variety.	Ookawa <i>et al.</i> (2010)		Enhanced culm strength and increased spikelet number
2	Maize				
	<i>Brittle stalk 2</i>	Functions in patterning of lignin-cellulosic interactions that maintain organ flexibility.	Sindhu <i>et al.</i> (2007)		
3	Wheat				
	<i>COMT</i>	Involved in lignin biosynthetic pathway related to stem rigidity and lodging character	Ma <i>et al.</i> (2002)	Stem, root and leaf tissues	Lodging
4	Arabidopsis				
	<i>SHINE</i>	A transcription factor from <i>Arabidopsis thaliana</i> in rice, causes 34% increase in cellulose and 45% reduction in lignin with no compromise in plant strength and performance.	Ambavaram <i>et al.</i> (2011)	Culm	No changes in phenotype. Rice <i>AtSHN</i> Lines have altered cell walls in mechanical tissues

2.7 Studies relating to yield and other related traits in kodo millet

Many attempts have been made to determine the yield factors through correlation and path analyses in kodo millet [Amawate (1965), Dhagat *et al.* (1971), and Dhagat (1978)]. Association analysis carried out by Verma and Singh (1982) indicates that plant height was positively correlated with grain yield in early and medium maturing genotypes of kodo millet. In early genotypes, plant height also showed positive correlation with

panicle length but it was negatively correlated with 1000 grain weight. Path analysis revealed that only plant height had direct influence on grain yield in all maturity groups. The panicle length contributed indirectly *via* plant height towards grain yield.

Parihar (1985) observed significant positive correlation between grain yield and days to flowering, days to maturity, plant height, grain-straw ratio, fodder yield, 1000 grain weight and grains per panicle. All these characters also exhibited positive association with fodder yield. Path analysis revealed that fodder yield, grain-straw ratio, racemes per ear and days to flowering are the major components of grain yield in kodo millet.

Significant positive association of grain yield with tillers per plant, racemes per ear, biological yield and harvest index was noted by Rao and Tiwari (1988). Tillers per plant showed significant positive association with ears per plant and positively and significantly associated among themselves. Yasin *et al.* (1988) found significant positive association between productive tillers per plant and grain yield.

Yadava and Jain (2006) based on studies with 261 germplasm accessions consisting of early (<100 days) and late (>100 days) groups in kodo millet reported that plant height, tillers per plant and biological yield had significant positive association with grain yield in early maturing genotypes, while plant height, tillers per plant, flag leaf length and biological yield exhibited significant positive association with grain yield in late maturing genotypes. Based on path analysis, they reported that, ear length, biological yield and tillers per plant in early maturing genotypes, while biological yield, harvest index, peduncle length and plant height in late maturing genotypes are the main components of grain yield in kodo millet. According to them, tillers per plant, plant height, panicle length, and biological yield are the major yield determining factors in kodo millet.

2.8 Variability, heritability and genetic advance studies in kodo millet

Janoria (1963) was the first to study the variability in different strains of kodo millet collected from Madhya Pradesh and classified them on the basis of leaf color, spike structure and density with grain yield. Amawate (1965), Salpute (1966), Tiwari and Janoria (1967), Amawate and Dabral (1970), Choudhary and Singh (1970) and Rao (1991) also observed wide range of variability for morphological characters and grain yield in land races of Madhya Pradesh.

Verma and Satpute (1969) and Ahluwalia *et al.* (1970) also recorded high heritable variability for days to flowering, ears per plant and tillers per plant in kodo millet. The magnitude of phenotypic coefficient of variation was high for dry matter production, grain yield per plant, panicle number per plant and basal tillers per plant whereas it was low for developmental traits like days to flowering and days to maturity in kodo millet.

Dhagat *et al.* (1971), Dhagat (1978), Parihar (1985), Sharma (1988), Rao and Tiwari (1988) and Kandaswamy *et al.* (1990) found maximum heritable variation for tillers per plant, ears per plant, ear weight, grains per ear, fodder yield and grain yield. They also noted low amount of heritable variation for plant height and phenological attributes, days to flowering and period from flowering to maturity in the genotypes of kodo millet studied by them.

2.9 Variability studies and genetic analysis for culm strength in other crops

Li (1998) reported that stem strength, stem diameter and pith diameter were controlled by both additive and non-additive gene effects.

Vijayalakshmi *et al.* (2008) studied genetic divergence in 128 low land rice germplasm accessions and reported high GCV and phenotypic coefficient of variation (PCV) for effective tillers per plant and test weight. High heritability observed for days to 50 per cent flowering, effective tillers per plant, plant height, panicle length, L/B (length/Breadth) ratio and test weight, ranging from 76.46% in panicle length to 98.9% in test weight. High heritability coupled with high genetic advance as percentage of means was observed for effective tillers per plant and test weight.

Dash *et al.* (2011) screened 225 Tropical japonicas (TJ) rice varieties and their derivatives for super plant type traits to be utilized as prospective donors for improving yield. Variability study for the parameters *viz.*, number of grains per panicle, plant height, tiller number and test weight exhibited comparatively higher phenotypic as well as genotypic coefficient of variation. The characters, test weight, plant height and days to flowering recorded high heritability, whereas, specific leaf weight, culm strength and flag leaf angle as moderate heritability. On the basis of estimated h^2 and GA, the traits *viz.*, plant height, flag leaf angle, culm strength, specific leaf weight and 100 grain weight could be regarded more amenable to selection.

A study was conducted by Mewa *et al.* (2013) to assess variability in some culm and grain yield characteristics of recombinant inbred lines of a cross between *Eragrostis tef* and *Eragrostis pilosa*. Grain yield revealed high genetic coefficient of variation (GCV) while lodging index revealed moderate. Moderate GCV was also recorded for plant height, panicle length and culm characters (first and second culm diameter and strength). Genetic advance was high for grain yield and lodging index. Moderate genetic advance was recorded for culm related characters. Lodging index revealed consistently strong association with second culm strength.

2.10 Diversity studies in kodo millet

The first study on genetic diversity in kodo millet was made by Dhagat (1978). The study indicated no relation between genetic and geographic diversity. However the germplasm lines from Georgia, Transvaal, Paraguay, Argentina and India were grouped in different clusters indicating highly diverse nature of these genotypes. Parihar (1985) studied genetic divergence among 100 genotypes using D^2 analysis. This study also evidenced the absence of any relation between genotypic and geographical divergence. Yadava and Jain (2006) studied 261 germplasm accessions comprising 133 early and 128 late maturing ones. They reported that the percent contribution to diversity was maximum by plant height followed by tillers per plant, flag leaf length, peduncle length and ear length in both early and late maturing types. The least contribution towards divergence was made by harvest index.

Materials and Methods

CHAPTER III

MATERIALS AND METHODS

The present investigation was primarily focused to identify kodo millet (*Paspalum scrobiculatum* L.) accession(s) suitable for mechanical harvesting. The study was carried out at the Millet Breeding Station farm, Centre for Plant Breeding and Genetics (CPBG), Tamil Nadu Agricultural University (TNAU), Coimbatore, between 2012 and 2014. The station is located at 11° N latitude and 77° E longitude, at an elevation of 411.98 meters above mean sea level with annual rain fall of 730 mm.

3.1 Materials

3.1.1 Phenotyping of kodo millet germplasm accessions based on morphological features [Kharif (Aug. - Dec.), 2012]

The study material consisted of 297 germplasm accessions of kodo millet sourced out from the Department of Millets, TNAU, Coimbatore, Tamil Nadu. The list of materials used for initial evaluation is presented in table 1. The checks used were CO3, a high yielding variety released by TNAU, and Adari a local land race under cultivation in Tamil Nadu.

3.1.2 Screening and selection of high culm strength genotypes based on morphological characteristics [Summer (Jan. - May), 2013]

A total of 41 accessions forwarded from the previous season were raised. This included 30 strong culm lines, nine weak culm lines and two checks viz., CO3 and Adari. The list of materials used along with their origin are presented in table 2.

3.1.3 Evaluation of morphological and biochemical characteristics for culm strength [Kharif (Aug. - Dec.), 2013]

A total of 15 accessions selected from previous season trial were raised. These include, Aamo 5, Aamo 13, Aamo 126, Aamo 210, Aamo 258, RK 50, Sel 21, TNPsc 156, TNPsc 176, TNPsc 183, TNPsc 201, Peruvargu, Thittakudi along with two checks (CO3 and Adari). Besides, seven other entries representing extreme phenotypes with respect to culm strength were raised for comparison. This comprised of six genotypes

Table 1. List of kodomillet germplasm accessions used for initial evaluation [*Kharif* (Aug. – Dec.), 2012]

Sl. No.	Accessions	Sl. No.	Accessions	Sl. No.	Accessions	Sl. No.	Accessions	Sl. No.	Accessions	Sl. No.	Accessions
1	Aamo 4	26	Aamo 101	51	Aamo 307	76	Aamo 379	101	RK 25/2	126	IPS 115
2	Aamo 5	27	Aamo 109	52	Aamo 308	77	Aamo 382	102	RK 31	127	IPS 117
3	Aamo 6	28	Aamo 126	53	Aamo 309	78	Aamo 384	103	RK 50	128	IPS 118
4	Aamo 8	29	Aamo 134	54	Aamo 210	79	Aamo 385	104	RK 51	129	IPS 122
5	Aamo 10	30	Aamo 173	55	Aamo 311	80	Aamo 394	105	RK 51/1	130	IPS 123
6	Aamo 13	31	Aamo 179	56	Aamo 312	81	Aamo 398	106	RK 65-18	131	IPS 125
7	Aamo 14	32	Aamo 182	57	Aamo 316	82	Aamo 400	107	RK 73	132	IPS 129
8	Aamo 27	33	Aamo 203	58	Aamo 318	83	Aamo 408	108	RK 79	133	IPS 131
9	Aamo 29	34	Aamo 207	59	Aamo 322	84	Aamo 413	109	RK 80	134	IPS 147
10	Aamo 31	35	Aamo 208	60	Aamo 325	85	Aamo 414	110	RK 82	135	IPS 160
11	Aamo 32	36	Aamo 211	61	Aamo 326	86	Aamo 416	111	RK 84	136	IPS 194
12	Aamo 33	37	Aamo 213	62	Aamo 327	87	Aamo 422	112	RK 92	137	DPS 95
13	Aamo 36	38	Aamo 214	63	Aamo 330	88	Aamo 423	113	RK 96	138	DPS 368
14	Aamo 37	39	Aamo 215	64	Aamo 335	89	Aamo 463	114	RK 96/1	139	DPS 637
15	Aamo 58	40	Aamo 224	65	Aamo 337	90	Aamo 536	115	RK 111	140	DPS637/1
16	Aamo 62	41	Aamo 243	66	Aamo 340	91	Aamo 591	116	RK 162	141	GPUK 3
17	Aamo 68	42	Aamo 267	67	Aamo 345	92	Aamo 592	117	RBK 73	142	GPUK 72
18	Aamo 72	43	Aamo 268	68	Aamo 347	93	Aamo 593	118	RBK 155	143	APK 1
19	Aamo 73	44	Aamo 271	69	Aamo 352	94	Aamo 594	119	RBK 155/1	144	Sel 1
20	Aamo 75	45	Aamo 275	70	Aamo 354	95	Aamo 595	120	ICK 76	145	Sel 2
21	Aamo 79	46	Aamo 258	71	Aamo 355	96	Aamo 596	121	ICK 86	146	Sel 3
22	Aamo 83	47	Aamo 280	72	Aamo 360	97	Aamo 597	122	ICK 1042	147	Sel 4
23	Aamo 89	48	Aamo 299	73	Aamo 362	98	RK 5	123	ICK 7114	148	Sel 5
24	Aamo 90	49	Aamo 303	74	Aamo 365	99	RK 25	124	IPS 102	149	Sel 6
25	Aamo 96	50	Aamo 306	75	Aamo 366	100	RK 25/1	125	IPS 113	150	Sel 7

Table 1 continued...

Sl. No.	Accessions	Sl. No.	Accessions	Sl. No.	Accessions	Sl. No.	Accessions	Sl. No.	Accessions	Sl. No.	Accessions
151	Sel 8	176	TNPSc 93	201	TNPSc 118	226	TNPSc 144	251	TNPSc 170	276	TNPSc 195
152	Sel 9	177	TNPSc 94	202	TNPSc 119	227	TNPSc 145	252	TNPSc 171	277	TNPSc 196
153	Sel 10	178	TNPSc 95	203	TNPSc 120	228	TNPSc 146	253	TNPSc 172	278	TNPSc 197
154	Sel 11	179	TNPSc 96	204	TNPSc 121	229	TNPSc 147	254	TNPSc 173	279	TNPSc 198
155	Sel 12	180	TNPSc 97	205	TNPSc 122	230	TNPSc 148	255	TNPSc 174	280	TNPSc 199
156	Sel 13	181	TNPSc 98	206	TNPSc 123	231	TNPSc 149	256	TNPSc 175	281	TNPSc 201
157	Sel 14	182	TNPSc 99	207	TNPSc 124	232	TNPSc 150	257	TNPSc 176	282	TNPSc 202
158	Sel 15	183	TNPSc 100	208	TNPSc 125	233	TNPSc 151	258	TNPSc 177	283	TNPSc 203
159	Sel 16	184	TNPSc 101	209	TNPSc 126	234	TNPSc 153	259	TNPSc 178	284	TNPSc 204
160	Sel 17	185	TNPSc 102	210	TNPSc 127	235	TNPSc 154	260	TNPSc 179	285	TNPSc 205
161	Sel 18	186	Aamo 30	211	TNPSc 128	236	TNPSc 155	261	TNPSc 180	286	TNPSc 206
162	Sel 19	187	TNPSc 104	212	TNPSc 129	237	TNPSc 156	262	TNPSc 181	287	TNPSc 211
163	Sel 20	188	TNPSc 105	213	TNPSc 130	238	TNPSc 157	263	TNPSc 182	288	TNPSc 215
164	Sel 21	189	TNPSc 106	214	TNPSc 131	239	TNPSc 158	264	TNPSc 183	289	TNPSc 217
165	<i>Thittakudi</i>	190	TNPSc 107	215	TNPSc 133	240	TNPSc 159	265	TNPSc 184	290	TNPSc 221
166	<i>Vridhachalam</i>	191	TNPSc 108	216	TNPSc 134	241	TNPSc 160	266	TNPSc 185	291	TNPSc 225
167	TNPSc 81	192	TNPSc 109	217	TNPSc 135	242	TNPSc 161	267	TNPSc 186	292	TNPSc 230
168	TNPSc 82	193	TNPSc 110	218	TNPSc 136	243	TNPSc 162	268	TNPSc 187	293	TNPSc 231
169	TNPSc 83	194	TNPSc 111	219	TNPSc 137	244	TNPSc 163	269	TNPSc 188	294	TNPSc 232
170	TNPSc 84	195	TNPSc 112	220	TNPSc 138	245	TNPSc 164	270	TNPSc 189	295	TNPSc 233
171	TNPSc 85	196	TNPSc 113	221	TNPSc 139	246	TNPSc 165	271	TNPSc 190	296	TNPSc 235
172	<i>Pudukooraipettai</i>	197	TNPSc 114	222	TNPSc 140	247	TNPSc 166	272	TNPSc 191	297	TNPSc 236
173	TNPSc 90	198	TNPSc 115	223	TNPSc 141	248	TNPSc 167	273	TNPSc 192	298	CO3 (check)
174	TNPSc 91	199	TNPSc 116	224	TNPSc 142	249	TNPSc 168	274	TNPSc 193	299	<i>Adari</i> (check)
175	TNPSc 92	200	TNPSc 117	225	TNPSc 143	250	TNPSc 169	275	TNPSc 194		

Table 2. List of kodomillet millet germplasm accessions used for evaluation in summer (Jan.-May), 2013.

Sl. No.	Accessions	Pedigree
A	<i>Strong culm genotypes</i>	
1	Aamo 5	Not known
2	Aamo 13	Not known
3	Aamo 126	Not known
4	Aamo 210	Not known
5	Aamo 258	Not known
6	RK 50	Not known
7	Sel 21	Not known
8	TNPSc 86*	IPS 85
9	TNPSc 147	Selection from TNPSc 51-600-5-1-1
10	TNPSc 155	Selection from <i>Athipatti</i> , Dharmapuri
11	TNPSc 156	Selection from TNPSc 95
12	TNPSc 176	Selection from Aamo 599
13	TNPSc 183	Selection from Aamo 608
14	TNPSc 201	Selection from <i>Peruvaragu</i>
15	TNPSc 215	IC 283797 (from NBPGR, Hyderabad)
16	TNPSc 217	IC 283805 (from NBPGR, Hyderabad)
17	TNPSc 221	IC 308984 (from NBPGR, Hyderabad)
18	TNPSc 225	IC 345002 (from NBPGR, Hyderabad)
19	TNPSc 231	IC 382888 (from NBPGR, Hyderabad)
20	TNPSc 232	IC 384076 (from NBPGR, Hyderabad)
21	TNPSc 233	IC 426676 (from NBPGR, Hyderabad)
22	TNPSc 235	IC 426678 (from NBPGR, Hyderabad)
23	TNPSc 236	IC 426679 (from NBPGR, Hyderabad)
24	<i>Athipatti</i> *	Land race from Athipatti, Tamil Nadu
25	<i>Naraiyur</i>	Land race from Naraiyur, Tiruvannamalai, Tamil Nadu
26	<i>Pacheri</i>	Land race from Pacheri, Sivaganga, Tamil Nadu
27	<i>Podivaragu</i>	Land race from Peraiyur, Tamil Nadu
28	<i>Peruvaragu</i> *	Land race from Peraiyur, Tamil Nadu.
29	<i>Thittakudi</i>	Land race from Thittakudi, Thanjavur, Tamil Nadu
30	<i>Venganoor</i>	Land race from Venganoor, Ariyalur, Tamil Nadu

Table 2 continued...

Sl. No.	Accessions	Pedigree
<i>B</i>	<i>Weak culm genotypes (used for comparison)</i>	
31	Aamo 8	Not known
32	Aamo 10	Not known
33	Aamo 29	Not known
34	Aamo 36	Not known
35	Aamo 37	Not known
36	Aamo 72	Not known
37	Aamo 398	Not known
38	TNPSc 106	Selection from mutant 51-600-5-1-1
39	TNPSc 170	Selection from DPS 72
<i>C</i>	<i>Checks</i>	
40	CO3 (for yield)	A variety from pure line selection (Gengia selection)
41	<i>Adari</i> (for culm strength)	Land race from Adari, Cuddalore, Tamil Nadu

*Included for evaluation from this season.

with weak culm characteristics viz., Aamo 8, Aamo 10, Aamo 36, Aamo 398, TNPsc 106 and TNPsc 170 and a land race Athipatti for strong culm.

3.1.4 Assessment of yield and degree of lodging in six selected kodo millet genotypes [Summer (Jan. - May), 2014]

Three high yielding and high culm strength lines viz., TNPsc 176, TNPsc 183 and Sel 21 were raised during this season, along with checks (CO3 and *Adari*) and one weak line (Aamo 10) for comparison to understand lodging behavior and yield potential of the genotypes.

3.1.5 Anatomy and elemental analysis for culm strength in select kodo millet genotypes [Summer (Jan. - May), 2014]

Culm anatomy was performed through histochemical staining and scanning electron microscopy (SEM). Besides, elemental analysis for silicon and potassium was taken up with SEM- EDX (electron dispersive x-ray) for the following genotypes, *Adari*, Aamo 10, Sel 21 and TNPsc 183.

3.1.6 Real-time quantification of gene expression in select kodo millet genotypes

Quantification of expression of the gene *FLEXIBLE CULM1* involved in lignin biosynthesis was performed in the following genotypes, *Adari*, Aamo 10, Sel 21 and TNPsc 183.

3.2 Methods

3.2.1 Phenotyping of kodo millet germplasm accessions based on morphological features [Kharif (Aug. - Dec.), 2012]

The experiment was laid out in augmented block design (ABD) (Federer 1956; 1961) with four blocks and the checks placed randomly in each block (Plate 1). Each accession was raised in a single row plot with a spacing of 22.5 x 10 cm spacing and all the recommended package of practices was adopted. Observations were recorded in five plants per genotype on 13 morphometric traits viz., days to 50 per cent flowering, plant height, number of productive tillers, inflorescence length, raceme number, raceme length, culm diameter, pulling force, internode length, recovery angle after bending, days to maturity, grain yield and test weight and four qualitative traits, viz., plant type, ear exertion

at dough stage, spikelet arrangement on rachis and degree of lodging, as described in tables 3 and 4.

Mean performance of all the genotypes for the characters studied was calculated. For traits relating to yield, (days to 50 per cent flowering, plant height, number of productive tillers, inflorescence length, raceme number, raceme length, days to maturity, grain yield and test weight) mean of test entries were compared with that of CO3, the check for yield and for traits relating to culm strength (culm diameter, internode length, recovery angle after bending and pulling force) the mean of test entries were compared with that of *Adari*.

Based on the mean performance, correlation and path analyses were carried out using pulling force as dependent trait and other traits *viz.*, days to 50 per cent flowering, plant height, number of productive tillers, culm diameter, internode length, recovery angle after bending, days to maturity and grain yield as independent traits. The traits which had true relationship with pulling force were used for further selection of culm strength.

3.2.2 Screening and selection of high culm strength genotypes based on morphological characteristics [Summer (Jan. - May), 2013]

All the 41 genotypes were raised in randomized block design (RBD) with two replications (Plate 2). In each replication, the genotypes were raised in 4 rows of 3 m length with a spacing of 22.5 x 10 cm. All the recommended agronomic practices were followed to ensure a uniform crop stand. Observations were recorded on 14 quantitative traits *viz.*, days to 50 per cent flowering, plant height, number of productive tillers, inflorescence length, raceme number, raceme length, culm diameter, pulling force, culm weight per unit length, internode length, recovery angle after bending, days to maturity, grain yield and test weight, and all the eight scorable traits, *viz.*, plant type, ear exertion at dough stage, spikelet arrangement on rachis, degree of chlorosis, stay green nature at maturity, shattering of inflorescence, uniformity in maturity and degree of lodging (Table 3 and 4). The genotypes which were significantly better with respect to culm strength and related traits were identified and forwarded to next season.

Besides analyzing the mean performance of all the genotypes, variability and diversity studies were carried out using 14 quantitative traits.

3.2.3 Evaluation of morphological and biochemical characteristics for culm strength [Kharif, (Aug. - Dec.), 2013]

The selected 22 (15 selected genotypes +7 extreme lines for comparison) genotypes including the two checks (CO3 and *Adari*) were raised in RBD with two replications in plots of ten rows per genotype per replication (Plate 3). Observations were recorded for the traits as mentioned in table 3 and 4.

Cellulose, hemicellulose and lignin were estimated per unit length of the dry culm as well as on the basis of the weight of dry culm. For the above estimation, the culm samples (second internode from the base of the plant) were collected at physiological maturity from each of the genotypes.

Based on the mean performance, correlation and path analyses were carried out using degree of lodging as dependent trait and the culm related traits such as cellulose per unit length of the culm, hemicellulose per unit length of the culm, lignin per unit length of the culm, pulling force, culm weight per unit length, culm diameter and recovery angle after bending as independent traits. Selection was mainly based on culm strength and yield. The biochemical data obtained on 22 genotypes were subjected to Analysis of variance (ANOVA) and subsequently to Duncan's multiple range test (DMRT) to identify the significantly differing genotypes.

3.2.4 Assessment of yield and degree of lodging in six selected kodo millet genotypes [Summer (Jan. - May), 2014]

A total of six genotypes *viz.*, three test entries (Sel 21, TNPsc 176 and TNpsc 183), two checks CO3 and *Adari* and one weak culm line Aamo 10 were raised in large individual plots in RBD with four replications to assess the plot yield and lodging behavior (Plate 4).

3.2.5 Anatomy and elemental analysis for culm strength in select kodo millet genotypes [Summer (Jan. - May), 2014]

All the six genotypes were subjected to histochemical staining using Phloroglucinol-HCl and TBO stains to visualize the localization of lignin in culm cross sections under light microscope.



Plate 1. Field view of kodo millet genotypes raised during *kharif* (Aug. - Dec.), 2012



Plate 2. Field view of kodo millet genotypes raised during summer (Jan. - May), 2013



Plate 3. Field view of kodo millet genotypes raised during *kharif* (Aug. - Dec.), 2013



Plate 4. Field view of kodo millet genotypes raised during summer (Jan. - May), 2014

Table 3. Description of the quantitative traits studied

Sl. No	Trait	Unit	Abbreviation used	Description
1	Days to 50 per cent flowering	days	DF	Counted as days from sowing to 50 per cent of the plants in flowering.
2	Plant height	cm	PH	Taken from base of the plant to tip of the panicle on the main tiller.
3	Number of productive tillers	count	Ptil	Total number of harvestable panicle bearing tillers per plant.
4	Inflorescence length	cm	InfloL	Measured from node of lowest raceme (thumb) to tip of the last raceme.
5	Raceme length	cm	RL	Length of the longest raceme (excluding thumb) in an inflorescence.
6	Raceme number	count	RN	Number of racemes above first (lowest) primary axis node in an inflorescence.
7	Culm diameter	mm	CD	Measured at middle of the second internode from the base of the plant on the main tiller, using digital Vernier caliper. The cross section of the internode is elliptical; hence, the shorter diameter was measured (Plate 5).
8	Pulling force	Newton	PF	It is the force required to bend the whole plant to an angle of 45° from the ground level and measured using force gauge (Torro <i>et al.</i> , 2011) after setting the gauge to read the force at peak-hold. This setting will give the maximum force required to displace or bend the plant from erect position to 45° angle (Plate 6).



Plate 5. Measurement of culm diameter using Vernier caliper



Plate 6. Measurement of pulling force using force gauge

Table 3 continued...

Sl. No	Trait	Unit	Abbreviation	Description
9	Internode length	cm	IntL	The length of the second internode from the base of the plant on the main tiller was measured
10	Recovery angle after bending	degrees	RA	The angle of recovery made by the plant after bending it to the ground level by holding the tip of the plant. To read the angle, a protractor is kept behind the plant (Plate 7).
11	Culm weight per unit length	mg/cm	WL	The dry weight of the culm from the second inter-node from the base of the plant on the main tiller divided by internode length.
12	Days to maturity	days	DM	Number of days taken from sowing to physiological maturity of 50 per cent of the main tillers
13	Grain yield per plant	g	GY	Mean grain yield of three random plants.
14	Test weight	g	TW	Weight of random samples of 1000 seeds from the total harvest of an accession.
15	Grain yield per plot	Kg	PY	Weight of grains harvested from a single plot

Except the trait, days to 50 per cent flowering, all the traits were recorded at 50 per cent physiological maturity of the crop.

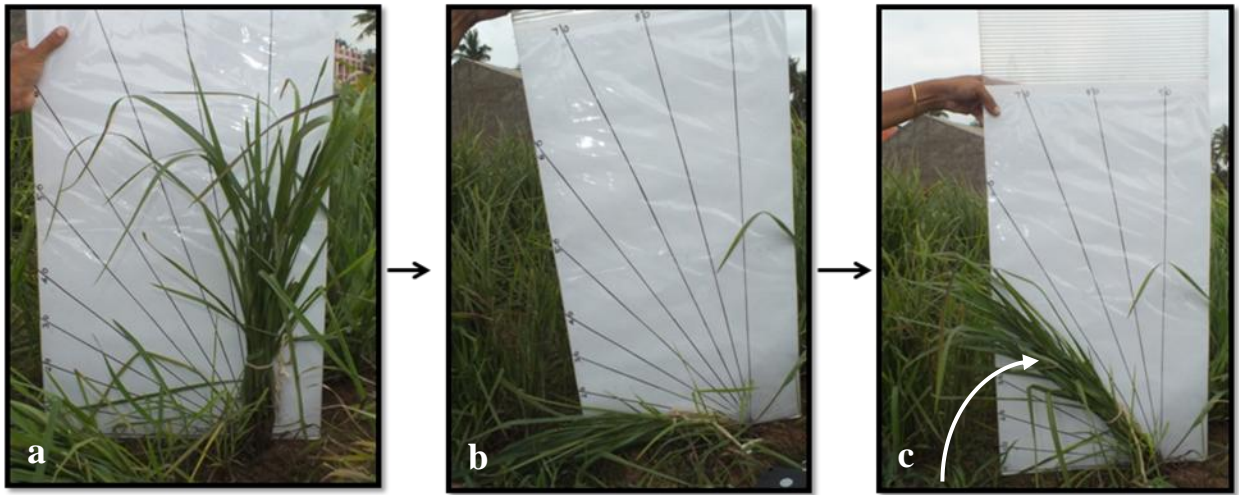


Plate 7. Measurement of recovery angle after bending

a. Plant at normal position; **b.** Plant was bent to the ground level gently by holding the tip of the plant; **c.** The plant was then slowly released. Now the angle made by the plant from the ground level was measured.

Table 4. Description of qualitative traits studied

Sl. No	Trait	Unit	Abbreviation	Description
1	Plant type	score	PT	1: Erect; 2: Decumbent; 3: Prostrate
2	Ear exertion at dough stage	score	EE	1: Complete; 2: Partial (Plate 8)
3	Spikelet arrangement on rachis	score	SA	1: Regular ; 2: Irregular; 3: Variable
4	Degree of chlorosis	score	DC	3: low; 5: intermediate; 7: high (Plate 9)
5	Stay green nature at maturity	score	SG	0: Absent; 1: Present
6	Shattering of inflorescence	score	SI	0: Absent; 1: Present (Plate 10)
7	Uniformity in maturity	score	UM	0: Absent; 1: Present (Plate 11)
8	Degree of lodging	score	DL	3: Low degree of lodging; 5: Intermediate degree of lodging; 7: High degree of lodging (Plate 12)

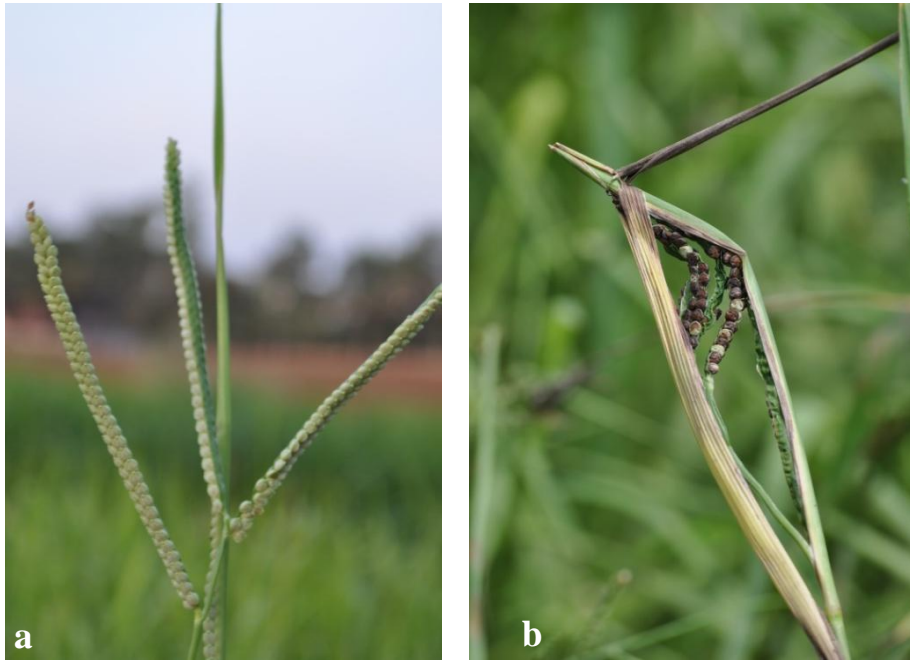


Plate 8. Ear exertion
a. Full (TNPsc 183); b. Partial (TNPsc 233)



Plate 9. Degree of chlorosis
a. High (score 7) in TNPsc 235; b. Low (score 3) in TNPsc 236



Plate 10. Shattering of inflorescence
a. Absent (Sel 21); b. Present (Aamo 10)



Plate 11. Uniformity in maturity
a. Present (Sel 21); b. Absent (Aamo 126)



Plate 12. Degree of lodging

a. High (score 7), in Aamo 10; **b.** Low (score 3), in Aamo 13

Ultra structure studies included analyzing cellular dimensions and elemental composition for silicon and potassium using Scanning Electron Microscope (SEM) with Electron Dispersive X-ray analysis (EDX) in select genotypes. For cellular dimensions and elemental analysis, measurements were taken at five different points in a cross section per sample.

3.2.6 Real-time quantification of gene expression in selected kodo millet genotypes

For differential gene expression studies, culm samples were collected on 80 days after sowing from selected kodo millet genotypes. The samples were harvested from the culm at a height of 20 cm from the ground level. The expression of the gene, *FLEXIBLE CULM 1 (CAD7)*, was analyzed through quantitative Reverse Transcription-Polymerase Chain Reaction (qRT-PCR).

3.3 Observations recorded

3.3.1 Morphological characters

Morphological characteristics studied included 15 quantitative traits and eight qualitative traits (Tables 3 and 4).

3.3.2 Biochemical characters

Estimation of cellulose, hemicellulose and lignin were done as per the protocol suggested by Goering and Van Soest (1975) and expressed in percentage. The culm samples were collected from the basal portion (second internode from base) of the main tiller of each plant at physiological maturity of the crop

Kokubo *et al.* (1989) in barley and Kong *et al.* (2013) in wheat estimated the cellulose and lignin content per unit length of the culm and recorded significant differences between genotypes. On similar lines, analyses were carried out to quantify the cellulose, hemicellulose and lignin content per unit length of the culm, *i.e.*, uniform length of the culm, from each second internode from the base of the plant and expressed in milligram per centimeter of the culm.

3.3.2.1 Cellulose, hemicellulose and lignin analyses

i. Principle

The plant materials were oven dried at 70° C and then grounded to give a particle size < 1 mm. The finely powdered material was used for determination of acid detergent fiber (ADF), neutral detergent fiber (NDF) and acid detergent lignin (ADL).

NDF was first determined after treatment with a neutral detergent solution (sodium lauryl sulphate and EDTA). The residue consisted of cellulose, hemicellulose and lignin. Then, ADF was determined by treatment of residue with an acid detergent solution Cetyl Tri-methyl Ammonium Bromide (CTAB) in sulfuric acid solution. The residue contained cellulose and lignin. Finally, ADL was determined after initial treatment for ADF measurement followed by removal of the cellulose fraction through extraction using 72 per cent H₂SO₄. The residue contained only lignin. A fraction of acid-soluble lignin and cellulose could be lost during this procedure (Van Soest 1987; Monties 1989). Acid resistant residue was recovered by filtration on a glass crucible with an asbestos filter, carefully washed and dried at 70° C for 24 hours to constant weight. This acid insoluble residue was insoluble lignin. After weighing, the residue was incinerated at 550 ° C for at least five hours and lignin was calculated after correcting for mineral elements (Van Soest 1987).

ii. Reagents for NDF, ADF and ADL determination

- a. *Neutral detergent solution*: A quantity of 18.61 g of CTAB and 6.81 g of sodium borate deca-hydrate were dissolved in 200 ml of distilled water by heating. To this solution, 30 g of sodium lauryl sulphate dissolved in 200 ml distilled water, 10 ml of 2-ethoxy ethanol and 4.5 g of disodium hydrogen phosphate dissolved in 100 ml of distilled water were added and the volume was made up to one litre, finally the pH was adjusted to 7.0.
- b. Deca-hydro-naphthalene (Decalin)
- c. Sodium sulphite (Na₂SO₃)
- d. Acetone
- e. Acid detergent solution: CTAB (20 g) was dissolved in one litre of 1N sulphuric acid
- f. 72 per cent H₂SO₄ (w/v)

iii. Procedure

a. Determination of NDF

To 1 g of the powdered sample in a refluxing flask, 10 ml of cold neutral detergent solution and 2 ml Decalin and 0.5 g sodium sulphite were added sequentially. The contents in the flask were allowed to boiling and refluxing for 60 min. The refluxed contents were filtered through sintered glass crucible (G-2) by suction and washed with hot water. Finally the residue was given two washings with acetone and then transferred to a pre-weighed crucible and dried at 100 °C for eight hours. The crucible was then cooled in a desiccator and weighed (Goering and Van Soest, 1975)

b. Determination of ADF

To 1 g of powdered sample taken in a round bottom flask, 100 ml of acid detergent solution was added. The mixture was then allowed to boil and refluxed for 1 h. The contents were then filtered through a pre-weighed sintered glass crucible (G-2) by suction and washed with hot water twice. The residue was given final washings with acetone. Acetone washing was continued until the filtrate was colorless. The residue was then dried at 100 °C for overnight and weighed after cooling in a desiccator.

c. Determination of ADL

To the ADF, 25-50 ml of 72 per cent sulphuric acid and 1 g asbestos were added and allowed to stand for 3 h with intermittent stirring with a glass rod. The solution was then diluted with distilled water and filtered through a pre-weighed Whatman no.1 filter paper. The residue was washed several times with water to get rid of the acid. The filter paper with residue was dried at 100 °C and weighed after cooling in a desiccator and transferred to a pre-weighed silica crucible. Then the contents were ashed in a muffle furnace at 550 °C for about 3 h, the crucible was then cooled in a desiccator and ash content was weighed. Simultaneously, a blank was run with 1 g asbestos and 72 per cent H₂SO₄ alone.

iv. Calculation

$$\text{Percent ADL} = \frac{(\text{Weight of 72\% H}_2\text{SO}_4 \text{ washed fibre} - \text{Ash}) \times 100}{\text{Weight of sample}}$$

Simple subtraction rules were used to calculate cellulose, hemicelluloses and lignin:

$$\text{ADF} - \text{ADL} = \text{Cellulose}$$

$$\text{NDF} - \text{ADF} = \text{Hemicellulose}$$

$$\text{ADL} = \text{Lignin}$$

The results for cellulose, hemicellulose and lignin were expressed as percentage of dry mass of plant material per cent dry matter. Cellulose, hemicellulose and lignin contents were also estimated for unit length of the culm by taking uniform length of 10 cm dry culm from each genotype and the contents expressed in milligrams per centimeter.

3.3.3 Anatomical studies of culm

The culm samples from the second internode from base of the plant were chosen for anatomical analysis. All the genotypes raised during Summer, 2014 were subjected to anatomical studies using light microscope. Only the extremes (*Adari* and *Aamo 10*) were subjected to SEM with EDX analysis for anatomical characterization of culm strength. The lines selected as suitable for mechanical harvesting (*Sel 21* and *TNPsc 183*) were further studied using SEM with EDX for confirmation of the results obtained.

3.3.3.1 Histochemical staining of culm cross section

In order to study the structural organization of the culm, cross sections were made so that enough light can be transmitted through the specimen to resolve the cell structures under the microscope. Free hand sectioning with razor blade was used for preparing specimens for microscopic viewing. Stains like Toluidine Blue O (TBO) and Phloroglucinol-HCl were used to study the cellular details and lignification of the cells respectively. The light microscope was equipped with a camera (Ultra scope 9.1_v) to capture the images.

3.3.3.1.1 Study on general cellular details using TBO stain

Since biological tissues have very little contrast, TBO stain was used to study the cellular details. The TBO is a polychromatic dye. Its reaction with carboxylated polysaccharides such as pectic acid give pinkish purple colour and with polyphenolic substances such as lignin produces bright blue color (O'Brien *et al.*, 1964).

Stain preparation

A quantity of 0.1 g of TBO was dissolved in 100 ml of 0.1 M benzoate buffer (benzoic acid 0.25 g, sodium benzoate 0.29 g, water 200 ml) with pH 4.4.

i. Staining procedure

The culm section was flooded with an aqueous solution of 0.1 per cent TBO solution for one minute. Then the excess stain was removed by using a piece of filter paper. The section was then flooded with water and excess water was removed. A drop of 30 per cent glycerol was added to the section and a cover glass was applied to examine it under the microscope.

3.3.3.1.2 Study on lignifications using Phloroglucinol-HCl stain

Lignin is a common constituent in the secondary wall of plant cells e.g., the walls of the xylem elements and sclerenchyma tissue. The cinnamaldehyde end group of lignin reacts with Phloroglucinol-HCl to give a red-violet color (Gahan, 1984)

i. Stain preparation

A quantity of 2 g of Phloroglucinol was added to 80 ml of 20 per cent ethanol and then 20 ml of concentrated HCl (12N) was added to it.

ii. Staining procedure

The stem bit sections were flooded with Phloroglucinol-HCl stain for two minutes. The lignified elements would turn red in a few minutes. The sections were transferred to a clean slide using a camel brush. A drop of 30 per cent glycerol solution was added and covered with a cover slip to be viewed under a microscope.

3.3.3.2 Ultrastructure of culm

3.3.3.2.1 Measurement of cellular features

Measurements were on number of vascular bundles, culm thickness (μm), sclerenchyma layer (arc) thickness, cell wall thickness (μm) of sclerenchyma cells in the fiber sheath and parenchyma tissue

3.3.3.2.2 Elemental composition using SEM-EDX

Elemental (Silicon and Potassium) composition (%) in different tissues (sclerenchyma layer in the outer region, sclerenchyma in the fiber sheath and parenchyma tissue walls) constituting the culm were measured to characterize the culm strength with respect to elemental composition in the following genotypes, *Adari*, Aamo 10, Sel 21 and TNPsc 183.

3.3.3.3 Real time gene expression analysis using qRT-PCR

3.3.3.3.1 Sample collection for RNA extraction

Culms from field grown samples were collected on 80 days after sowing. The samples were harvested from the culm at a height of 20 cm from the ground level. The samples were instantly frozen in liquid nitrogen and stored in – 80 °C before being taken up for RNA extraction.

3.3.3.3.2 Isolation of total RNA from culm tissue

The RNA was isolated using TRIzol reagent (Bio Basic Inc. cat. no. BS409/ BS410). The samples were ground using liquid nitrogen and transferred to an RNase free eppendorf tube. To this 1 ml of TRIzol reagent was added and centrifuged at 12,000 rpm for 10 min at 4 °C. The supernatant was taken in a separate tube, incubated at room temperature (RT) for 5 min and 0.2 ml of chloroform was added. A vigorous shaking/vortexing was given for 15 sec, incubated at RT for 3 min, followed by centrifugation at 12,000 rpm for 15 min at 4 °C. To the aqueous phase, 0.5 ml of isopropyl alcohol was added, incubated at RT for 10 min and centrifuged at 12,000 rpm for 10 min at 4 °C. The supernatant was discarded, the pellet was washed with 1 ml of 75 per cent ethanol and centrifuged at 7500 rpm for 5 min at 4 °C. Finally, the pellet was air dried and dissolved in 30 µl of RNase free water and further stored at - 80 °C. The RNA was treated with DNase enzyme to remove genomic DNA from RNA preparation.

It was noted that the pestle and mortars used for grinding, and the tips and tubes used in the extraction processes were treated with RNase inhibitor (DEPC) for 24 hrs and double autoclaved prior to use. The pestle and mortar were pre-chilled in the refrigerator

before grinding the sample. The working table was cleaned with 70 per cent ethanol to avoid any RNase contamination. The micropipettes were also cleaned with 70 per cent ethanol, after removing the tip ejector.

3.3.3.3.3 Quality and quantity check of the extracted total RNA

Isolated total RNA samples were resolved on 1.2 per cent agarose gels (Sambrook *et al.*, 1989) for checking the quality. Required amount of agarose was boiled in 1X TAE (0.9 M Tris-Acetate, 0.002 M EDTA, pH 8.0) in a microwave oven until complete dissolution. The molten agarose was cooled to 60 °C, 1-2 µl ethidium bromide (0.5 mg/ml stock) was added and mixed gently avoiding bubble formation. The mixture was poured into the gel cast with suitable combs. Required amount of total RNA was mixed with 6X loading dye (0.72 ml formamide, 0.16 ml 10X electrophoresis buffer, 0.26 ml formalin, 0.1 ml glycerol, 0.08 ml saturated bromophenol blue, 0.18 ml sterile distilled water) in 1:6 ratio. The samples were loaded into the wells and electrophoresed at 70 volts. After electrophoresis, the RNA profile was visualised on a UV-transilluminator. The quantification of RNA was taken up with Nano Drop Spectrophotometer (Thermo Scientific Inc.). Finally, A_{260}/A_{280} value was determined.

3.3.3.3.4 First strand cDNA synthesis

The RT-PCR reaction had essentially two parts i) first strand cDNA synthesis from mRNA using M-MuLV reverse transcriptase ii) the first strand cDNA synthesized was used as the template for second strand synthesis for making into double stranded DNA. The first strand cDNA was synthesized from the isolated RNA samples. An equal concentration of RNA (1µg) was taken in all reactions for cDNA synthesis. Fermentas Revert AidTM first strand cDNA synthesis kit (cat. no. 1621) was used and the reaction was carried out in Eppendorf thermal cycler. The second strand synthesis was carried out with gene specific primers.

3.3.3.3.5 Design of primers for quantitative gene expression analysis

The gene encoding the enzyme cinnamyl-alcohol dehydrogenase (CAD), which is associated with lignin biosynthesis, was chosen for real time gene expression analysis. In the absence of gene sequence in kodo millet, rice gene *OsCAD7 (FLEXIBLE CULM1)* reported by Li *et al.* (2009a), was used as query to find orthologous sequences from other

cereal members. The PSI-BLAST was carried out on National Centre for Biotechnology Information (NCBI) database and at least five full length coding sequences (CDS) of *CAD7* orthologs were retrieved. Further, multiple sequence alignment (MSA) was constructed for the orthologous sequences using the tool CLUSTALW. Conserved motifs were earmarked in the multiple alignments and forward and reverse primers for real time gene expression analysis in kodo millet were picked up from the strictly conserved segments in MSA. An internal control gene, *actin* was also used in this analysis. Rice *actin*, was used as query to retrieve orthologous sequences from other cereal members. Forward and reverse primers for amplifying *actin* in kodo millet were designed as it was done for *CAD7*.

Orthologous gene sequences used for designing primers for *CAD7* (test gene)

1	<i>Oryza sativa ssp. japonica</i>	NP_001053848.1
2	<i>Oryza brachyantha</i>	XP_006652804.1
3	<i>Setaria italica</i>	XP_004976783.1
4	<i>Brachypodium distachyon</i>	XP_003580533.1
5	<i>Sorghum bicolor</i>	XP_002447082.1

Orthologous gene sequences used for designing primers for *actin* (reference gene)

1	<i>Oryza sativa ssp. japonica</i>	NP_001051086.1
2	<i>Oryza brachyantha</i>	XP_006650501.1
3	<i>Setaria italica</i>	XP_004981971.1
4	<i>Brachypodium distachyon</i>	XP_003560589.1
5	<i>Zea mays</i>	NP_001150462.1

The following were the list of primers used for quantitative reverse transcription-Polymerase chain reaction (qRT-PCR)

qFC-F	5' – CAGGAGATGATGGACTTCG – 3'	<i>CAD7</i>
qFC-R	5' – ACGAAGCGGTACCTGACG – 3'	
qAct-F	5' – TGGCTCCTAGCAGCATGAAG – 3'	<i>actin</i>
qAct-R	5' – ATGGATCCTCCAATCCAGAC – 3'	

Real time analysis PCR was carried out in BioRad CFX96 machine (facility availed from the Department of Crop Physiology, TNAU, Coimbatore). Three biological replicates and two technical replicates were maintained for each of the genotypes. Each biological replicate was constituted by pooling four individual culms.

The real time reaction was prepared as below:

Roche FastStart Essential SYBR Green I Master mix (2X)	5 μ l
Template (cDNA)	1 μ l (100 ng)
Primer (Forward)	1 μ l (5 pico mole)
Primer (Reverse)	1 μ l (5 pico mole)
MilliQ water	2 μ l
Total volume/reaction	10 μ l

The following conditions were followed for RT- PCR analysis:

Initial denaturation	95° C, 3 min
Cycle denaturation	95° C, 10 sec
Annealing - Extension	55° C, 30 sec
Number of cycles	40

Relative gene expression was calculated by the formula,

$$\text{Relative gene expression} = \frac{2^{\text{ct (test gene)}}}{2^{\text{ct (reference gene)}}$$

Where,

ct = cycle threshold

3.4 Statistical Analysis

The mean values of five plants were computed for each replication and utilized for further statistical analysis. Mean, Variance and Standard error, were calculated by adopting the method suggested by Panse and Sukhatme (1967).

3.4.1 ANOVA for Augmented block design

ANOVA for augmented block design was done using the online statistical tool available at Indian Agricultural Statistics Research Institute (IASRI) website (<http://www.iasri.res.in/spadweb/default.aspx>).

3.4.2 ANOVA for RBD

The data collected were subjected to ANOVA in RBD as suggested by Panse and Sukhatme (1967). This was used to split up the variance into its three major components *viz.*, replication, treatment and error variance.

ANOVA for RBD

Sources of variation	Df	SS	MS	Expectation	F calculated
Replications (r)	r-1	RSS	RMS		RMS/EMS
Treatments (t)	t-1	TSS	TMS (=MS ₁)	$\sigma^2e + \sigma^2g$	TMS/EMS
Error	(r-1)(t-1)	ESS	EMS (=MS ₂)	σ^2e	-
Total	rt-1	Total SS	-		-

Where,

$$\begin{aligned}
 R &= \text{number of replications} \\
 t &= \text{number of genotypes} \\
 df &= \text{degrees of freedom} \\
 SS &= \text{sum of squares} \\
 MS &= \text{mean sum of squares} \\
 \text{Total SS} &= \sum Y_{ij}^2 - CF, \\
 \text{RSS} &= 1/t \sum R_j^2 - CF, \\
 \text{TSS} &= 1/r \sum T_i^2 - CF \\
 \text{where, } i &= 1, 2, 3, \dots, t; \\
 j &= 1, 2, 3, \dots, r \\
 F &= (\text{Grand total})^2 / rt \\
 \text{ESS} &= \text{Total SS} - \text{RSS} - \text{TSS} \\
 \text{RMS} &= \text{RSS} / (r-1) \\
 \text{TMS} &= \text{TSS} / (t-1) \\
 \text{EMS} &= \text{ESS} / (r-1)(t-1)
 \end{aligned}$$

The significance of the different variance components was assessed in comparison with F table (Snedecor and Cochran, 1967). After finding the significant differences between treatments, Critical Difference (CD) was used to find the significant treatments by the standard error pertaining to RBD analysis using the formulae,

$$SE_{(d)} = \sqrt{2EMS/r}$$

$$CD = t_{[\alpha, (r-1)(t-1)]} SE_{(d)}$$

Where,

$$SE_{(d)} = \text{Standard Error}$$

$$CD = \text{Critical Difference}$$

$$= \text{tabulated value of 't' @ } \alpha \text{ level of significance and } (r-1)(t-1) \text{ degrees of freedom}$$

3.4.3 Estimation of genetic parameters

The genetic parameters like genotypic variance, phenotypic variance, and genotypic coefficient of variation, phenotypic coefficient of variation, heritability and genetic advance were calculated as given below.

3.4.3.1 Variance

The phenotypic and genotypic variances were calculated by utilizing the mean sum of square values (MS_1 and MS_2) from variance table (Johnson *et al.*, 1955).

$$\text{Genotypic variance } (\sigma^2_g) = \frac{(MS_1 - MS_2)}{r}$$

$$\text{Phenotypic variance } (\sigma^2_p) = MS_1$$

$$\text{Environmental variance } (\sigma^2_e) = MS_2$$

3.4.3.2 Phenotypic and Genotypic coefficients of variation (PCV and GCV)

PCV and GCV were estimated using the formula suggested by Burton (1952) and expressed in percentage.

$$PCV (\%) = \frac{\sqrt{\sigma^2_p}}{\text{Mean}} \times 100$$

$$\text{GCV (\%)} = \frac{\sqrt{\sigma^2_g}}{\text{Mean}} \times 100$$

The estimates of PCV and GCV were categorized based on the scale given below (Sivasubramanian and Madhavmenon, 1973).

<15%	-	Low
15-30%	-	Medium
>30%	-	High

3.4.3.3 Heritability (h²)

It is the ratio of the genetic variance to the total variance. Heritability in broad sense (h²) was calculated according to Lush (1940) and expressed in percentage.

$$\text{Heritability in broad sense (h}^2\text{) (\%)} = \frac{\sigma^2_g}{\sigma^2_p} \times 100$$

The range of heritability was categorized as below, as suggested by Johnson *et al.* (1955).

0 – 30	-	Low
31 – 60	-	Medium
>61	-	High

3.4.3.4 Genetic advance (GA)

Genetic advance was worked out based on the formula suggested by Johnson *et al.* (1955).

$$\text{GA} = \frac{\sigma^2_g}{\sigma^2_p} \times k \times \sqrt{\sigma^2_p}$$

Where,

k = Selection differential at 5% selection intensity = 2.06 (Falconer, 1981).

$\sqrt{\sigma^2_p}$ = Phenotypic standard deviation.

3.4.3.5 Genetic Advance as percentage of mean

$$\text{GA \%} = \frac{\text{GA}}{\text{Mean}} \times 100$$

This was categorized, as below, as suggested by Johnson *et al.* (1955)

>20	- High
10–20	- Medium
<10	- Low

3.4.4 Association analysis

3.4.4.1 Estimation of correlation coefficient

Pearson's linear correlation coefficient (r) was calculated for all the combinations of traits. Since the trait 'degree of lodging' was recorded as a categorical data, Point biserial correlation coefficient (r_{pb}) (<http://vassarstats.net/pbcorr.html>) suggested by Das Gupta (1960) was used to estimate the correlation between degree of lodging and other continuous data. Microsoft office excel 2007 was used to draw the polynomial regression curves for different combination of traits to study the trend in their relationship.

Since the aim of this study is to identify traits related to culm strength, which is complex trait, governed by many factors, pulling force, which quantifiably measures culm strength, was given importance in association studies. The association of other culm strength related traits with pulling force and their inter-relationships assumes importance for selection and evaluation. The traits such as inflorescence length, raceme number and raceme length were excluded from association analysis to avoid ambiguity, as they are mainly related to yield.

Phenotypic correlation (r_p)

It is the association between two variables which was directly observed and calculated by

$$r_p = \frac{\text{Cov } p_{xy}}{\sqrt{\sigma^2_{px} \cdot \sigma^2_{py}}}$$

Where,

Cov p_{xy} = phenotypic covariance between two traits (x and y)
 σ^2_{px} =phenotypic variance of x
 σ^2_{py} =phenotypic variance of y

3.4.4.2 Path coefficient analysis

Phenotypic correlation coefficients were partitioned into direct and indirect effects using path analysis suggested by Dewey and Lu (1959). By keeping pulling force as dependent variable and other nine traits, viz., days to 50 per cent flowering, plant height, number of productive tillers, culm diameter, internode length, recovery angle after bending, days to maturity, grain yield per plant and test weight, as independent variables, simultaneous equations which expressed the basic relationship between path co-efficients, were solved to estimate the direct and indirect effects.

The direct and indirect effects were classified based on the following scale (Lenka and Mishra, 1973).

>1.0	-	Very high
0.30-1.00	-	High
0.20-0.29	-	Moderate
0.10-0.19	-	Low
0.00-0.09	-	Negligible

3.4.5 Diversity by multivariate analysis of quantitative traits

3.4.5.1 Hierarchical Cluster analysis (HCA)

The data on 14 quantitative traits for all the 41 genotypes raised during Summer, 2013 were subjected to multivariate hierarchical cluster analysis. The mean data over replications were computed. The traits which showed significant variation based on “F” test alone were standardized to unit variance. Distance matrix was constructed using Euclidean distance coefficients, e_{ij} . Cluster analysis was carried out for the morphological traits based on the genetic distance matrix of the 41

genotypes, applying Un-weighted Pair-Group Method using Arithmetic averages (UPGMA) method. The entire statistical analysis was done using DARwin 5.0 software (Perrier *et al.*, 2003; <http://darwin.cirad.fr/>).

3.4.5.2 Principal component analysis (PCA)

Principal component analysis (PCA) was also used to complement cluster analysis and to investigate patterns of genetic diversity. The PCA was done using the software MULTIBASE 2014 (www.numericaldynamics.com). The variance and cumulative percentage contribution to variation of different traits to eigen vector were also computed using the software.

3.4.6 Duncan's Multiple Range test (DMRT)

If 'F' ratio in ANOVA is significant, this implicates that two or more of the means are different. But, a multiple comparison test, like DMRT will indicate which genotype or treatment means are different from each other. The SEM elemental content data were evaluated by analysis of variance. Where the 'F' test was significant, means were separated by DMRT ($P \leq 0.05$). Results shown are means of five measurements in each tissue type per genotype tested.

Results

CHAPTER IV

RESULTS

The present investigation included 297 germplasm accessions of kodo millet and two checks (CO3 and *Adari*) for initial screening and selection of culm strength in un-replicated design and further evaluation of the selects in subsequent replicated trials to identify the genotypes which were consistent in their performance to suit mechanical harvesting. Biochemical, anatomical and molecular investigations were also conducted to characterize factors contributing to culm strength. The results of the present study are as follows:

4.1 Phenotyping and association studies in Kodo millet germplasm accessions for culm strength related traits [Kharif (Aug. - Dec.), 2012]

4.1.1 Mean performance of genotypes for 13 quantitative traits

The analysis of variance of treatments, checks, test entries (treatments excluding checks) and check *vs* test entries of 13 quantitative traits are presented in table 5. The check varieties differed significantly for all the traits except for internode length, pulling force, and grain yield per plant. Significant differences existed among the germplasm accessions for all the traits studied.

The mean performance of 297 germplasm accessions including checks is presented in table 6.

4.1.1.1 Days to 50 per cent flowering

Days to 50 per cent flowering ranged between 64 (Aamo 29) and 127 days (TNPsc 199) with an overall mean of 78.21 days. A total of 125 test entries were significantly earlier than the check, CO3 and 24 accessions were significantly longer in duration than the check CO3 (Table 6).

4.1.1.2 Plant height

Among the test entries, the genotypes, IPS 131 and GPUK 3 had the lowest plant height (20.33 cm). The maximum for this trait was recorded by *Thittakudi* (70.20 cm). The general mean recorded was 40.83 cm. Of the test entries, 192 accessions were significantly shorter than the check CO3 and only three lines, *viz.*, TNPsc 231, Aamo 5 and *Thittakudi* were significantly taller than CO3.

Table 5. ANOVA for 13 quantitative traits in 297 kodo millet germplasm accessions with two checks in ABD I [Kharif, (Aug.- Dec.), 2012]

Sl. No	Traits	Mean sum of squares				
		Treatments (df =298)	Checks (df =1)	Test entries (df =296)	Check x Test entries (df =1)	Error (df =6)
1	Days to 50 per cent flowering	83.05*	465.12**	77.42*	1369.13**	14.79
2	Plant height	110.62**	269.93**	101.79**	2565.39**	11.86
3	Numbers of productive tillers	6.77**	8.14*	6.78**	2.25	0.87
4	Inflorescence length	3.84*	34.56**	3.31*	127.83**	0.69
5	Raceme number	0.59*	34.98**	0.39	24.24**	0.12
6	Raceme length	0.80**	1.64**	0.74**	18.62**	0.06
7	Culm diameter	0.37*	0.75*	0.34*	7.65**	0.07
8	Internode length	5.77*	2.21	5.79*	3.13	0.84
9	Pulling force	1.30**	0.35	1.30**	2.35**	0.08
10	Recovery angle after bending	121.23*	153.12*	120.95*	172.79*	18.40
11	Days to maturity	91.05**	512.00**	84.31**	1664.03**	7.90
12	Grain yield per plant	62.72*	49.21	61.86*	331.04**	10.01
13	Test weight	0.30*	0.87*	0.26*	9.68**	0.06

** significant at 1% level; *significant at 5 % level

Table 6. Mean performance of 297 kodo millet germplasm accessions and two checks for 13 quantitative traits and four qualitative traits [Kharif, (Aug.- Dec.), 2012]

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
1	Aamo 4	72	52.60	7.80	11.30	3.80	5.90	2.94	3.54	11.83	45.00	102	13.83	4.30	1	1	1	5
2	Aamo 5	73	65.00	15.00	15.20	4.20	8.40	4.94	6.99	13.50	56.67	103	20.16	4.80	1	1	2	3
3	Aamo 6	72	61.00	17.20	13.40	3.60	5.90	2.92	4.71	12.83	55.00	105	16.57	3.85	1	1	1	7
4	Aamo 8	66	58.20	8.80	12.80	3.40	6.20	2.94	5.29	12.83	45.00	106	14.34	3.80	1	1	3	7
5	Aamo 10	65	44.40	7.60	9.80	3.40	6.10	3.15	4.47	12.00	60.00	102	11.39	4.48	3	1	3	7
6	Aamo 13	72	62.40	13.80	14.80	4.00	8.10	4.23	7.78	11.63	60.00	110	25.85	5.05	1	1	2	3
7	Aamo 14	77	49.40	7.80	11.20	3.80	5.70	2.82	4.96	10.83	45.00	116	12.35	4.33	1	1	1	5
8	Aamo 27	66	29.00	11.80	8.40	3.60	4.80	3.20	1.75	9.00	46.67	103	18.79	4.57	1	2	3	5
9	Aamo 29	64	48.20	18.20	10.80	3.80	5.20	2.62	1.95	15.17	36.67	103	23.36	3.70	1	1	1	7
10	Aamo 31	72	57.40	18.20	11.60	3.60	5.10	2.62	2.32	12.17	60.00	113	25.82	4.05	1	1	1	5
11	Aamo 32	66	55.20	10.40	12.70	4.00	5.20	3.24	1.60	15.17	45.00	108	11.60	4.43	1	1	1	5
12	Aamo 33	67	60.40	15.40	11.80	3.60	5.30	3.19	2.22	19.50	51.67	103	18.15	4.43	1	2	1	5
13	Aamo 36	66	48.20	10.20	11.80	3.80	6.40	3.33	2.30	15.33	43.33	103	18.32	4.52	1	2	3	7
14	Aamo 37	70	42.60	13.00	10.20	3.20	5.40	2.76	1.74	14.33	46.67	108	17.95	4.37	1	2	1	7
15	Aamo 58	66	51.00	12.75	10.75	3.20	4.75	3.70	3.51	15.67	53.33	103	16.07	4.10	1	1	1	5
16	Aamo 62	65	48.80	9.80	11.80	4.00	5.40	3.20	2.96	12.17	48.33	104	10.95	3.86	1	1	1	5
17	Aamo 68	72	62.00	15.20	11.00	3.80	5.50	3.30	3.31	16.00	45.00	109	17.31	4.07	1	2	1	5
18	Aamo 72	68	42.40	7.00	8.10	3.60	4.50	3.05	1.10	11.67	45.00	103	6.50	3.61	3	2	1	7
19	Aamo 73	65	39.60	20.00	9.00	3.60	5.70	2.77	2.40	8.50	46.67	103	20.88	4.06	3	2	1	3
20	Aamo 75	66	40.80	12.60	9.30	3.40	5.20	2.72	2.97	10.00	50.00	105	14.04	3.91	1	2	1	3
21	Aamo 79	71	58.40	16.00	12.60	4.00	5.50	3.42	3.72	12.00	55.00	113	25.36	4.31	1	1	1	3
22	Aamo 83	72	50.60	8.20	11.80	3.60	4.50	3.05	2.81	12.17	53.75	109	11.18	4.15	1	1	1	5
23	Aamo 89	74	39.80	9.00	12.00	3.40	5.50	2.64	2.96	10.17	58.33	112	11.87	3.89	1	2	1	5
24	Aamo 90	72	48.80	9.60	11.80	4.00	4.00	3.11	2.32	12.33	50.00	110	9.06	4.19	1	2	1	3
25	Aamo 96	73	35.20	15.40	9.60	3.20	5.50	2.82	2.21	11.17	58.33	113	17.82	3.67	1	1	1	3

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
26	Aamo 101	70	42.00	12.60	11.10	4.00	6.70	3.05	2.51	11.67	63.33	108	17.26	4.09	1	2	1	3
27	Aamo 109	79	46.20	11.60	10.20	3.80	6.00	3.03	2.01	12.50	63.33	117	14.83	3.85	1	2	1	3
28	Aamo 126	76	55.00	8.40	11.20	4.00	6.00	4.95	3.55	10.33	58.75	118	16.97	5.05	1	1	2	3
29	Aamo 134	79	39.60	9.20	11.40	4.00	5.80	3.11	3.46	11.00	60.00	119	15.35	3.84	1	2	1	3
30	Aamo 173	84	50.40	15.60	10.40	3.60	4.80	3.24	1.85	11.00	45.00	120	27.30	4.37	1	1	1	3
31	Aamo 179	70	25.00	9.60	10.00	3.80	5.20	3.00	2.65	5.67	71.67	103	15.44	4.56	1	1	3	3
32	Aamo 182	72	39.60	15.00	10.80	3.40	5.90	2.50	4.66	12.33	65.00	102	14.69	3.83	1	2	1	3
33	Aamo 203	73	38.20	8.60	9.00	3.40	4.40	2.59	2.57	10.50	61.67	115	16.49	4.25	1	2	1	3
34	Aamo 207	72	42.60	12.40	10.00	4.00	5.70	3.09	2.61	10.00	53.33	111	14.99	4.43	1	1	1	3
35	Aamo 208	74	49.40	12.80	11.40	4.00	5.10	2.72	2.38	11.00	51.67	113	14.13	4.29	1	2	1	3
36	Aamo 211	79	55.20	9.40	9.90	3.60	5.00	3.16	1.57	11.67	53.33	114	21.74	4.13	1	1	1	5
37	Aamo 213	84	43.60	8.60	8.40	3.40	4.50	2.61	1.71	9.83	58.33	118	11.70	4.07	1	2	1	5
38	Aamo 214	79	36.00	8.20	10.40	3.60	5.60	3.11	2.32	8.83	56.67	116	24.40	4.30	1	1	1	3
39	Aamo 215	80	37.00	7.20	10.54	7.20	6.00	2.90	1.98	8.50	51.67	116	14.81	3.98	1	1	1	3
40	Aamo 224	79	41.40	7.80	10.94	3.40	6.14	2.90	2.57	9.17	55.00	118	17.45	3.83	1	2	1	3
41	Aamo 243	78	44.20	9.40	12.20	3.80	6.64	2.90	1.56	11.00	48.33	118	18.97	4.10	1	1	1	5
42	Aamo 267	76	33.00	5.80	11.10	4.00	6.14	2.56	1.27	9.50	50.00	116	12.23	3.89	1	1	1	3
43	Aamo 268	78	46.20	7.00	11.20	3.60	5.80	3.29	2.10	10.67	53.33	118	7.77	4.11	1	1	1	5
44	Aamo 271	79	52.40	7.80	11.50	4.00	6.30	3.03	2.77	12.67	50.00	118	18.31	4.31	1	1	1	3
45	Aamo 275	76	49.00	8.40	12.30	3.80	6.40	2.88	3.04	10.83	50.00	118	17.30	4.60	1	1	1	3
46	Aamo 258	75	50.60	8.40	13.10	3.80	7.60	4.14	6.58	8.17	56.25	113	20.96	5.06	1	1	2	3
47	Aamo 280	81	41.80	8.40	11.20	3.40	6.20	2.83	2.92	10.83	45.00	116	19.39	4.08	1	1	1	3
48	Aamo 299	80	36.60	7.80	11.00	3.60	5.80	2.70	2.79	10.67	56.67	118	13.62	4.06	1	1	1	3
49	Aamo 303	81	51.20	8.00	11.70	4.00	5.80	2.69	3.69	10.50	50.00	118	12.79	4.07	1	1	1	3
50	Aamo 306	79	47.40	8.00	11.90	3.40	6.14	3.41	3.41	9.17	58.33	118	11.44	4.28	1	1	1	3
51	Aamo 307	75	43.80	7.60	11.00	3.60	6.70	3.09	3.33	7.83	63.33	120	15.46	4.33	1	1	2	3
52	Aamo 308	85	28.20	5.40	10.80	3.40	5.70	2.63	3.13	7.00	63.75	121	11.11	3.74	1	2	1	3

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
53	Aamo 309	85	49.80	7.00	10.40	3.40	5.30	2.33	2.29	9.50	51.67	130	18.83	3.91	1	2	1	3
54	Aamo 210	81	45.40	6.00	10.70	3.80	5.80	3.68	4.20	6.50	53.33	126	24.33	4.93	1	1	2	3
55	Aamo 311	85	42.60	8.40	10.50	3.60	5.60	3.02	3.80	8.83	51.67	130	21.54	4.03	1	1	1	3
56	Aamo 312	79	30.60	5.20	10.60	3.80	5.50	2.70	3.04	8.33	53.33	124	19.32	3.83	1	1	1	3
57	Aamo 316	85	25.80	6.20	9.30	3.20	5.20	2.77	3.28	6.33	66.67	130	7.68	3.93	1	1	1	3
58	Aamo 318	79	26.60	6.00	9.60	3.40	6.00	2.69	2.19	7.00	61.67	124	16.35	3.95	1	1	1	3
59	Aamo 322	78	24.00	5.00	8.33	3.33	4.67	2.67	1.97	3.25	60.00	124	5.98	3.52	1	1	1	3
60	Aamo 325	79	22.00	6.50	10.00	4.00	6.00	2.82	1.51	5.50	57.50	124	5.04	3.29	1	1	1	3
61	Aamo 326	98	23.00	6.00	9.00	3.00	6.00	1.72	1.60	5.50	60.00	143	7.15	3.76	1	1	1	3
62	Aamo 327	79	27.00	6.00	9.50	3.00	5.50	3.01	2.57	8.67	55.00	124	6.20	3.98	1	1	1	3
63	Aamo 330	80	32.67	6.33	9.67	3.67	5.33	2.77	2.07	8.33	58.33	124	11.93	3.74	1	1	1	3
64	Aamo 335	85	26.00	7.00	9.50	3.00	6.75	2.42	2.45	6.50	67.50	130	5.18	3.90	1	1	1	3
65	Aamo 337	84	26.00	7.00	9.50	4.00	5.50	2.98	2.03	6.00	55.00	130	7.60	4.03	1	1	1	3
66	Aamo 340	98	25.00	7.00	10.00	3.00	6.00	2.92	0.88	4.50	80.00	143	3.12	3.76	1	1	1	3
67	Aamo 345	85	34.00	6.50	10.50	3.00	5.50	2.53	1.34	6.00	52.50	130	8.07	4.08	1	1	1	3
68	Aamo 347	81	29.00	6.00	11.00	3.50	6.00	2.49	2.70	7.00	57.50	126	11.52	4.08	1	1	1	3
69	Aamo 352	85	26.33	6.00	11.67	3.33	6.00	2.59	1.54	7.33	65.00	130	12.52	4.08	1	1	1	3
70	Aamo 354	84	27.50	6.33	12.00	3.33	7.50	2.50	1.72	7.75	50.00	129	5.70	3.46	1	1	1	3
71	Aamo 355	85	30.67	7.00	12.17	3.67	6.33	2.60	0.91	9.33	57.50	129	13.31	3.84	1	1	1	3
72	Aamo 360	86	24.67	6.67	9.67	3.33	5.67	2.75	1.28	6.83	67.50	129	8.99	3.51	1	1	1	3
73	Aamo 362	98	33.67	5.00	12.00	4.00	6.67	2.80	1.52	6.00	65.00	129	11.92	4.32	1	1	1	3
74	Aamo 365	71	37.60	5.60	11.20	4.00	5.80	2.54	3.19	10.00	55.00	117	14.40	4.02	1	1	1	3
75	Aamo 366	72	39.00	5.60	11.60	3.40	6.40	2.01	2.75	9.67	45.00	117	10.65	4.68	1	1	1	5
76	Aamo 379	83	44.80	6.20	10.60	3.60	6.40	2.54	2.45	10.17	61.67	128	10.83	4.16	1	1	1	3
77	Aamo 382	84	51.00	5.80	12.00	3.80	5.80	2.75	2.21	10.67	48.33	128	25.84	4.18	1	1	1	5
78	Aamo 384	83	60.60	6.80	11.80	3.80	6.20	3.16	2.38	14.67	40.00	128	16.95	4.31	1	1	1	5
79	Aamo 385	70	38.00	6.40	10.60	3.80	6.00	2.34	0.88	9.67	50.00	115	19.73	4.17	1	2	1	3

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
80	Aamo 394	71	51.60	7.80	11.20	3.60	6.24	2.51	1.63	12.33	46.67	120	19.69	3.75	1	1	1	7
81	Aamo 398	72	52.60	8.20	11.00	3.40	6.20	2.46	2.11	14.17	40.00	117	21.10	4.20	1	1	1	7
82	Aamo 400	75	45.80	7.00	11.60	3.80	6.24	3.25	1.70	11.83	35.00	123	10.81	4.16	1	1	1	7
83	Aamo 408	72	54.20	8.00	11.10	4.20	5.90	2.37	2.34	15.33	45.00	129	17.95	4.16	1	1	1	5
84	Aamo 413	73	43.60	7.00	11.00	4.00	6.40	3.33	2.27	10.17	43.33	117	12.76	4.46	1	1	1	3
85	Aamo 414	72	52.60	7.60	11.80	3.40	6.20	2.85	2.96	11.50	50.00	118	12.00	4.64	1	2	1	5
86	Aamo 416	72	35.20	7.00	11.00	4.00	6.00	2.99	2.68	6.00	53.33	116	13.81	4.20	1	1	3	5
87	Aamo 422	74	43.00	7.20	10.80	3.40	6.04	2.68	4.49	9.63	63.33	117	14.61	4.41	1	1	1	3
88	Aamo 423	72	41.60	6.40	10.60	3.60	6.20	2.56	2.27	11.33	46.67	115	17.49	4.07	1	1	1	5
89	Aamo 463	77	37.00	7.00	11.00	3.60	5.70	2.72	3.25	10.50	53.33	122	25.13	3.94	1	1	1	3
90	Aamo 536	78	53.80	7.20	11.60	3.40	6.04	3.25	2.86	11.67	53.33	123	32.19	4.39	1	1	1	5
91	Aamo 591	77	35.40	7.60	10.80	3.60	5.60	2.48	2.81	12.00	35.00	123	9.97	3.74	1	2	1	3
92	Aamo 592	78	40.80	6.20	11.40	4.00	6.10	2.37	3.30	8.67	66.67	123	21.98	4.01	1	2	1	5
93	Aamo 593	72	32.40	6.00	11.20	3.40	6.00	2.33	1.86	10.17	65.00	117	13.97	3.88	1	2	1	3
94	Aamo 594	73	39.60	6.20	11.20	3.60	6.20	2.06	2.34	12.13	56.67	118	11.44	3.96	1	1	1	3
95	Aamo 595	70	38.20	7.20	10.80	3.80	6.10	2.57	2.67	10.00	65.00	117	12.98	4.06	1	2	1	3
96	Aamo 596	72	51.40	5.60	11.20	3.40	5.80	2.75	2.89	12.67	55.00	119	15.51	3.89	1	1	1	5
97	Aamo 597	74	56.80	6.40	12.00	3.80	6.30	3.34	2.07	12.17	50.00	117	26.88	4.38	1	1	1	5
98	RK 5	114	40.75	5.00	16.75	2.50	8.00	3.18	3.52	8.00	66.67	154	9.13	5.28	1	1	1	5
99	RK 25	72	38.80	6.00	12.00	4.00	6.54	2.22	3.37	11.83	58.33	117	32.76	4.03	1	1	1	3
100	RK 25/1	70	29.20	6.00	10.40	3.40	5.20	2.48	3.52	5.83	58.33	115	15.98	4.21	1	1	2	3
101	RK 25/2	78	41.60	6.20	12.00	3.40	6.70	3.24	1.62	10.83	41.67	124	18.91	4.34	1	1	1	5
102	RK 31	79	29.60	6.00	11.50	3.50	6.13	2.77	2.19	9.50	53.33	124	13.94	4.39	1	1	1	3
103	RK 50	80	55.20	8.00	21.00	5.60	8.30	3.85	3.53	9.50	50.00	125	31.49	6.03	1	1	1	3
104	RK 51	79	45.60	5.00	10.80	3.40	5.60	2.84	3.30	9.83	52.50	125	23.43	4.16	1	1	1	3
105	RK 51/1	81	30.80	5.80	11.40	3.80	5.80	2.85	1.62	9.50	53.33	125	15.87	5.03	3	1	1	3
106	RK 65-18	72	43.60	5.60	12.20	3.80	6.24	2.55	2.72	11.00	65.00	117	19.74	4.77	3	1	1	3

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
107	RK 73	81	54.00	4.80	12.40	3.80	5.90	2.93	2.22	8.83	45.00	126	25.69	4.19	1	1	1	5
108	RK 79	75	39.00	6.40	12.00	3.60	6.00	2.70	3.66	10.33	61.67	120	14.29	3.87	1	1	1	3
109	RK 80	72	37.40	5.60	11.20	3.80	6.00	2.67	2.32	9.38	73.33	117	27.60	4.02	1	1	1	3
110	RK 82	72	37.40	5.60	9.40	3.80	5.70	2.93	1.73	9.00	53.33	117	19.97	4.03	1	2	3	5
111	RK 84	80	43.80	6.60	12.80	3.40	6.60	3.46	4.00	7.67	51.67	125	27.50	4.16	1	1	1	3
112	RK 92	82	33.80	6.40	10.40	3.80	5.80	3.37	2.07	9.83	58.33	127	16.61	3.88	1	1	1	5
113	RK 96	100	43.00	4.40	12.20	2.80	7.00	3.00	2.70	9.83	58.33	145	14.60	4.02	1	1	1	3
114	RK 96/1	70	29.60	5.00	11.20	3.20	5.20	2.20	2.17	7.33	66.67	115	19.48	4.51	1	1	3	3
115	RK 111	79	53.40	5.40	11.40	3.00	6.00	2.83	2.38	11.17	53.75	125	26.51	4.09	1	1	1	3
116	RK 162	80	47.20	4.80	10.40	3.80	5.80	2.87	2.02	9.83	61.67	125	21.58	3.99	1	1	1	5
117	RBK 73	79	34.60	5.20	12.00	3.40	6.40	2.90	1.68	7.67	61.67	124	19.61	4.16	1	1	1	3
118	RBK 155	72	31.80	4.00	11.20	3.80	5.50	2.11	0.90	10.83	58.33	117	12.94	3.36	1	1	1	3
119	RBK 155/1	81	46.40	4.80	11.00	3.80	5.90	2.53	2.37	7.17	50.00	126	16.44	4.34	1	1	1	3
120	ICK 76	85	30.67	6.00	10.00	3.00	5.50	2.41	2.33	6.83	58.33	130	4.47	3.71	1	1	1	3
121	ICK 86	79	50.60	5.80	11.60	3.40	6.00	2.91	2.05	10.50	56.67	124	15.50	4.60	1	1	1	3
122	ICK 1042	72	30.80	7.20	11.80	3.80	6.44	2.85	1.47	7.17	58.33	117	19.67	4.43	1	1	1	3
123	ICK 7114	85	42.80	5.60	11.00	3.80	5.90	2.96	2.06	9.17	56.67	130	18.82	4.00	1	1	1	3
124	IPS 102	75	39.20	5.60	12.50	4.00	6.60	2.84	1.49	10.88	58.33	120	21.96	3.74	1	1	1	3
125	IPS 113	70	21.00	5.80	9.50	3.75	5.75	2.90	2.60	6.33	61.67	117	16.58	4.13	1	1	3	3
126	IPS 115	72	22.50	4.20	9.40	3.40	5.60	3.03	3.57	7.17	80.00	117	11.59	4.23	1	1	3	3
127	IPS 117	71	23.00	4.00	9.80	3.20	5.50	2.63	3.32	5.17	71.67	117	6.69	4.12	1	1	3	3
128	IPS 118	72	31.67	5.00	11.00	4.00	6.00	3.32	3.21	5.83	76.67	117	13.55	4.04	1	1	2	3
129	IPS 122	79	33.60	4.60	11.00	3.60	5.80	3.14	2.24	7.83	73.33	124	10.90	4.72	1	1	3	3
130	IPS 123	75	30.80	5.60	12.40	3.60	6.08	3.61	2.97	5.73	75.00	120	16.79	4.54	1	1	2	3
131	IPS 125	72	34.50	4.80	12.00	3.20	5.80	3.23	1.45	6.17	66.67	117	10.66	4.70	1	1	2	3
132	IPS 129	75	26.33	3.75	10.50	3.25	5.68	3.27	1.13	7.00	75.00	120	6.12	4.28	1	1	2	3
133	IPS 131	72	20.33	4.00	10.75	3.00	5.63	2.68	2.30	6.00	70.00	117	6.89	3.85	1	1	3	3

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
134	IPS 147	85	23.00	4.50	9.00	4.00	5.50	2.24	3.95	5.83	75.00	130	4.72	4.09	1	1	1	3
135	IPS 160	83	27.67	5.67	11.00	3.33	5.73	2.70	3.07	5.33	67.50	128	6.78	4.12	1	1	1	3
136	IPS 194	100	30.00	7.00	11.00	2.00	5.00	1.85	1.84	5.75	65.00	145	1.11	2.75	1	1	1	3
137	DPS 95	82	25.33	5.00	10.67	3.33	5.33	2.53	1.92	5.75	67.50	127	3.32	4.03	1	1	1	3
138	DPS 368	85	30.00	4.67	12.33	3.67	6.33	2.24	2.01	7.67	75.00	129	6.98	3.81	1	1	1	3
139	DPS 637	87	29.67	4.33	11.67	3.00	5.67	2.47	1.15	8.00	67.50	129	7.77	3.86	1	1	1	3
140	DPS637/1	83	22.67	3.33	10.67	3.00	5.33	2.01	1.42	7.00	77.50	129	4.57	3.48	1	1	1	3
141	GPUK 3	85	20.33	4.00	10.33	3.33	5.33	2.49	1.54	3.50	70.00	129	6.92	3.93	1	1	3	3
142	GPUK 72	83	28.33	6.00	11.33	3.67	6.00	3.00	1.60	6.83	77.50	129	12.68	4.42	1	1	1	3
143	APK 1	72	37.00	7.33	12.67	3.33	7.00	2.76	2.17	7.17	60.00	129	12.63	4.48	1	1	1	3
144	Sel 1	72	32.33	6.67	12.67	3.67	6.00	2.87	2.10	8.00	70.00	129	10.32	4.24	1	1	1	3
145	Sel 2	75	34.33	5.67	12.33	3.00	6.67	3.05	1.71	7.83	62.50	129	8.69	4.26	1	1	1	3
146	Sel 3	79	29.33	5.67	11.67	3.33	6.33	2.54	1.64	7.33	72.50	129	11.20	3.51	1	1	1	3
147	Sel 4	81	24.33	4.67	12.00	4.00	6.33	2.83	3.72	5.17	65.00	129	6.15	3.85	1	1	1	3
148	Sel 5	72	43.20	6.40	10.80	3.00	6.20	2.91	1.69	10.50	45.00	117	14.59	4.34	1	1	3	5
149	Sel 6	72	50.20	6.40	12.60	2.80	6.24	2.90	1.19	11.50	33.33	117	23.31	4.24	1	1	1	5
150	Sel 7	79	38.80	4.80	12.40	3.00	7.00	2.73	2.16	9.33	58.33	124	24.75	3.80	1	1	1	3
151	Sel 8	79	52.00	5.00	12.40	3.00	7.24	2.88	2.61	10.33	43.33	125	16.67	4.13	1	1	1	5
152	Sel 9	80	45.20	5.00	12.80	3.40	6.20	3.09	2.33	10.17	43.33	125	16.91	4.04	1	1	1	3
153	Sel 10	72	40.20	6.40	10.80	3.20	6.50	2.66	1.54	10.67	63.33	117	10.81	4.21	1	1	2	5
154	Sel 11	74	47.20	5.40	12.00	2.60	6.12	3.13	2.93	11.75	33.33	117	16.11	4.55	1	1	1	5
155	Sel 12	72	42.60	5.80	11.40	2.40	6.24	2.59	1.79	12.50	53.33	117	13.15	3.86	1	1	1	5
156	Sel 13	73	60.40	6.00	15.80	3.20	8.00	3.85	4.01	10.00	60.00	117	21.98	4.42	1	1	1	5
157	Sel 14	72	45.60	5.00	11.80	2.20	6.60	2.80	2.39	13.50	45.00	117	14.38	4.03	1	1	1	5
158	Sel 15	71	30.00	7.00	8.60	3.00	5.20	2.81	3.78	7.63	70.00	117	21.33	4.70	1	1	2	3
159	Sel 16	72	39.40	5.20	11.60	3.00	6.10	2.93	3.19	11.17	50.00	117	18.38	4.34	1	1	1	5
160	Sel 17	81	43.80	6.20	11.80	2.40	6.40	2.81	2.29	10.50	40.00	126	37.39	3.97	1	1	1	5

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
161	Sel 18	72	25.40	6.40	8.90	3.00	5.40	2.55	2.19	6.38	58.33	117	13.67	4.26	1	1	3	3
162	Sel 19	72	39.20	5.40	9.50	3.20	5.80	2.79	2.76	8.83	50.00	121	30.31	4.37	1	1	3	7
163	Sel 20	70	24.60	6.80	9.30	2.80	5.80	2.63	3.35	8.00	76.67	115	12.89	4.58	1	1	3	3
164	Sel 21	72	48.40	5.40	10.60	3.20	6.50	4.01	2.43	9.17	31.67	112	38.05	5.55	1	1	2	5
165	<i>Thittakudi</i>	83	70.20	8.20	19.60	3.60	7.64	4.02	2.32	11.00	41.67	128	18.70	5.91	1	1	1	5
166	<i>Vridhachalam</i>	81	42.67	7.00	14.67	2.67	7.00	3.27	1.86	11.33	46.67	126	14.59	4.83	1	1	3	3
167	TNPSc 81	98	55.50	7.50	15.63	5.50	7.75	4.40	3.99	4.50	40.00	129	28.55	5.98	1	1	1	3
168	TNPSc 82	72	27.20	7.50	10.70	3.00	5.60	2.67	4.05	8.00	63.33	117	21.34	4.39	1	1	3	3
169	TNPSc 83	73	26.60	7.20	9.50	2.80	6.00	2.79	2.51	7.50	60.00	117	15.77	4.09	1	1	1	3
170	TNPSc 84	72	25.20	7.00	10.10	2.80	5.60	3.02	1.33	6.75	61.67	117	21.29	4.32	1	1	3	3
171	TNPSc 85	70	28.60	7.60	11.80	3.00	6.10	2.46	2.78	7.00	65.00	118	21.35	4.71	1	1	3	3
172	<i>Pudukooraipettai</i>	71	48.00	7.60	13.60	2.80	6.50	2.48	2.13	11.17	36.67	118	23.87	4.66	1	1	1	7
173	TNPSc 90	70	36.80	7.00	10.80	2.20	6.10	2.50	2.61	8.63	55.00	120	27.08	4.47	1	1	1	5
174	TNPSc 91	72	28.60	5.60	10.80	2.20	6.10	2.33	2.22	7.00	70.00	118	26.45	3.88	1	1	1	3
175	TNPSc 92	73	35.40	6.00	11.20	2.60	5.86	2.52	1.57	11.00	43.33	118	29.75	3.99	1	1	1	5
176	TNPSc 93	72	39.00	6.60	10.70	3.00	6.00	2.73	2.22	9.83	56.67	121	16.16	4.24	3	1	3	5
177	TNPSc 94	70	30.60	8.20	10.80	3.20	6.06	2.73	2.15	7.17	53.33	116	23.12	4.44	1	1	2	3
178	TNPSc 95	69	39.80	7.40	10.60	2.60	6.30	2.98	1.83	11.17	48.33	106	19.84	4.23	3	1	3	5
179	TNPSc 96	82	35.60	7.00	10.80	2.80	6.04	2.08	1.82	9.00	51.67	121	22.57	3.91	1	1	1	3
180	TNPSc 97	79	46.80	7.40	10.80	3.00	6.00	2.40	2.22	16.33	43.33	121	20.50	3.90	1	1	1	5
181	TNPSc 98	71	42.00	7.60	11.80	2.60	7.10	2.75	1.21	11.00	45.00	121	19.32	3.99	1	1	1	5
182	TNPSc 99	70	38.00	5.60	11.00	2.60	5.70	2.68	2.06	11.50	42.50	121	10.99	3.77	1	1	1	5
183	TNPSc 100	72	44.00	7.60	12.20	2.80	6.04	2.90	2.12	12.13	47.50	121	25.36	4.61	1	1	1	5
184	TNPSc 101	97	43.00	7.20	13.60	2.80	7.80	3.47	2.42	9.38	52.50	128	11.18	5.55	1	1	1	5
185	TNPSc 102	70	49.60	7.60	12.60	3.20	6.44	3.02	2.41	12.33	60.00	121	22.92	4.11	1	1	1	5
186	Aamo 30	80	48.80	7.80	12.20	3.00	6.40	2.81	1.48	12.67	42.50	119	27.75	4.29	1	1	1	5
187	TNPSc 104	80	39.80	6.20	11.20	2.80	6.40	2.67	1.20	11.50	67.50	116	20.67	3.71	1	1	1	3

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
188	TNPSc 105	78	38.20	6.40	11.60	3.00	6.20	2.30	1.67	10.83	47.50	121	16.02	3.72	1	1	1	5
189	TNPSc 106	72	40.20	6.40	11.40	2.40	6.00	2.40	1.17	13.67	37.50	113	15.30	4.08	1	1	1	7
190	TNPSc 107	77	41.80	7.80	9.60	3.25	5.00	2.97	1.61	9.50	55.00	121	23.00	3.76	1	1	1	5
191	TNPSc 108	72	37.60	6.00	11.20	2.60	6.60	2.47	2.00	11.13	65.00	110	24.01	3.94	1	1	1	5
192	TNPSc 109	70	39.60	7.00	11.80	6.80	6.80	3.07	2.69	11.17	50.00	116	19.75	4.55	1	1	1	5
193	TNPSc 110	73	40.40	6.60	10.60	2.80	6.20	2.25	1.51	12.67	40.00	113	12.92	3.91	1	1	1	5
194	TNPSc 111	70	33.60	5.00	10.00	2.80	6.00	2.22	2.20	11.50	67.50	118	11.32	3.80	1	1	1	5
195	TNPSc 112	71	37.80	6.00	10.80	2.80	6.40	2.17	2.10	11.00	75.00	121	16.82	4.20	1	2	1	5
196	TNPSc 113	85	51.20	6.20	13.60	3.60	6.80	3.58	2.02	10.33	35.00	131	15.05	5.11	1	1	1	5
197	TNPSc 114	84	40.40	4.40	10.60	2.40	5.80	2.66	1.68	10.17	47.50	129	11.31	3.83	1	1	1	5
198	TNPSc 115	85	43.60	5.40	12.00	2.80	6.00	4.02	2.35	7.75	45.00	130	12.39	5.55	1	1	1	3
199	TNPSc 116	86	47.60	5.60	14.20	3.40	7.30	3.68	1.75	8.00	42.50	130	14.48	5.37	1	1	1	5
200	TNPSc 117	85	37.80	4.80	11.60	3.00	6.30	3.40	1.58	7.25	70.00	130	8.98	5.02	1	1	1	3
201	TNPSc 118	95	46.67	6.00	14.67	3.33	7.83	3.95	2.57	8.17	47.50	140	16.28	5.01	1	1	1	3
202	TNPSc 119	80	39.60	4.20	11.20	2.60	5.90	2.39	1.28	11.75	40.00	125	13.41	3.76	1	1	1	5
203	TNPSc 120	79	38.40	5.20	11.40	3.00	6.44	2.80	1.97	8.88	55.00	125	10.97	3.64	1	1	1	5
204	TNPSc 121	81	38.20	4.40	12.80	2.80	6.80	3.22	2.28	10.17	65.00	125	8.77	4.62	1	1	1	3
205	TNPSc 122	82	31.80	3.40	7.60	3.00	3.80	2.06	1.44	8.00	70.00	127	4.32	3.70	1	1	1	3
206	TNPSc 123	78	29.67	5.67	9.33	2.00	5.00	2.78	2.45	7.25	67.50	123	11.36	4.14	1	1	3	3
207	TNPSc 124	79	28.67	6.33	11.00	3.00	5.17	3.05	2.19	6.17	60.00	123	14.05	3.79	1	1	2	3
208	TNPSc 125	100	32.00	5.00	13.00	4.00	6.00	2.93	0.86	7.50	30.00	145	7.77	5.19	3	2	1	3
209	TNPSc 126	99	27.33	4.67	8.67	2.00	5.00	1.93	1.45	6.83	57.50	145	7.36	3.72	1	1	1	3
210	TNPSc 127	80	36.33	7.00	14.33	3.00	7.50	2.99	2.26	9.50	55.00	125	14.66	4.99	1	1	3	3
211	TNPSc 128	78	28.67	5.00	13.00	2.33	7.00	2.71	2.19	7.13	60.00	123	11.97	4.19	1	1	1	3
212	TNPSc 129	80	30.33	5.67	11.33	2.33	6.17	2.44	1.04	8.03	67.50	125	13.73	3.61	1	1	1	3
213	TNPSc 130	80	30.33	5.33	9.75	3.33	5.83	2.39	2.27	7.67	52.50	125	16.94	3.84	1	1	1	3
214	TNPSc 131	98	36.00	6.00	15.50	3.00	8.00	2.41	1.32	10.75	67.50	143	15.19	4.96	1	1	1	3

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
215	TNPSc 133	82	33.33	5.67	14.33	3.00	6.17	3.03	2.25	5.67	70.00	127	13.88	3.90	1	1	1	3
216	TNPSc 134	85	29.33	5.00	12.00	2.33	5.83	2.36	3.09	7.50	65.00	130	12.75	3.80	1	1	1	3
217	TNPSc 135	85	34.50	6.50	13.00	2.50	7.50	2.68	0.92	8.00	75.00	130	3.21	4.22	3	1	1	3
218	TNPSc 136	78	34.33	7.00	11.00	3.33	6.33	3.30	0.93	8.83	47.50	123	13.84	4.36	3	1	2	3
219	TNPSc 137	81	37.00	6.50	13.50	3.00	7.00	3.50	2.92	10.00	40.00	126	26.42	4.19	3	1	1	3
220	TNPSc 138	70	33.20	6.00	11.60	3.80	6.40	3.07	2.59	8.00	60.00	118	11.39	4.19	1	1	3	3
221	TNPSc 139	73	32.60	5.20	10.75	3.50	5.50	2.50	1.46	9.00	47.50	120	15.02	3.87	1	1	1	3
222	TNPSc 140	72	32.60	5.60	11.40	4.60	6.00	2.83	3.06	9.33	72.50	118	20.65	4.40	3	1	3	3
223	TNPSc 141	69	34.80	5.00	11.00	4.20	6.10	2.70	3.31	8.33	50.00	116	21.62	4.45	1	1	1	3
224	TNPSc 142	70	32.20	5.00	11.20	3.80	5.40	2.61	2.96	7.17	60.00	121	16.21	3.44	1	1	1	3
225	TNPSc 143	69	44.00	5.60	12.60	4.20	6.70	2.53	1.64	9.17	45.00	116	21.61	4.75	1	1	3	5
226	TNPSc 144	70	31.40	5.20	11.40	3.40	5.40	2.37	1.25	8.17	42.50	121	20.41	4.01	3	2	1	5
227	TNPSc 145	71	37.00	7.60	11.80	3.60	6.00	2.33	1.95	8.75	47.50	110	24.31	4.08	1	1	1	7
228	TNPSc 146	70	42.20	7.40	11.80	4.20	5.90	2.80	1.43	11.17	37.50	121	13.97	4.13	1	1	1	5
229	TNPSc 147	82	52.60	8.40	12.80	4.40	7.00	3.27	3.27	8.50	55.00	129	15.73	5.54	1	1	1	5
230	TNPSc 148	70	35.40	5.80	10.80	4.00	5.80	2.88	2.40	8.83	57.50	108	25.69	4.28	3	1	3	5
231	TNPSc 149	82	52.40	7.00	14.80	4.20	7.80	3.23	3.15	9.83	62.50	129	10.13	5.25	1	1	1	5
232	TNPSc 150	70	31.60	6.00	10.80	4.60	6.20	2.56	3.48	9.67	52.50	110	15.50	4.17	1	1	3	5
233	TNPSc 151	71	32.80	6.60	10.40	4.00	5.40	2.64	1.72	8.00	70.00	113	15.99	4.25	1	1	2	3
234	TNPSc 153	70	38.20	5.60	10.20	4.00	5.40	2.60	1.10	8.83	55.00	121	11.83	3.53	1	1	1	3
235	TNPSc 154	82	33.00	7.00	11.00	3.00	6.00	2.56	1.82	8.50	60.00	118	7.47	3.83	1	1	1	3
236	TNPSc 155	97	53.20	7.60	15.00	4.00	7.60	5.14	7.36	8.50	55.00	142	32.88	5.25	1	1	1	3
237	TNPSc 156	98	57.80	7.00	15.20	4.40	8.20	4.97	4.35	7.33	25.00	123	17.35	5.06	1	1	1	7
238	TNPSc 157	70	39.20	6.80	10.80	4.20	5.50	2.38	1.66	8.33	48.33	118	20.18	3.74	1	1	1	5
239	TNPSc 158	72	35.80	6.40	12.20	3.40	6.20	3.04	1.77	10.50	32.50	113	10.47	4.08	1	1	1	3
240	TNPSc 159	70	36.60	7.40	11.40	3.80	5.80	2.48	2.44	8.00	55.00	116	5.49	4.20	1	1	1	3
241	TNPSc 160	71	34.80	8.00	12.40	4.00	6.20	3.02	4.21	8.00	80.00	121	14.03	4.21	3	2	2	3

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
242	TNPSc 161	70	38.40	6.60	11.60	3.40	6.20	2.42	2.71	9.00	37.50	116	21.35	3.76	1	1	1	5
243	TNPSc 162	81	45.00	7.00	11.80	3.80	6.40	2.72	1.93	11.17	57.50	116	32.50	4.47	1	1	1	5
244	TNPSc 163	70	30.80	6.00	11.00	3.40	6.00	2.70	1.11	10.17	22.50	113	13.63	4.13	1	1	1	5
245	TNPSc 164	70	44.20	9.20	12.20	3.80	6.20	3.07	2.41	8.67	27.50	109	57.55	4.20	3	1	1	7
246	TNPSc 165	72	38.00	8.00	11.40	3.60	5.90	2.68	3.04	8.67	62.50	108	21.39	3.73	3	1	1	7
247	TNPSc 166	71	37.80	7.40	11.40	3.40	5.80	2.15	1.66	8.17	52.50	113	14.68	3.84	1	1	1	7
248	TNPSc 167	70	33.00	7.20	11.20	4.00	6.00	1.99	2.91	8.33	50.00	111	18.21	3.76	3	2	1	5
249	TNPSc 168	73	33.60	8.20	11.20	3.40	6.20	2.17	2.38	8.50	52.50	120	21.16	4.42	1	1	1	3
250	TNPSc 169	70	37.60	7.80	11.20	3.60	6.40	1.85	2.78	7.33	42.50	115	24.98	4.49	3	1	1	5
251	TNPSc 170	70	41.00	7.40	13.80	3.40	7.40	2.65	1.61	9.38	20.00	113	25.69	3.97	1	1	1	7
252	TNPSc 171	72	50.60	7.00	12.20	3.40	6.60	3.54	1.40	10.75	30.00	118	33.06	4.64	1	2	1	5
253	TNPSc 172	76	50.20	7.40	11.40	4.00	5.60	3.08	3.93	8.17	57.50	118	35.75	4.03	1	2	1	5
254	TNPSc 173	82	38.40	6.20	11.40	3.60	5.80	2.75	3.38	9.67	72.50	121	18.37	3.75	1	1	1	3
255	TNPSc 174	82	54.00	6.40	12.00	3.00	6.00	2.90	2.14	8.63	55.00	118	30.22	4.52	1	1	1	3
256	TNPSc 175	79	42.60	5.20	13.00	3.80	6.80	2.37	2.48	11.00	67.50	117	26.62	3.89	1	1	1	5
257	TNPSc 176	81	51.60	7.80	11.80	3.80	7.00	3.74	4.71	8.25	50.00	115	34.86	5.73	1	1	3	3
258	TNPSc 177	72	42.00	6.40	11.20	3.40	6.40	2.63	1.61	9.38	45.00	115	19.26	4.70	1	1	1	3
259	TNPSc 178	72	35.20	6.20	11.40	3.60	6.00	1.76	1.70	7.25	55.00	116	14.82	4.24	1	1	1	3
260	TNPSc 179	72	42.80	4.80	11.00	3.40	5.60	2.29	1.22	6.33	50.00	122	13.00	4.21	1	1	1	3
261	TNPSc 180	85	50.20	6.40	12.20	3.00	6.60	2.84	1.46	9.38	45.00	120	24.24	4.70	1	2	1	5
262	TNPSc 181	70	32.00	6.60	11.20	3.60	5.80	2.40	2.30	8.00	45.00	122	18.54	4.15	1	1	1	5
263	TNPSc 182	70	43.20	5.80	11.20	4.00	6.34	2.19	1.17	7.33	60.00	121	14.12	4.40	1	1	1	3
264	TNPSc 183	83	61.80	7.00	13.80	3.50	8.20	3.24	2.83	8.30	65.00	115	40.44	5.22	1	1	1	3
265	TNPSc 184	78	53.20	6.60	11.80	3.40	5.80	3.21	3.51	13.13	42.50	116	27.58	4.41	1	2	1	5
266	TNPSc 185	77	59.20	6.00	13.00	3.20	6.60	2.96	2.21	13.80	50.00	116	29.13	4.31	1	2	1	5
267	TNPSc 186	70	51.20	5.80	12.00	3.60	6.60	2.53	2.52	10.00	60.00	115	9.39	4.46	1	2	1	5
268	TNPSc 187	80	53.80	7.40	11.80	3.40	6.40	2.88	2.57	12.13	65.00	118	42.40	4.35	1	2	1	5

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
269	TNPSc 188	72	48.20	5.20	12.60	3.40	6.40	3.09	1.87	12.50	60.00	121	33.29	4.21	1	2	1	5
270	TNPSc 189	85	52.00	6.20	12.60	3.00	6.60	3.00	1.70	10.75	32.50	122	28.89	4.31	1	2	1	3
271	TNPSc 190	82	50.20	7.40	12.40	3.80	6.20	2.41	2.07	11.67	50.00	122	18.52	4.01	1	2	1	3
272	TNPSc 191	78	42.40	5.60	12.40	3.40	6.80	2.67	2.19	10.38	50.00	122	39.78	4.13	3	2	1	3
273	TNPSc 192	78	34.40	4.20	11.00	3.60	6.20	2.66	1.09	8.88	57.50	123	10.87	4.08	3	2	1	3
274	TNPSc 193	84	40.00	7.20	11.80	3.80	7.60	2.93	1.64	8.25	42.50	129	21.00	4.26	1	1	1	3
275	TNPSc 194	81	38.80	4.00	12.00	4.00	5.90	2.31	1.51	9.63	47.50	122	17.45	4.26	1	1	1	3
276	TNPSc 195	82	43.00	5.40	12.60	3.20	6.10	2.13	1.96	10.33	50.00	122	31.11	4.33	1	1	1	3
277	TNPSc 196	80	44.80	5.00	13.20	3.20	6.50	2.39	2.22	11.00	62.50	125	20.64	4.31	1	1	1	3
278	TNPSc 197	81	41.25	4.80	12.40	3.40	6.60	2.62	2.23	10.83	67.50	122	20.56	4.17	1	1	1	3
279	TNPSc 198	84	35.20	5.00	11.80	3.60	5.80	2.39	1.43	9.88	70.00	122	10.73	3.98	1	1	1	3
280	TNPSc 199	127	35.00	11.00	15.00	2.00	9.00	4.49	4.76	7.00	62.50	172	12.71	3.85	3	1	1	3
281	TNPSc 201	85	41.33	6.67	13.33	3.00	8.00	4.79	6.67	8.50	75.00	129	12.44	4.47	1	1	3	3
282	TNPSc 202	94	48.50	10.50	18.00	3.50	8.50	4.57	4.71	8.00	57.50	139	20.56	5.09	1	1	1	3
283	TNPSc 203	100	51.50	12.00	16.50	3.00	8.25	3.59	5.80	8.50	52.50	139	23.10	5.57	1	1	1	3
284	TNPSc 204	99	58.00	14.50	17.50	3.00	7.50	3.94	4.77	4.33	50.00	144	20.82	4.92	3	1	1	3
285	TNPSc 205	98	57.00	10.50	17.50	3.00	8.50	4.02	6.37	6.67	57.50	142	23.76	4.83	3	1	1	3
286	TNPSc 206	99	55.00	8.50	16.50	3.00	8.25	5.00	5.34	6.38	57.50	142	14.40	5.02	3	1	1	3
287	TNPSc 211	85	30.00	7.00	13.00	4.00	6.00	3.26	1.42	4.50	60.00	130	7.75	4.65	1	1	1	3
288	TNPSc 215	94	55.60	5.00	16.60	3.40	9.00	5.45	2.25	6.75	50.00	129	11.19	5.92	1	1	1	5
289	TNPSc 217	97	57.20	5.00	14.00	4.80	7.40	4.45	3.90	7.50	50.00	129	25.83	6.06	2	1	1	3
290	TNPSc 221	80	49.75	8.25	15.00	5.25	7.50	3.55	5.32	7.75	47.50	125	45.00	5.69	1	1	1	5
291	TNPSc 225	80	58.60	6.20	14.00	4.80	7.00	3.85	3.83	8.50	57.50	123	25.23	5.31	1	1	1	3
292	TNPSc 230	80	47.00	10.50	14.50	5.00	8.75	3.83	3.49	8.50	45.00	125	22.82	5.88	2	2	1	5
293	TNPSc 231	98	64.33	11.33	19.33	3.00	10.00	3.70	4.93	9.83	70.00	129	27.12	5.12	1	1	1	3
294	TNPSc 232	99	52.80	6.40	13.00	4.20	7.24	4.00	3.81	8.17	50.00	144	12.63	4.87	1	1	1	3
295	TNPSc 233	101	43.50	8.50	14.50	3.50	7.25	3.39	2.45	9.50	52.50	129	21.55	5.52	1	1	1	5

Table 6 continued...

Sl.No.	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		DF	PH	Ptil	InfloL	RN	RL	CD	PF	IntL	RA	DM	GY	TW	PT	EE	SA	DL
296	TNPSc 235	101	55.00	11.00	12.75	5.50	8.25	3.37	2.54	8.33	35.00	129	15.61	5.12	1	1	1	5
297	TNPSc 236	85	61.40	7.00	12.00	4.00	7.00	3.78	4.95	7.17	52.50	130	17.16	4.53	1	1	1	5
Check	CO3	83.75	53.05	6.65	13.68	3.15	7.30	3.60	3.29	9.27	45.83	127.75	21.38	5.72	1	1	1	5
Check	<i>Adari local</i>	99.00	64.67	8.67	17.83	7.33	8.21	4.21	2.87	8.22	54.58	143.75	26.34	5.06	1	1	1	3
General mean		78.21	40.83	7.12	11.73	3.49	6.22	2.92	2.54	9.38	54.89	121.23	17.39	4.28				
SE		0.51	0.59	0.15	0.11	0.04	0.05	0.03	0.07	0.14	0.63	0.54	0.45	0.03				
Test entries mean		78.12	40.71	7.12	11.70	3.48	6.21	2.91	9.38	54.92	121.13	17.34	4.28	2.53				
Check mean		91.38	58.86	7.66	15.75	5.24	7.75	3.91	8.75	50.21	135.75	23.86	5.39	3.08				
CD (p=0.05)																		
	Test entries	13.33	11.93	3.24	2.87	1.24	0.86	0.97	1.03	3.18	14.86	9.75	10.96	0.91				
	Checks	6.66	5.97	1.62	1.44	0.62	0.43	0.49	0.51	1.59	7.43	4.87	5.48	0.46				
	Test Vs Checks	10.53	9.43	2.56	2.28	0.99	0.67	0.77	0.81	2.51	11.75	7.71	8.67	0.72				

Abbreviations used: DF- Days to 50 per cent flowering (days); PH- Plant height (cm); Ptil- Number of productive tillers (count); InfloL- Inflorescence length (cm); RN- Raceme number (count); RL-Raceme length (cm); CD- Culm diameter (mm); PF- Pulling force (PF); IntL- Internode length (cm); RA- Recovery angle after bending (degrees); DM- Days to maturity (days); GY- Grain yield per plant (g);TW-Test weight (g); PT- Plant type; EE- Ear exertion at dough stage; SA- Spikelet arrangement on rachis; DL- Degree of lodging.

4.1.1.3 Number of productive tillers

The mean value for this trait ranged from 3.33 (DPS 637/1) to 20 (Aamo 73). The general mean recorded was 7.12. Significantly more number of productive tillers than the check CO3 was observed in 35 entries.

4.1.1.4 Inflorescence length

General mean recorded for this trait was 11.73 cm. Maximum inflorescence length was recorded by RK 50 with 21 cm and a minimum was recorded in TNPsc 122 (7.6 cm). The genotypes, TNPsc 203, TNPsc 206, TNPsc 215, RK 5, TNPsc 204, TNPsc 205, TNPsc 202, TNPsc 231, *Thittakudi* and RK 50 had significantly longer inflorescence length than the check CO3.

4.1.1.5 Raceme number

The general mean observed among the genotypes was 3.49, with a maximum number in Aamo 215 (7.2) and least in IPS 194, TNPsc 123, TNPsc 126, TNPsc 199 (2.0). Twenty one genotypes had significantly greater number of racemes than the check CO3.

4.1.1.6 Raceme length

The maximum raceme length of 10 cm was observed in TNPsc 231 and minimum in TNPsc 122 (3.8 cm). A total of 18 genotypes exceeded the check significantly. The general mean for this trait was 6.22 cm.

4.1.1.7 Culm diameter

The genotype TNPsc 215 recorded the highest value (5.45 mm) whereas Aamo 326 recorded the lowest (1.72 mm) of all. The genotypes Aamo 126, TNPsc 156, TNPsc 206, TNPsc 155 and TNPsc 215 had significantly higher culm diameter than the check *Adari*.

4.1.1.8 Pulling force

The range of pulling force observed was 0.86 N in TNPsc 125 to 7.78 N in Aamo 13. The general mean for this trait was 2.54 N. A total of 38 genotypes registered significantly higher pulling force than the check *Adari*.

4.1.1.9 Internode length

The internode length ranged from 3.25 cm (Aamo 322) to 19.5 cm (Aamo 33). Thirteen genotypes had significantly shorter internode than the check, *Adari*.

4.1.1.10 Recovery angle after bending

The highest recovery angle after bending (80°) was recorded by three genotypes viz., Aamo 340, IPS 115 and TNPsc 160 and the lowest by TNPsc 170 (20°). Of all, 49 genotypes had significantly better recovery angle than the check *Adari*.

4.1.1.11 Days to maturity

The genotype Aamo 4 was the earliest to mature within 102 days duration and the genotype TNPsc 199 recorded the longest duration of 172 days among the test entries studied. The mean for this trait was 121 days and 17 genotypes were significantly late in maturity and 149 genotypes were significantly earlier when compared with the check CO3.

4.1.1.12 Grain yield per plant

Among the test entries, maximum grain yield per plant (57.55 g) was recorded by the genotype TNPsc 164, while the minimum yield was 1.11 g in IPS 194. The overall average grain yield per plant was 17.39 g. The test entries TNPsc 174, Sel 19, TNPsc 195, RK 50, Aamo 536, TNPsc 162, RK 25, TNPsc 155, TNPsc 171, TNPsc 188, TNPsc 176, TNPsc 172, Sel 17, Sel 21, TNPsc 191, TNPsc 183, TNPsc 187, TNPsc 221 and TNPsc 164 were identified as higher yielders when compared with the check CO3.

4.1.1.13 Test weight

The general mean for this trait was 4.28 g. Mean values registered a range of 2.75 (IPS 194) to 6.06 g (TNPsc 217). None of the genotypes were significantly better in test weight than the check CO3. But, seven genotypes (TNPsc 176, TNPsc 230, *Thittakudi*, TNPsc 215, TNPsc 81, RK 50 and TNPsc 217) had higher mean than that of the check, though not significant.

4.1.2 Frequency distribution of four qualitative traits

The frequency distribution for four qualitative traits which were observed in discrete classes is described below.

4.1.2.1 Plant type

Among the 297 test entries, 270 were erect, two were decumbent and the remaining 25 were prostrate in their growth habit.

4.1.2.2 Ear exertion

Majority of the genotypes (252 nos.) were full in their ear exertion and the remaining (45 nos.) being partial in ear exertion.

4.1.2.3 Spikelet arrangement

Most of the genotypes (245 nos.) had regular spikelet arrangement, but, 19 genotypes had irregular arrangement and the remaining 33 genotypes had variable spikelet arrangement.

4.1.2.4 Degree of lodging

Lodging was severe in 19 genotypes and low in 182 genotypes and it was moderate in 96 genotypes.

4.1.3 Association studies

Estimation of the phenotypic correlation coefficients and the direct and indirect effects of these traits on each other were studied using the mean data.

4.1.3.1 Correlation between culm strength and its components

The pulling force, which quantifiably represents the culm strength, exhibited the highest positive correlation with culm diameter (0.544) followed by plant height (0.379), number of productive tillers per plant (0.324), grain yield per plant (0.219) and days to 50 per cent flowering (0.155). But, pulling force had no correlation with internode length, recovery angle after bending and degree of lodging.

4.1.3.2 Correlation between lodging and its component traits

Plant elasticity represented by recovery angle after bending had significant negative correlation with internode length (-0.392) and degree of lodging (-0.470). The

correlation between internode length and degree of lodging was positive and highly significant (0.490).

4.1.3.3 Correlation between grain yield and its components

Grain yield per plant had significant positive correlation with plant height (0.465), number of productive tillers per plant (0.191), culm diameter (0.206), internode length (0.250), degree of lodging (0.240) and pulling force (0.219), and negative association with recovery angle after bending (-0.316) (Table 7).

4.1.3.4 Inter correlation among other traits

4.1.3.4.1 Days to 50 per cent flowering

Day to 50 per cent flowering was positively correlated with culm diameter (0.351), days to maturity (0.857) and pulling force (0.155), and negatively correlated with internode length (-0.359) and degree of lodging (-0.250).

4.1.3.4.2 Plant height

Plant height was positively and significantly correlated with most of the traits such as number of productive tillers (0.416), culm diameter (0.521), internode length (0.556), degree of lodging (0.390), grain yield per plant (0.465) and pulling force (0.379), but negatively correlated with recovery angle after bending (-0.432). The traits, days to 50 per cent flowering and days to maturity showed insignificant correlation with plant height.

4.1.3.4.3 Number of productive tillers

The number of productive tillers had significant positive correlation with all the traits except day to 50 per cent flowering. It had positive correlations with plant height (0.416), culm diameter (0.257), internode length (0.369), degree of lodging (0.100), grain yield per plant (0.191) and pulling force (0.324) and negative correlation with recovery angle after bending (-0.176) and days to maturity (-0.320).

4.1.3.4.4 Culm diameter

Culm diameter had significant positive correlation with six traits *viz.*, days to 50 per cent flowering (0.351), plant height (0.521), number of productive tillers (0.257), days to maturity (0.193), grain yield per plant (0.206) and pulling force (0.544). It had

Table 7. Simple phenotypic correlation coefficients between pulling force and other related traits [Kharif, (Aug.- Dec.), 2012]

Traits	DF	PH	Ptil	CD	IntL	RA	DL	DM	GY
PH	0.088								
Ptil	-0.113	0.416**							
CD	0.351**	0.521**	0.257**						
IntL	-0.359**	0.556**	0.369**	-0.024					
RA	0.095	-0.432**	-0.176**	-0.135*	-0.392**				
DL	-0.250**	0.390**	0.100*	0.010	0.490**	-0.470**			
DM	0.857**	-0.090	-0.320**	0.193**	-0.453**	0.160**	-0.290**		
GY	-0.107	0.465**	0.191**	0.206**	0.250**	-0.316**	0.240**	-0.190**	
PF	0.155*	0.379**	0.324**	0.544**	-0.026	0.103	-0.090	0.036	0.219**

*significant at 5% level; **significant at 1% level

Abbreviations used: DF- Days to 50 per cent flowering; PH- Plant height; Ptil- Number of productive tillers; CD- Culm diameter; IntL- Internode length; RA- Recovery angle after bending; DL- Degree of lodging; DM- Days to maturity; GY- Grain yield per plant; PF- Pulling force.

negative correlation with recovery angle after bending (-0.135). The traits internode length and degree of lodging had insignificant correlation with culm diameter.

4.1.3.4.5 Internode length

Internode length exhibited positive correlation with plant height (0.556), number of productive tillers (0.369), degree of lodging (0.490) and grain yield per plant (0.250). But, it was negative with days to 50 per cent flowering (-0.359), recovery ability after bending and days to maturity (-0.453). Its relationships with culm diameter and pulling force were not significant.

4.1.3.4.6 Recovery angle after bending

Recovery angle after bending had insignificant correlation with days to 50 per cent flowering (0.095) and pulling force (0.103). But, it exhibited significant positive correlation with days to maturity (0.160) and negative correlation with plant height (-0.432), number of productive tillers (-0.176), culm diameter (-0.135), internode length (-0.392), degree of lodging (-0.470) and grain yield per plant (-0.316).

4.1.3.4.7 Days to maturity

This trait showed positive association with days to 50 per cent flowering (0.857), culm diameter (0.193) and recovery angle after bending (0.160), but negatively correlated with number of productive tillers (-0.320), internode length (-0.453), degree of lodging (-0.290) and grain yield per plant (0.190). Its correlations with plant height and pulling force were not significant.

4.1.4 Path analysis

Path analysis was carried out to find out the direct and indirect effects of associated traits with pulling force (Table 8), which was considered as dependent variable. Path analysis revealed that culm diameter (0.378) had the highest positive direct effect on pulling force followed by plant height (0.292) and recovery angle after bending (0.255). The traits number of productive tillers per plant and internode length exhibited low level of positive (0.188) and negative (-0.170) effects respectively on pulling force. The traits, days to 50 percent flowering (-0.054), degree of lodging (-0.067), days to

Table 8. Direct and indirect effects of different traits on pulling force [Kharif, (Aug.- Dec.), 2012]

Traits	Effect via									Correlation with pulling force
	DF	PH	Ptil	CD	IntL	RA	DL	DM	GY	
DF	-0.054	0.026	-0.021	0.133	0.061	0.024	0.017	-0.020	-0.011	0.155*
PH	-0.005	0.292	0.078	0.197	-0.094	-0.110	-0.026	0.002	0.046	0.379**
Ptil	0.006	0.121	0.188	0.097	-0.063	-0.045	-0.007	0.007	0.019	0.324**
CD	-0.019	0.152	0.048	0.378	0.004	-0.034	-0.001	-0.004	0.020	0.544**
IntL	0.019	0.162	0.069	-0.009	-0.170	-0.100	-0.033	0.010	0.025	-0.026
RA	-0.005	-0.126	-0.033	-0.051	0.067	0.255	0.032	-0.004	-0.031	0.103
DL	0.013	0.114	0.019	0.004	-0.083	-0.120	-0.067	0.007	0.024	-0.068
DM	-0.046	-0.026	-0.060	0.073	0.077	0.041	0.019	-0.023	-0.019	-0.090
GY	0.006	0.136	0.036	0.078	-0.042	-0.081	-0.016	0.004	0.099	0.219**

Residual effect = 0.759

*significant at 5% level; **significant at 1% level

Abbreviations used: DF- Days to 50 per cent flowering; PH- Plant height; Ptil- Number of productive tillers; CD- Culm diameter; IntL- Internode length; RA- Recovery angle after bending; DL- Degree of lodging; DF- Days to maturity; GY- Grain yield per plant; PF- Pulling force.

maturity (-0.023) and grain yield per plant (0.099) exhibited negligible level of direct effect on pulling force.

The indirect effects on pulling force were either low or negligible for all the traits studied.

4.2 Selection for high culm strength, bold seeded and erect genotypes [Summer (Jan.- May), 2013]

From the 297 germplasm accessions evaluated during *Kharif*, 2012, 41 were forwarded to analyzed for consistency of performance with respect to culm strength and yield during Summer (Jan. – May), 2013. The genotypes included thirty germplasm accessions, identified as having good culm strength, two checks (CO3 and *Adari*) and nine weak culm genotypes for comparison.

4.2.1 Mean performance of genotypes for 14 quantitative traits

The result of analysis of variance for the 14 quantitative characters studied in the selected genotypes is presented in Table 9. Mean square of all the characters, showed significant difference ($P < 0.05$) among the tested genotypes for all the traits except for number of productive tillers per plant. Hence, the trait, number of productive tillers per plant was excluded from further analysis as there was no significant variation among genotypes for this trait.

The mean performance of 41 selected germplasm accessions is presented in table 10 along with the mean performance of the checks over replications.

4.2.1.1 Days to 50 per cent flowering

Days to 50 per cent flowering ranged between 62 days (Aamo 29) and 101.50 days (TNPsc 215) with an overall mean of 83.15 days (Table 10). Eight test entries *viz.*, Aamo 398, TNPsc 170, TNPsc 106, Aamo 10, Aamo 72, Aamo 37, Aamo 29 and Aamo 36 were significantly earlier than the check, CO3 and five accessions *viz.*, TNPsc 155, TNPsc 215, *Peruvaragu*, *Adari*, TNPsc 217 were significantly longer in duration than the check.

Table 9. ANOVA for 14 traits in 41 genotypes of kodo millet in RBD [Summer (Jan.-May), 2013]

Sl. No.	Traits	Mean sum of squares		
		Replications (df=1)	Treatments (df=40)	Error (df=40)
1	Days to 50 per cent flowering	10.976	303.756**	34.676
2	Plant height	1.478	158.539**	68.471
3	Numbers of productive tillers	0.883	1.712	1.323
4	Inflorescence length	0.478	6.413**	2.069
5	Raceme number	0.195	0.474**	0.209
6	Raceme length	7.219**	2.397**	0.777
7	Culm diameter	0.052	1.530**	0.198
8	Pulling force	0.000	4.311**	0.582
9	Culm weight per unit length	7.689**	72.851**	1.828
10	Internode length	4.637	5.745*	3.191
11	Recovery angle after bending	34.697	195.162**	71.822
12	Days to maturity	0.049	241.764**	48.324
13	Grain yield per plant	49.826**	69.830**	5.863
14	Test weight	3.114**	0.560**	0.226

*significant at 5% level; **significant at 1% level

Table 10. Mean performance of 41 germplasm accessions of kodo millet for 14 quantitative traits [Summer (Jan.-May), 2013]

Sl. No	Genotypes	1 DF	2 PH	3 Ptil	4 infloL	5 RN	6 RL	7 CD	8 PF	9 WL	10 IntL	11 RA	12 DM	13 GY	14 TW
A	Lines with better culm strength														
1	Aamo 5	78.5	52.17	5.75	14.25	3.33	7.63	4.13	4.05	12.55	10.83	54.17	115.0	21.60	4.63
2	Aamo 13	74.5	50.33	5.67	13.67	3.17	7.42	4.43	3.93	13.45	10.17	50.00	116.0	15.85	4.35
3	Aamo 126	77.0	54.67	6.83	13.67	3.17	7.25	4.58	4.22	12.00	11.08	51.67	116.0	16.57	4.38
4	Aamo 210	75.0	56.17	6.17	13.25	3.67	7.75	4.51	4.71	13.60	10.75	45.00	113.0	23.31	4.50
5	Aamo 258	75.5	54.83	6.00	15.00	4.00	7.75	4.70	5.20	13.80	11.58	51.67	114.0	26.56	4.50
6	RK 50	85.5	56.17	6.50	17.33	4.33	8.12	3.82	3.02	9.15	11.75	40.83	125.5	19.49	4.67
7	Sel 21	74.5	50.00	6.83	13.33	3.33	7.33	3.85	5.44	9.50	9.75	40.00	114.0	25.87	4.83
8	TNPSc 86	83.0	57.17	8.33	16.00	3.17	8.62	3.75	5.13	6.70	10.42	40.00	128.5	18.48	5.23
9	TNPSc 147	87.0	54.67	8.17	16.17	3.00	8.25	3.38	2.98	8.25	9.67	39.17	127.5	15.14	4.72
10	TNPSc 155	101.5	67.42	6.42	16.08	2.83	8.17	5.16	6.41	13.84	10.83	32.50	123.0	5.80	4.91
11	TNPSc 156	91.0	57.17	7.50	16.50	2.83	7.92	4.24	2.85	23.55	12.67	55.00	127.5	25.82	5.02
12	TNPSc 176	78.0	50.83	6.33	11.67	3.33	6.92	3.99	5.54	7.55	9.75	29.17	118.0	18.71	5.53
13	TNPSc 183	83.0	61.17	7.17	15.17	3.00	8.17	4.05	2.47	8.90	10.25	47.50	115.0	18.37	4.19
14	TNPSc 201	87.0	58.08	8.17	13.58	3.67	7.33	4.06	4.47	19.10	11.17	51.25	132.5	14.44	4.44
15	TNPSc 215	101.5	53.42	6.50	14.58	3.50	7.08	4.25	4.50	14.18	10.17	44.17	131.0	9.65	5.48
16	TNPSc 217	99.0	53.92	6.83	15.08	3.33	7.08	4.31	5.19	15.19	10.33	50.00	129.0	8.85	5.44
17	TNPSc 221	94.5	44.92	6.00	15.67	3.33	8.13	2.97	2.75	9.25	10.04	43.33	124.5	12.95	5.18
18	TNPSc 225	82.0	52.67	7.50	14.83	2.83	8.37	2.70	4.64	8.96	12.40	49.17	126.0	12.85	5.21
19	TNPSc 231	88.0	61.33	7.08	19.17	2.83	9.50	3.36	2.69	9.00	15.50	42.50	129.5	10.12	4.72
20	TNPSc 232	86.0	62.33	8.17	14.83	2.83	8.50	2.70	4.24	5.02	14.17	60.00	142.5	9.33	4.35
21	TNPSc 233	88.0	46.83	6.67	14.42	3.17	8.17	3.14	4.29	13.14	10.00	54.17	134.5	15.01	5.10
22	TNPSc 235	94.5	51.33	8.17	15.08	3.00	8.33	2.89	5.01	6.30	10.75	44.17	141.0	9.02	5.18
23	TNPSc 236	89.0	45.67	7.33	13.50	2.67	6.50	2.83	2.93	11.25	10.42	25.00	137.0	11.63	3.88
24	<i>Athipatti</i>	97.0	67.83	6.08	15.25	2.67	7.83	5.22	3.84	32.70	9.67	45.83	125.0	4.49	4.42
25	<i>Naraiyur</i>	91.0	49.67	7.33	13.83	2.17	8.67	3.71	3.61	18.85	15.00	50.00	137.0	20.69	4.22

Table 10 continued...

Sl. No.	Genotypes	1 DF	2 PH	3 Ptil	4 infloL	5 RN	6 RL	7 CD	8 PF	9 WL	10 IntL	11 RA	12 DM	13 GY	14 TW
26	<i>Pacheri</i>	96.5	53.00	8.08	13.50	3.17	9.58	3.42	3.75	6.45	12.33	42.50	142.5	21.22	4.59
27	<i>Peruvaragu</i>	100.5	61.00	6.33	14.33	2.75	7.42	5.25	6.18	15.40	8.92	62.50	134.0	14.74	5.17
28	<i>Podivaragu</i>	88.0	74.00	7.00	14.67	3.33	8.08	5.58	4.01	15.01	11.08	56.67	132.5	13.32	4.67
29	<i>Thittakudi</i>	97.5	54.83	7.25	17.75	2.92	10.25	4.32	4.54	13.45	12.58	53.75	130.5	16.09	4.96
30	<i>Venganoor</i>	92.0	61.50	8.67	18.67	2.67	11.17	3.07	4.82	17.58	12.50	37.50	137.5	15.97	3.90
B Lines with poor culm strength															
1	Aamo 8	76.0	40.75	4.50	13.33	2.83	7.03	3.20	1.46	6.30	8.75	68.34	110.5	13.05	4.01
2	Aamo 10	64.0	37.33	6.08	12.58	2.58	6.25	3.20	1.53	5.65	9.12	45.83	109.0	6.78	3.63
3	Aamo 29	62.0	37.67	6.50	12.08	2.17	6.42	2.48	1.96	5.53	10.83	46.67	110.0	8.72	3.70
4	Aamo 36	62.0	39.92	6.25	12.63	3.00	6.42	2.78	1.53	5.40	11.08	23.33	110.0	7.11	4.36
5	Aamo 37	62.5	37.00	5.75	12.42	2.58	6.33	2.51	1.24	5.43	10.50	31.67	109.5	7.82	4.38
6	Aamo 72	63.0	38.25	7.58	11.50	3.25	6.25	2.74	1.48	5.69	10.42	39.17	110.5	12.44	3.86
7	Aamo 398	67.0	48.83	6.50	14.08	3.17	6.83	3.12	1.45	6.70	12.25	47.50	112.0	11.58	3.72
8	TNPSc 106	65.5	38.17	8.25	11.58	2.83	6.58	2.04	1.32	5.50	11.08	42.50	110.5	7.51	4.07
9	TNPSc 170	65.5	47.67	6.50	15.83	2.00	6.92	2.49	1.36	5.70	16.17	25.83	111.5	14.35	3.71
C Checks															
1	CO3	86.0	42.83	7.75	15.42	2.50	8.92	2.93	2.27	6.80	10.08	52.50	129.0	22.18	4.24
2	<i>Adari</i>	99.5	58.83	5.75	15.50	4.08	8.67	4.20	3.67	23.40	9.42	50.00	145.0	6.71	4.19
Mean		83.15	52.25	6.88	14.58	3.05	7.80	3.66	3.58	11.36	11.13	45.43	124.07	14.39	4.54
SE		1.92	1.39	0.14	0.28	0.08	0.17	0.14	0.23	0.34	0.26	1.54	1.72	0.92	0.08
CD (0.05)		11.90	16.72	NA	2.91	0.93	1.78	0.90	1.54	2.73	3.61	17.13	14.05	4.89	0.96

Abbreviations used: DF- Days to 50 per cent flowering (days); PH- Plant height (cm); ProT- Number of productive tillers (count); InfloL- Inflorescence length (cm); RN- Raceme number (count); RL- Raceme length (cm); CD- Culm diameter (mm); PF- Pulling force (N); WL- culm weight per unit length (mg/cm); IntL- Internode length (cm); RA- Recovery angle after bending (degrees); DM- Days to maturity (days); GY- Grain yield per plant (g); TW- Test weight (g).

4.2.1.2 Plant height

Among the 41 accessions, Aamo 37 had the lowest value (37 cm) for plant height. The maximum for this trait was recorded by *Podivaragu* (74 cm). The general mean recorded was 52.25 cm. The genotypes, *Podivaragu*, *Athipatti*, TNPsc 155, TNPsc 232, *Venganoor*, TNPsc 231, TNPsc 183, *Peruvaragu* were on par with CO3, in height (Table 10).

4.2.1.3 Number of productive tillers

No significant differences existed between genotypes for this trait. The overall mean for number of productive tillers per plant was 6.88.

4.2.1.4 Inflorescence length

General mean recorded for this trait was 14.58 cm; a maximum length was recorded in TNPsc 231 (19.17 cm) and a minimum in Aamo 72 (11.50 cm). The genotypes TNPsc 231 and *Venganoor* had significantly longer inflorescence length than the check CO3 (Table 10).

4.2.1.5 Raceme length

A maximum raceme length of 11.17 cm was noticed in *Venganoor* and a minimum raceme length of 6.25 cm was observed in Aamo 10 and Aamo 72. The land race *Venganoor* alone exceeded the check, significantly, for the trait raceme length. The general mean for this trait was 7.80 cm (Table 10).

4.2.1.6 Raceme number

The general mean observed for the genotypes was 3.05, with the maximum number in RK 50 (4.33) and least in TNPsc 170 (2.00). The entries, RK 50, Adari, Aamo 258, Aamo 210, TNPsc 201, TNPsc 215 had significantly greater number of racemes than the check CO3 (Table 10).

4.2.1.7 Culm diameter

The genotype *Podivaragu* recorded the maximum diameter (5.58 mm) where as TNPsc 106 recorded the least (2.04 mm). The genotypes *Athipatti* (5.22 mm), *Podivaragu* (5.58 mm), *Peruvaragu* (5.25 mm) and TNPsc 155 (5.16 mm) had significantly higher culm diameter than the check, *Adari*. But compared to CO3, eighteen

lines (*Podivaragu*, *Peruvaragu*, *Athipatti*, TNPsc 155, Aamo 258, Aamo 126, Aamo 210, Aamo 13, *Thittakudi*, TNPsc 217, TNPsc 215, TNPsc 156, *Adari*, Aamo 5, TNPsc 201, TNPsc 183, TNPsc 176 and Sel 21) had significantly better culm diameter (Table 10).

4.2.1.8 Pulling force

The range of pulling force observed was from 1.24 N (Aamo 37) to 6.41 N (TNPsc 155). The general mean for this trait was 3.58 N. Four genotypes, viz., Sel 21 (5.44 N), TNPsc 155 (6.41 N), TNPsc 176 (5.54 N) and *Peruvaragu* (6.18 N) registered significantly higher pulling force than the check, *Adari*. Twenty one genotypes (TNPsc 155, *Peruvaragu*, TNPsc 176, Sel 21, Aamo 258, TNPsc 217, TNPsc 86, TNPsc 235, *Venganoor*, Aamo 210, TNPsc 225, *Thittakudi*, TNPsc 215, TNPsc 201, TNPsc 233, TNPsc 232, Aamo 126, Aamo 5, *Podivaragu*, Aamo 13, *Athipatti*) were better than CO3 with respect to culm strength (Table 10).

4.2.1.9 Culm weight per unit length

The range registered for this trait was from 5.02 mg/cm (TNPsc 232) to 32.70 mg/cm (*Athipatti*). The land race, *Athipatti* was the only genotype which had significantly higher culm weight per unit length when compared with *Adari*. But the lines *Athipatti*, TNPsc 156, *Adari*, TNPsc 201, *Naraiyur*, *Venganoor*, *Peruvaragu*, TNPsc 217, *Podivaragu*, TNPsc 215, TNPsc 155, Aamo 258, Aamo 210, Aamo 13, *Thittakudi*, TNPsc 233, Aamo 5, Aamo 126 and TNPsc 236 exhibited higher value for this trait than the check CO3 (Table 10).

4.2.1.10 Internode length

The internode length ranged from 8.75 cm (Aamo 8) to 16.17 cm (TNPsc 170). None of the test entries had significantly shorter internode than both the checks, *Adari* and CO3 (Table 10).

4.2.1.11 Recovery angle after bending

The highest recovery angle after bending (68.34°) was recorded by the genotype, Aamo 8 and the least by Aamo 36 (23.33°). Among all the genotypes studied, Aamo 8 had better recovery angle after bending than the check *Adari* (Table 10).

4.2.1.12 Days to maturity

The genotype, Aamo 10 was the earliest to mature with 109 days duration and the genotype *Adari* was recorded a longest duration of 145 days among the test entries studied. The mean for this trait was around 124.07 days. *Adari* had significantly longer duration than the check CO3 and 12 other genotypes (Aamo 258, Sel 21, Aamo 210, Aamo 398, TNPsc 170, TNPsc 106, Aamo 8, Aamo 72, Aamo 29, Aamo 36, Aamo 37, Aamo 10) were significantly early to mature when compared with the check CO3 (Table 10).

4.2.1.13 Grain yield per plant

Among the test entries, the maximum yield (26.56 g) was recorded in Aamo 258 and the minimum yield (4.49 g / plant) was observed in the genotype *Athipatti*. The average yield was observed to be 14.39 g per plant. The genotypes, Aamo 258, Sel 21, TNPsc 156, Aamo 210, Aamo 5, *Pacheri*, *Naraiyur*, RK 50, TNPsc 176, TNPsc 86 and TNPsc 183, were on par with the check CO3 with respect to this trait (Table 10).

4.2.1.14 Test weight

The general mean for this trait was 4.54 g. Mean values of genotypes registered a range of 3.63 g (Aamo 10) to 5.53 g (TNPsc 176). The genotypes, TNPsc 176, TNPsc 215, TNPsc 217, TNPsc 86 and TNPsc 225 were significantly better in test weight than the check CO3 (Table 10).

4.2.2 Frequency distribution of eight qualitative traits in 41 genotypes

The frequency distribution for eight qualitative traits (Table 11) is described below.

4.2.2.1 Plant type

Among the 41 test entries, 23 were erect, seven were decumbent and the remaining 11 were prostrate in their growth habit.

4.2.2.2 Ear exertion at dough stage

Majority of the genotypes (36) were full in their ear exertion and the remaining (5) being partial in ear exertion.

4.2.2.3 Spikelet arrangement on rachis

Most of the genotypes (30) had regular spikelet arrangement, three genotypes had irregular arrangement and the remaining (8) genotypes had variable spikelet arrangement.

Table 11: Performance of 41 germplasm accessions for eight qualitative traits [Summer (Jan.-May), 2013]

Sl. No	Genotypes	1	2	3	4	5	6	7	8
		PT	EE	SA	DC	SG	SI	UM	DL
A	Lines with better culm strength								
1	Aamo 5	1	1	3	3	1	0	1	5
2	Aamo 13	1	1	2	3	1	0	0	5
3	Aamo 126	1	1	3	5	1	0	0	5
4	Aamo 210	1	1	2	5	1	0	1	7
5	Aamo 258	1	1	3	3	1	0	1	5
6	RK 50	1	1	1	5	1	0	0	3
7	Sel 21	1	1	2	3	1	0	1	5
8	TNPSc 86	1	1	1	7	0	0	0	3
9	TNPSc 147	2	1	1	7	0	1	0	5
10	TNPSc 155	1	1	1	3	1	1	0	3
11	TNPSc 176	1	1	3	5	1	0	1	5
12	TNPSc 183	1	1	1	5	1	0	1	3
13	TNPSc 201	2	1	3	3	1	0	0	3
14	TNPSc 215	1	2	1	5	0	1	0	5
15	TNPSc 217	2	2	1	5	1	0	0	3
16	TNPSc 221	1	1	1	5	1	0	1	5
17	TNPSc 225	1	1	1	5	1	0	0	3
18	TNPSc 231	1	1	1	7	1	0	0	3
19	TNPSc 232	3	1	1	7	0	0	1	5
20	TNPSc 233	1	1	1	5	0	0	1	5
21	TNPSc 235	3	1	1	5	0	1	0	5
22	TNPSc 236	1	1	1	5	0	0	0	7
23	<i>Athipatti</i>	3	1	1	3	1	1	0	5
24	<i>Naraiyur</i>	3	1	1	3	1	0	0	7
25	<i>Pacheri</i>	3	1	1	3	1	1	0	5

Table 11 continued...

Sl. No.	Genotypes	15 PT	16 EE	17 SA	18 DC	19 SG	20 SI	21 UM	22 DL
26	<i>Peruvaragu</i>	3	1	1	3	1	0	0	3
27	<i>Podivaragu</i>	3	1	1	3	1	1	0	3
28	<i>Thittakudi</i>	1	1	1	3	1	0	0	3
29	<i>Venganoor</i>	3	1	1	5	0	1	0	7
B	Lines with poor culm strength								
1	Aamo 8	2	1	3	3	0	0	0	7
2	Aamo 10	3	1	3	7	0	1	0	7
3	Aamo 29	2	1	1	3	0	0	0	7
4	Aamo 36	2	2	3	3	1	1	0	7
5	Aamo 37	1	2	1	3	0	1	1	7
6	Aamo 72	3	2	1	3	1	1	0	7
7	Aamo 398	1	1	1	3	1	1	1	7
8	TNPSc 106	1	1	1	3	1	1	0	7
9	TNPSc 156	1	1	1	5	1	0	0	3
10	TNPSc 170	2	1	1	3	0	1	0	7
C	Checks								
1	CO3	1	1	1	5	0	0	0	5
2	<i>Adari</i>	3	1	1	3	1	1	0	5

Abbreviations used: PT- Plant type; EE- Ear exertion at dough stage; SA- Spikelet arrangement on rachis; DC- Degree of chlorosis; SG- Stay green nature at maturity; SI- Shattering of inflorescence; UM- Uniformity in maturity; DL- Degree of lodging

4.2.2.4 Degree of chlorosis

Among the 41 genotypes, only five exhibited severe chlorosis and the remaining were low (21 genotypes) to intermediate (15 genotypes) with respect to this trait.

4.2.2.5 Stay green nature at maturity

Out of the 41 genotypes, only 27 remained green at maturity.

4.2.2.6 Shattering of inflorescence

Shattering of inflorescence was observed in 16 genotypes, while 25 had non-shattering inflorescence.

4.2.2.7 Uniformity in maturity

Only 11 genotypes exhibited uniformity in maturity out of the 41 genotypes studied.

4.2.2.8 Degree of lodging

Lodging was severe in 13 genotypes and low in 12 genotypes and it was moderate in 16 genotypes.

4.2.3 Genetic analysis for variability, heritability and genetic advance

4.2.3.1 Variance components

The extent of variability in respect of range, phenotypic and genotypic coefficients of variation, is presented in table 12.

4.2.3.1.1 Phenotypic and genotypic variances

The difference between phenotypic and genotypic variances was less for all the traits except for days to 50 per cent flowering, plant height, days to maturity and recovery angle after bending (Table 12). Days to 50 per cent flowering expressed highest genotypic and phenotypic variances (134.54 and 169.22 days, respectively) followed by days to maturity (96.72 and 145.04 days, respectively). The magnitude of genotypic and phenotypic variances were the lowest for the trait, raceme number (0.13 and 0.34, respectively) followed by test weight (0.17 and 0.39g, respectively).

Table 12. Genetic parameters for 13 quantitative traits in kodo millet genotypes [Summer (Jan.-May), 2013]

Sl No	Traits	Minimum	Maximum	Mean±SE	Vg	Vp	GCV %	PCV %	Heritability %	GA	GA (%) of mean
1	Days to 50 per cent flowering	62.00	101.50	83.15 ±1.92	134.54	169.22	13.95	15.65	79.51	21.31	25.62
2	Plant height	37.00	74.00	52.25 ±1.39	45.03	113.51	12.84	20.39	39.68	8.71	16.66
3	Inflorescence length	11.50	19.17	14.58 ±0.28	2.17	4.24	10.11	14.12	51.21	2.17	14.90
4	Raceme number	2.00	4.33	3.05 ±0.08	0.81	1.59	11.94	16.15	51.04	1.32	16.98
5	Raceme length	6.25	11.17	7.80 ±0.17	0.13	0.34	11.54	19.17	38.80	0.47	15.32
6	Culm diameter	2.04	5.58	3.66 ±0.14	0.67	0.86	22.30	25.40	77.08	1.48	40.33
7	Pulling force	1.24	6.41	3.58 ±0.23	1.86	2.45	38.18	43.74	76.21	2.46	68.67
8	Culm weight per unit length	5.02	32.70	11.36 ±0.94	35.51	37.34	52.46	53.79	95.10	11.97	105.38
9	Internode length	8.75	16.17	11.13 ±0.26	1.28	4.47	10.16	19.00	28.58	1.24	11.18
10	Recovery angle after bending	23.33	68.34	45.43 ±1.54	61.67	133.49	17.29	25.43	46.20	11.00	24.20
11	Days to maturity	109.00	145.00	124.07 ±1.72	96.72	145.04	7.93	9.71	66.68	16.54	13.33
12	Grain yield per plant	4.49	26.56	14.39 ±0.92	31.98	37.85	39.29	42.74	84.51	10.71	74.40
13	Test weight	3.63	5.53	4.54 ±0.08	0.17	0.39	8.99	13.80	42.49	0.55	12.08

4.2.3.1.2 Phenotypic and genotypic co-efficient of variation

In the present study, the traits days to 50 per cent flowering, inflorescence length, raceme length, culm diameter, pulling force, culm weight per unit length, days to maturity, grain yield per plant and test weight showed narrow differences between phenotypic and genotypic coefficients of variation. The rest of the traits (plant height, number of racemes, internode length and recovery angle after bending) exhibited wide differences between PCV and GCV (Table 12, Fig. 1).

High GCV and PCV estimates were recorded for pulling force (38.18% and 43.74%, respectively), culm weight per unit length (52.46% and 53.79%, respectively) and grain yield per plant (39.29% and 42.74%, respectively). Low GCV was recorded for traits such as days to 50 per cent flowering (13.95%), plant height (12.84%), inflorescence length (10.11%), raceme length (11.54%), raceme number (11.94%), internode length (10.16%), days to maturity (7.93%) and test weight (8.99%) and low PCV for inflorescence length (14.12%), days to maturity (9.71%) and test weight (13.80%). Moderate level of GCV was obtained for culm diameter (22.30%) and recovery angle after bending (17.29%) and moderate level of PCV was recorded for days to 50 per cent flowering (15.65%), plant height (20.39%), raceme length (16.15%), raceme number (19.17%), culm diameter (25.40%), internode length (19.00%) and recovery angle after bending (25.43%) (Table 12).

4.2.3.2 Heritability in broad sense and genetic advance

The estimates of heritability (broad sense), genetic advance and genetic advance as percentage of mean are furnished in Table 12. The genotypes under study showed low level of heritability for the trait, internode length (28.58%). The highest heritability was recorded for culm weight per unit length (95.10%) followed by grain yield per plant (84.51%), days to 50 per cent flowering (79.51%), culm diameter (77.08%), pulling force (76.21%) and days to maturity (66.68%) indicating high influence of genetic components. Remaining traits such as plant height (39.68%), inflorescence length (51.21%), raceme length (51.04%), raceme number (38.80%), recovery angle after bending (46.20%) and test weight (42.49%) recorded medium level of heritability (Table 12, Fig. 2).

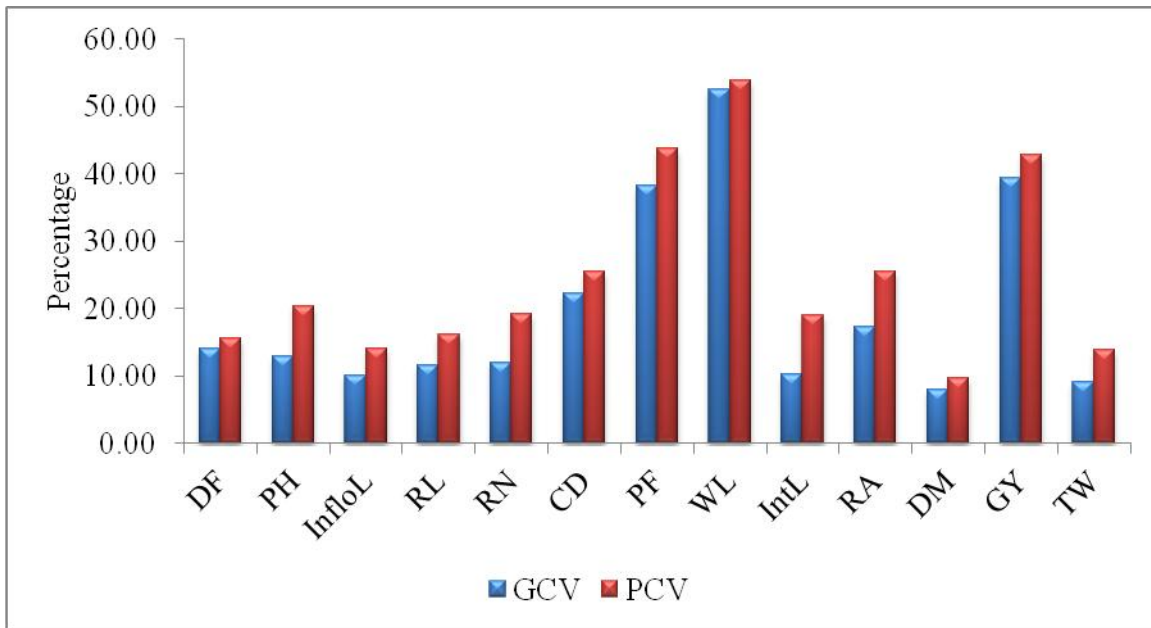


Figure 1. GCV and PCV for 13 quantitative traits in kodo millet [Summer (Jan. - May), 2013]

Pulling force (PF), culm weight per unit length (WL) and grain yield per plant (GY) recorded high GCV and PCV

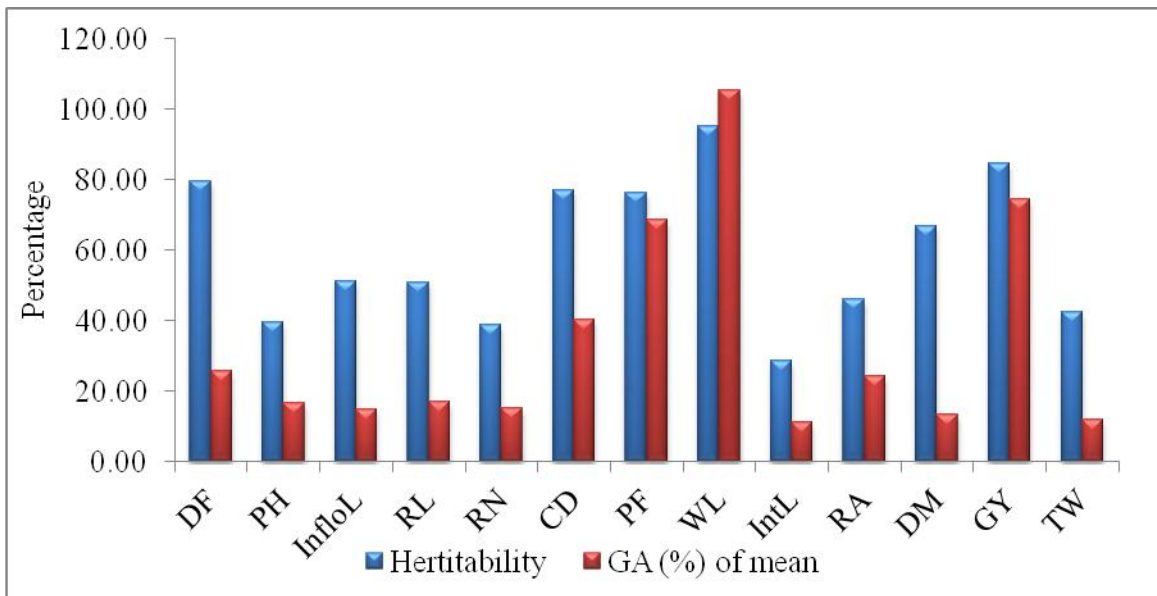


Figure 2. Heritability (broad sense) and genetic advance as per cent of mean for 13 traits in kodo millet [Summer (Jan. - May), 2013]

Culm weight per unit length (WL), grain yield per plant (GY) and pulling force (PF) recorded high heritability along with high genetic advance (GA) over mean

The genetic advance ranged from 0.47 for raceme number to 21.31 for days to 50 per cent flowering (Table 12).

The genetic advance as percentage of mean ranged from 11.18 per cent (internode length) to 105.38 per cent (culm weight per unit length). All the traits studied were of either moderate or high genetic advance category. Those traits which were under moderate genetic advance (as percentage of mean) were plant height (16.66%), inflorescence length (14.90%), raceme length (16.98%), raceme number (15.32%), internode length (11.18%), days to maturity (13.33%), and test weight (12.08%) and those under high genetic advance over mean category were days to 50 per cent flowering (25.62%), culm diameter (40.33%), pulling force (68.67%), culm weight per unit length (105.38%), recovery angle after bending (24.20%) and grain yield per plant (74.40%) (Table 12; Fig. 2).

High heritability along with high genetic advance was recorded for days to 50 per cent flowering, grain yield per plant and other culm related traits such as culm diameter, pulling force and culm weight per unit length. For the remaining traits the genetic advance over mean were moderate coupled with low (internode length) or moderate (plant height, inflorescence length, raceme length, raceme number and test weight) or high (days to maturity) heritability (Table 12; Fig. 2).

4.2.4 Genetic divergence based on quantitative traits

Genetic diversity was analyzed among 41 kodo millet genotypes on the basis of 13 quantitative traits using multivariate analyses, namely Hierarchical Cluster Analysis (HCA) and Principal Component Analysis (PCA).

The Euclidean distances among 41 genotypes were computed (Table 13). The mean phenotypic distance of genotypes was 4.89. Euclidean distance in pair wise comparison ranged between 0.874 and 8.987, from a total of 820 pairs (Fig. 3). The lowest distance coefficient was exhibited by Aamo 13 and Aamo 126 (0.874), followed by TNPsc 217 and TNPsc 215 (0.955), and Aamo 5 and Aamo 126 (1.32). The distance coefficient value was high for TNPsc 170, a weak culm line, with other strong culm lines such as *Peruvaragu* (8.99), *Adari* (8.86) and *Athipatti* (8.48).

Table 13. Euclidean distances among 41 genotypes based on 13 morphological traits [Summer (Jan.-May), 2013]

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	
2	1.45																						
3	1.32	0.87																					
4	1.60	2.01	1.87																				
5	2.09	2.96	2.70	1.61																			
6	3.47	4.01	3.91	3.56	3.26																		
7	2.21	2.63	2.70	1.89	2.77	4.18																	
8	3.03	3.43	3.34	3.39	3.77	3.25	3.02																
9	3.09	3.06	3.22	3.76	4.41	3.22	3.60	2.07															
10	5.05	4.74	4.50	5.06	5.59	5.31	5.39	3.85	4.21														
11	3.40	4.12	3.93	4.20	4.08	4.46	4.75	4.21	4.14	5.46													
12	3.78	3.72	3.76	3.38	4.47	5.01	2.40	3.43	4.12	5.06	5.85												
13	2.28	2.31	2.24	2.89	3.61	3.54	3.47	3.19	2.26	4.68	3.91	4.64											
14	2.67	2.60	2.46	2.86	3.30	3.62	3.81	3.64	3.53	4.40	3.73	4.40	3.44										
15	3.75	3.72	3.67	4.18	4.66	4.03	4.30	3.20	3.28	3.54	4.47	3.80	4.27	2.87									
16	3.61	3.58	3.48	4.19	4.56	4.30	4.36	3.22	3.46	3.34	4.28	4.10	4.28	2.83	0.96								
17	3.38	3.53	3.76	4.24	4.74	3.38	3.98	2.81	1.89	4.90	4.44	4.16	3.40	3.76	2.80	3.10							
18	3.23	3.50	3.39	4.11	4.60	4.22	3.88	2.48	2.74	4.62	4.19	4.14	3.60	3.64	3.53	3.35	2.69						
19	5.45	5.69	5.43	6.12	6.11	4.52	6.55	4.44	4.19	5.20	4.82	7.03	4.68	5.47	5.53	5.44	4.68	3.92					
20	4.77	4.91	4.56	5.58	5.81	5.14	5.91	4.36	4.23	5.56	5.23	6.36	4.49	4.27	5.04	4.83	4.65	3.05	3.81				
21	2.84	3.05	3.23	3.81	4.25	4.01	3.64	2.69	2.58	4.90	3.88	4.11	3.62	2.78	2.71	2.63	2.16	2.26	5.17	3.95			
22	4.30	4.31	4.30	4.99	5.49	4.43	4.65	2.61	2.69	4.23	5.13	4.41	4.34	3.84	2.89	2.94	2.57	2.24	4.56	3.41	2.24		
23	4.98	4.33	4.58	5.26	6.23	5.44	4.93	4.83	3.58	5.60	5.93	4.89	4.47	4.51	4.76	5.09	4.09	4.56	6.04	5.30	4.39	4.18	
24	5.09	4.83	4.95	5.28	5.48	4.51	6.19	5.08	4.55	4.98	5.25	6.56	5.02	3.09	3.90	3.99	4.67	5.42	6.01	5.45	4.18	4.67	
25	5.55	4.98	5.02	5.76	6.35	6.50	6.63	5.93	5.38	4.08	4.96	6.83	5.22	4.31	4.72	4.46	5.93	5.98	6.49	6.59	5.48	6.02	
26	4.48	4.62	4.42	5.15	5.49	5.81	5.49	4.97	4.71	5.85	3.52	6.28	4.62	4.37	5.47	5.33	5.13	4.00	4.64	4.05	4.34	4.87	
27	3.97	4.35	4.21	4.32	4.75	4.03	4.40	3.13	3.22	5.28	4.48	4.87	3.87	3.85	4.29	4.55	3.58	3.31	4.59	3.64	3.19	3.13	
28	3.78	3.79	3.34	3.96	4.21	4.49	5.11	4.06	4.25	3.87	4.09	5.55	3.63	2.94	3.82	3.60	5.04	4.66	5.34	4.65	4.31	4.80	
29	4.12	4.17	3.95	4.70	4.99	5.80	4.92	4.14	4.60	4.01	4.49	5.20	4.70	3.69	3.32	2.78	4.92	4.63	6.64	5.40	3.68	4.32	
30	4.15	4.61	4.44	4.90	4.76	4.23	5.34	3.28	3.65	4.28	3.51	6.11	4.10	4.31	4.27	3.98	3.94	3.49	3.33	4.16	3.62	3.78	
31	5.82	5.95	5.93	6.16	6.25	5.36	6.53	4.70	4.52	5.25	5.14	7.43	5.06	5.49	6.20	6.07	5.45	4.90	3.76	4.89	5.29	5.00	
32	3.58	3.79	4.03	4.67	5.11	4.78	4.36	3.87	2.84	6.32	4.41	5.59	3.15	4.73	5.20	5.21	3.30	3.59	5.12	4.49	3.22	4.05	
33	3.98	3.61	3.97	5.09	5.69	6.04	5.12	5.76	4.65	7.42	6.09	6.13	3.97	5.20	5.86	5.73	4.56	5.00	7.12	5.76	4.48	5.71	
34	5.13	4.16	4.62	5.71	6.67	6.76	5.53	6.38	5.12	7.49	7.32	5.97	4.78	5.96	6.51	6.53	5.32	5.66	7.55	6.57	5.62	6.23	
35	5.33	4.57	4.90	6.00	6.95	7.16	5.68	6.48	5.38	7.76	7.24	6.14	5.08	6.23	6.89	6.85	5.58	5.32	7.29	6.14	5.71	6.20	
36	5.40	4.66	5.01	5.56	6.62	6.05	5.21	5.82	4.83	7.07	7.34	4.89	5.12	6.02	6.10	6.39	4.88	5.24	6.89	6.69	5.74	5.84	
37	5.39	4.66	5.06	5.84	6.90	6.56	5.35	6.03	4.91	7.42	7.27	5.23	5.13	6.22	6.31	6.49	4.90	5.17	7.12	6.63	5.59	5.88	
38	4.68	4.02	4.41	4.95	5.93	5.92	4.77	6.01	4.98	7.65	7.05	5.14	4.70	5.49	6.24	6.45	4.97	5.37	7.42	6.46	5.39	6.08	
39	3.76	3.27	3.38	4.34	4.99	4.72	4.71	5.17	4.05	6.61	5.73	5.53	3.30	4.56	5.66	5.69	4.39	4.36	5.51	4.87	4.79	5.38	
40	5.17	4.53	4.88	5.72	6.70	6.46	5.46	6.19	5.07	7.70	7.20	5.63	5.02	5.86	6.37	6.50	4.91	4.99	7.08	6.03	5.35	5.81	

41 | 6.40 6.16 6.10 6.84 7.40 6.86 6.74 6.69 5.84 7.70 6.93 7.24 5.69 7.17 7.78 7.82 6.43 5.72 5.44 6.18 7.06 6.90

Table 13 continued...

	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40
24	5.62																	
25	6.15	4.20																
26	4.95	5.97	5.82															
27	4.53	4.98	6.61	3.67														
28	6.18	3.99	4.08	5.20	4.67													
29	6.27	4.83	4.54	5.50	5.12	3.20												
30	6.21	4.76	5.42	4.08	3.57	4.06	4.38											
31	6.02	5.34	5.99	4.62	4.43	5.70	6.48	3.42										
32	4.36	5.90	6.78	4.09	3.40	5.65	5.53	4.22	5.00									
33	5.46	6.66	7.14	6.18	6.00	6.23	6.12	6.47	7.75	3.81								
34	4.44	7.44	7.47	6.74	6.76	7.22	7.38	7.64	8.21	4.96	2.99							
35	4.48	8.01	7.86	6.16	6.61	7.57	7.66	7.62	8.03	4.84	3.47	1.66						
36	3.92	7.59	7.80	6.81	6.36	7.47	8.01	7.61	7.98	5.62	5.12	3.09	3.24					
37	4.05	7.86	7.87	6.65	6.49	7.65	7.88	7.66	8.15	5.19	4.37	2.33	2.24	1.36				
38	4.19	7.30	7.90	6.57	6.13	7.19	7.66	7.64	8.19	5.02	3.68	2.17	2.53	2.27	2.11			
39	4.23	6.32	6.83	5.36	5.31	5.78	6.83	6.14	6.71	4.21	3.34	2.90	3.00	3.29	3.26	2.47		
40	4.27	7.54	7.93	6.38	6.23	7.48	7.80	7.55	8.06	4.96	3.65	2.15	1.79	2.33	1.61	1.63	2.68	
41	5.17	8.86	8.48	5.59	6.63	8.06	8.99	7.29	6.95	5.80	6.70	5.50	4.64	4.55	4.61	5.25	4.21	4.87

1	Aamo 5	11	TNPsc 156	21	TNPsc 233	31	Venganoor	41	TNPsc 170
2	Aamo 13	12	TNPsc 176	22	TNPsc 235	32	CO3		
3	Aamo 126	13	TNPsc 183	23	TNPsc 236	33	Aamo 8		
4	Aamo 210	14	TNPsc 201	24	Adari	34	Aamo 10		
5	Aamo 258	15	TNPsc 215	25	Athipatti	35	Aamo 29		
6	RK 50	16	TNPsc 217	26	Naraiyur	36	Aamo 36		
7	Sel 21	17	TNPsc 221	27	Pacheri	37	Aamo 37		
8	TNPsc 86	18	TNPsc 225	28	Podivargu	38	Aamo 72		
9	TNPsc 147	19	TNPsc 231	29	Peruvaragu	39	Aamo 398		
10	TNPsc 155	20	TNPsc 232	30	Thittakudi	40	TNPsc 106		

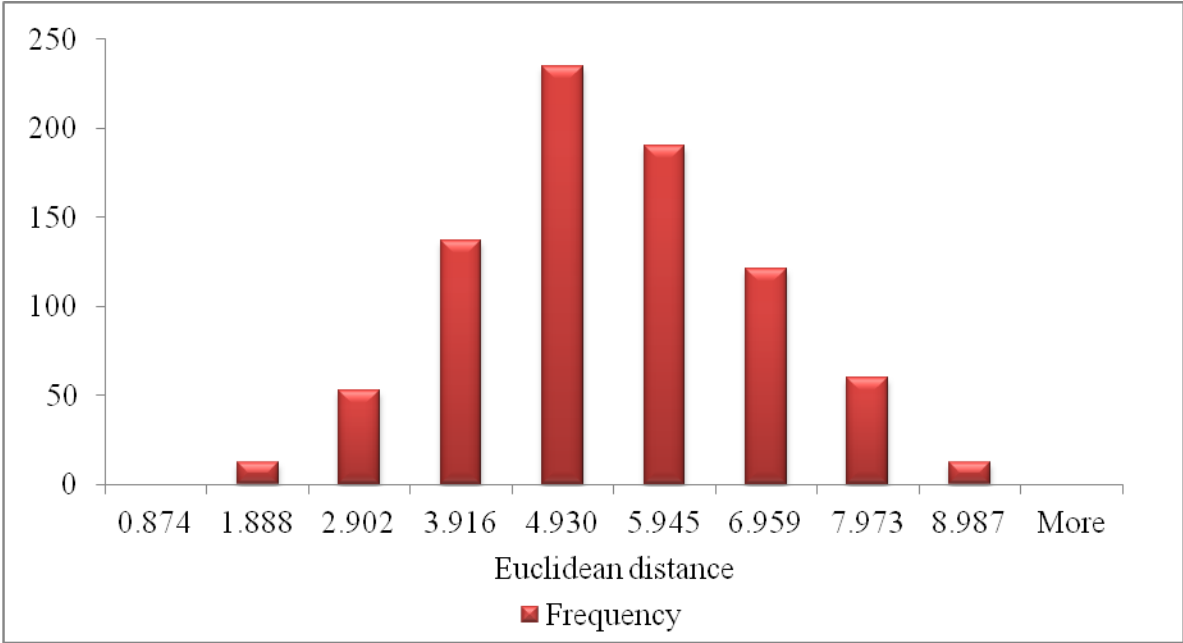


Figure 3. Frequency distribution of Euclidean distance between 41 kodo millet genotypes for 13 quantitative traits [Summer (Jan. - May), 2013]

4.2.4.1 Hierarchical Cluster Analysis (HCA)

The result of HCA utilizing 13 quantitative traits on 41 genotypes is presented as a tree (Fig. 4). Five clusters could be recognized from the tree. The composition of each cluster is furnished in table 14. The cluster I was the largest of all with 20 genotypes and cluster II, III and IV are moderate in size with 5, 6 and 9 genotypes, respectively. Cluster V had only one genotype. The cluster mean values for each trait are presented in table 15.

4.2.4.1.1 Cluster means of the traits

Cluster means for the 13 morphological traits are presented in table 15.

Cluster I

This cluster exhibited the maximum mean values for raceme number (3.30), grain yield per plant (17.31 g) and test weight (4.82 g) (Table 15).

Cluster II

This cluster showed the maximum values for days to 50 per cent flowering (97.30 days), plant height (65.82 cm), culm diameter (5.08 mm), pulling force (4.82 N) and culm weight per unit length (20.07 mg/cm). It showed the minimum values for internode length (9.98 cm) and grain yield per plant (9.01 g) (Table 15).

Cluster III

This cluster recorded the maximum values for inflorescence length (16.79 cm), raceme length (9.33 cm), recovery angle after bending (49.79°) and days to maturity (134.08 days) (Table 15).

Cluster IV

This cluster registered only the minimum values for traits such as plant height (40.40 cm), inflorescence length (12.63 cm) and raceme length (6.51 cm) (Table 15).

Cluster V

This cluster which had only one genotype was with minimum values for most of the traits such as days to 50 per cent flowering (65.5 days), raceme number (2.00), culm diameter (2.49 mm), pulling force (1.36N), culm weight per unit length (5.70 mg/cm),

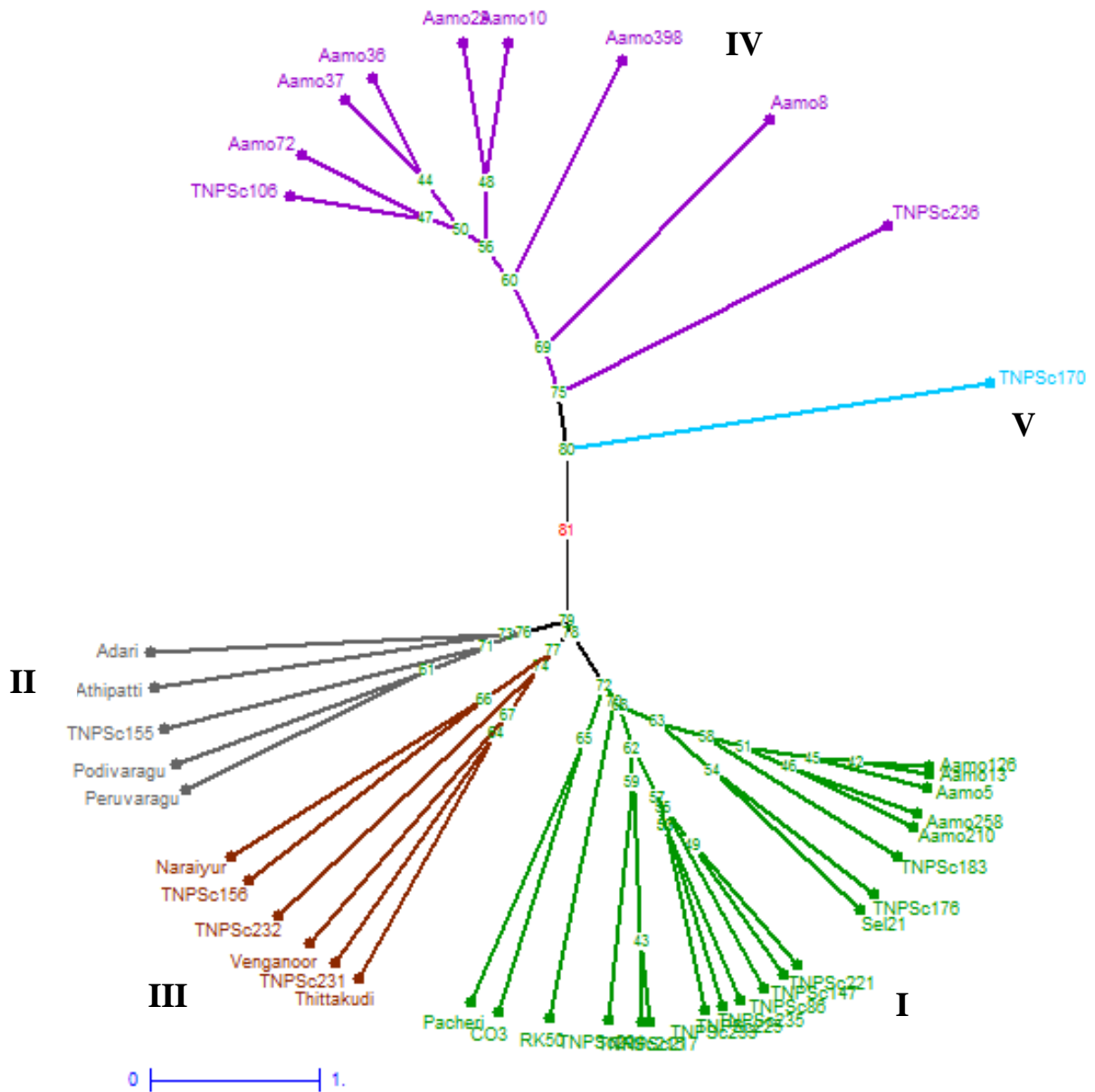


Figure 4. Clustering pattern of 41 kodo millet accessions based on Euclidean distances [Summer (Jan. - May), 2013]

Kodo millet germplams and land races with high culm strength were grouped under cluster II and III. Weak culm lines with high degree of lodging fall into cluster IV and V

Table 14. Cluster composition of 41 kodo millet accessions for 13 morphological traits [Summer (Jan.-May), 2013]

Cluster	Number of genotypes	Genotypes
I	20	Aamo 5, Aamo 13, Aamo 126, Aamo 210, Aamo 258, RK 50, Sel 21, TNPSc 86, TNPSc 147, TNPSc 176, TNPSc 183, TNPSc 201, TNPSc 215, TNPSc 217, TNPSc 221, TNPSc 225, TNPSc 233, TNPSc 235, <i>Pacheri</i> , CO3
II	5	TNPSc 155, <i>Adari</i> , <i>Athipatti</i> , <i>Podivaragu</i> , <i>Peruvaragu</i>
III	6	TNPSc 156, TNPSc 231, TNPSc 232, <i>Naraiyur</i> , <i>Thittakudi</i> , <i>Venganoor</i>
IV	9	TNPSc 106, TNPSc 236, Aamo 8, Aamo 10, Aamo 29, Aamo 36, Aamo37, Aamo 72, Aamo 398
V	1	TNPSc 170

Table 15. Cluster mean for 13 morphological traits in 41 kodo millet germplasm accessions [Summer (Jan.-May), 2013]

Cluster	DF	PH	InfloL	RN	RL	CD	PF	WL	IntL	RA	DM	GY	TW
I	85.03	52.76	14.58	3.30	7.91	3.79	4.18	10.74	10.66	46.02	124.63	17.31	4.82
II	97.30	65.82	15.17	3.13	8.03	5.08	4.82	20.07	9.98	49.50	131.90	9.01	4.67
III	90.92	57.81	16.79	2.71	9.33	3.57	3.79	14.58	13.74	49.79	134.08	16.34	4.53
IV	67.89	40.40	12.63	2.79	6.51	2.77	1.66	6.38	10.49	41.11	113.22	9.63	3.96
V	65.5	47.67	15.83	2.00	6.92	2.49	1.36	5.70	16.17	25.83	111.50	14.35	3.71

(maximum value for each trait is given in bold).

Abbreviations used: DF- Days to 50 per cent flowering (days); PH- Plant height (cm); InfloL- Inflorescence length (cm); RN- Raceme number (count); RL- Raceme length (cm); CD- Culm diameter (mm); PF- Pulling force (N); WL- Culm weight per unit length (mg/cm); IntL- Internode length (cm); RA- Recovery angle after bending (degrees); DM- Days to maturity (days); GY- Grain yield per plant (g); TW- Test weight (g)

recovery angle after bending (25.83°), days to maturity (111.50 days) and test weight (3.71 g) (Table 15).

4.2.4.2 Principal component analysis (PCA)

Grouping of the traits through PCA was carried out to examine the percentage contribution of each trait to total genetic variation. The PCA revealed that the first four principal components accounted for 39, 16, 9, and 9 per cent of total variability, respectively and cumulatively accounted for 74 per cent (Table 16). The first principal component correlated with days to 50 per cent flowering (-0.39), plant height (-0.38) and pulling force (-0.34). The traits such as inflorescence length (0.38), raceme number (-0.37), raceme length (0.38) and culm diameter (-0.34) contributed more to the second component. Grain yield per plant (-0.80) alone made substantial contribution to the third component. The fourth component was highly contributed by culm weight per unit length (-0.45) and test weight (0.54).

The scatter plot of the first Principal Components Axis (PC1) against the second Principal Component Axis (PC2) revealed that all the 41 kodo millet accessions were ordered into five distinct PCA clusters which were in conformity with the clusters obtained through hierarchical cluster analysis, except for the land race *Pacheri*, which got grouped in cluster III instead of cluster I (Fig. 5).

4.3 Analysis of select biochemical parameters for culm strength and evaluation for yield [Kharif, (Aug. - Dec.), 2013]

4.3.1 Mean performance of genotypes for 15 quantitative traits

The result of analysis of variance on 15 quantitative traits for the 22 genotypes (13 lines selected from previous season based on yield and strength, two checks, CO3 and *Adari*, and *Athipatti*, identified as having good culm strength, but a poor yielder and six weak culm lines) is presented in table 17. Mean square of all the characters studied, showed significant difference ($P < 0.05$) among the tested genotypes except for number of productive tillers per plant, indicating the presence of variability for the remaining 14 traits. Hence, subsequent analysis was carried out excluding the trait, number of productive tillers per plant.

Table 16. Loadings of the 13 quantitative traits onto four principal components, with eigen values and variance [Summer (Jan.-May), 2013]

Variables	Comp 1	Comp 2	Comp 3	Comp 4
Days to 50 per cent flowering	-0.39	0.06	0.26	0.12
Plant height	-0.38	0.03	0.03	-0.18
Inflorescence length	-0.29	0.38	-0.08	0.00
Raceme number	-0.16	-0.37	-0.26	0.17
Raceme length	-0.31	0.38	-0.17	0.04
Culm diameter	-0.31	-0.34	-0.02	-0.30
Pulling force	-0.34	-0.20	-0.05	0.25
Culm weight per unit length	-0.29	-0.11	0.24	-0.45
Internode length	-0.02	0.54	-0.25	-0.09
Recovery angle after bending	-0.16	-0.13	-0.11	-0.48
Days to maturity	-0.32	0.23	0.23	0.18
Grain yield per plant	-0.10	-0.06	-0.80	-0.05
Test weight	-0.27	-0.20	-0.03	0.54
Variance %	39	16	9	9
Cumulative variance %	39	55	64	73
Eigen value	4.76	1.95	1.15	1.05

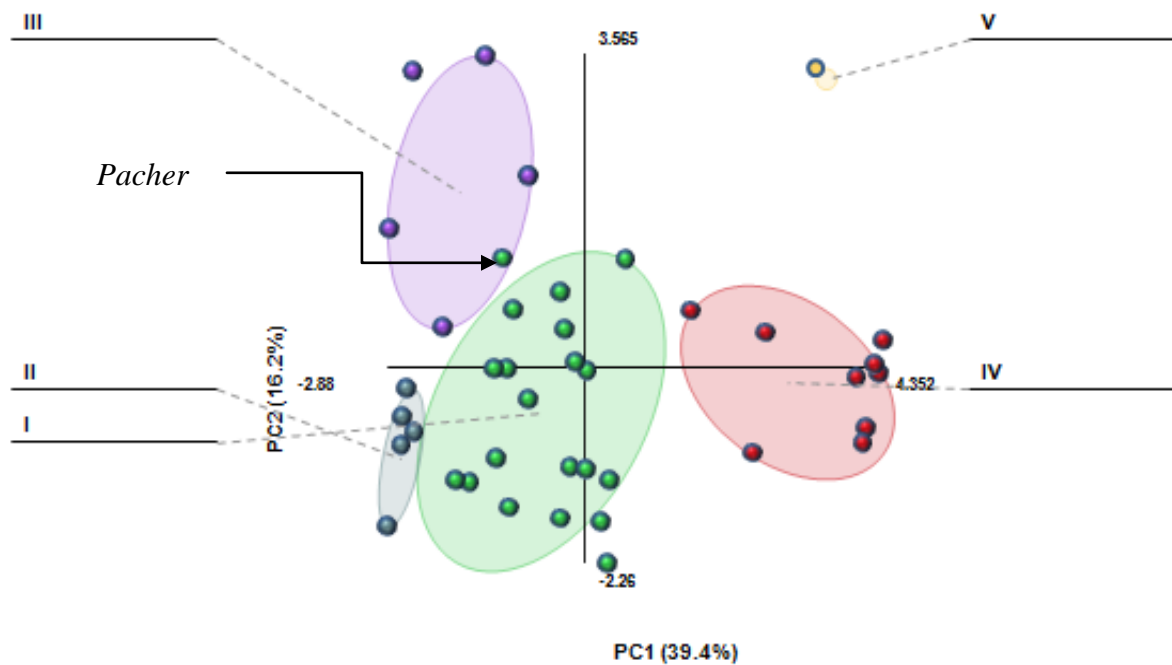


Figure 5. 2D plot based on correlation of each germplasm accession with the two principal components [Summer (Jan. - May), 2013]

The PCA clusters were in conformity with the clusters obtained through HCA, except for the land race, Pacheri, which got grouped in cluster III instead of cluster I.

Table 17. ANOVA for 15 traits in 22 kodo millet germplasm accessions in RBD [Kharif, (Aug.- Dec.), 2013]

Sl. No.	Traits	Mean sum of squares		
		Replications (df=1)	Treatments (df=21)	Error (df=21)
1	Days to 50 per cent flowering	1.455	296.939**	2.121
2	Plant height	17.413**	257.425**	9.437
3	Numbers of productive tillers	1.224	1.331	1.134
4	Inflorescence length	1.397	2.692**	0.421
5	Number of racemes	0.003	0.941**	0.179
6	Raceme length	0.984	1.666**	0.369
7	Culm diameter	0.146	1.335**	0.071
8	Pulling force	0.027	1.655**	0.392
9	Culm weight per unit length	0.110	100.919**	2.273
10	Internode length	5.005	8.035**	1.457
11	Recovery angle after bending	39.463	275.797**	35.694
12	Days to maturity	149.114*	183.616**	25.018
13	Grain yield per plant	4.259	23.926**	6.025
14	Grain yield per plot	0.010	0.28**	0.030
15	Test weight	0.000	1.033**	0.059

*significant at 5% level; **significant at 1% level

Mean performance 22 germplasm accessions are presented in table 18.

4.3.1.1 Days to 50 per cent flowering

Days to 50 per cent flowering ranged between 59.50 days (Aamo 10) and 99.50 days (*Athipatti*). The entries *viz.*, Aamo 5, Aamo 13, Aamo 126, Aamo 210, Aamo 258, Sel 21, TNPsc 176, TNPsc 106, TNPsc 170, Aamo 8, Aamo 10, Aamo 36, Aamo 398 were significantly earlier than the check, CO3 and TNPsc 156, *Peruvaragu*, *Thittakudi* and *Athipatti* were significantly longer in duration than the check. The general mean for this trait was 77.77 days (Table 18).

4.3.1.2 Plant height

Among the test entries, the genotypes, Aamo 398 had the lowest value (34.25 cm) for plant height. The maximum for this trait was recorded in *Thittakudi* (72.00 cm). The general mean recorded was 54.70 cm. Four test entries *viz.*, Aamo 13, Aamo 258, Sel 21, TNPsc 176 and six weak culm lines *viz.*, TNPsc 106, TNPsc 170, Aamo 8, Aamo 10, Aamo 36 and Aamo 398 were significantly shorter than the check CO3 and only *Thittakudi* was significantly taller than CO3 (Table 18).

4.3.1.3 Number of productive tillers per plant

No significant differences existed between genotypes for this trait. The overall mean for number of productive tillers per plant was 7.96 (Table 18).

4.3.1.4 Inflorescence length

General mean recorded for this trait was 8.36 cm; the maximum was recorded by TNPsc 156 with 10.42 cm length and the minimum was recorded in Aamo 398 with inflorescence length of 6.25 cm. None of the genotypes had significantly longer inflorescence than the check CO3, but the test entries *viz.*, Sel 21 and TNPsc 176 and weak culm lines *viz.*, TNPsc 106, TNPsc 170, Aamo 8, Aamo 10, Aamo 36 and Aamo 398 had significantly shorter inflorescence length than the check (Table 18).

4.3.1.5 Raceme length

The maximum raceme length of 8.12 cm was noticed in TNPsc 201 and the minimum raceme length of 4.88 cm was observed in TNPsc 106 and none of the

Table 18. Mean performance of 22 kodo millet germplasm accessions for 15 quantitative traits [Kharif, (Aug.- Dec.), 2013]

Sl No	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
		DF	PH	Ptil	InfloL	RL	RN	CD	PF	WL	IntL	RA	DM	GY	YP	TW
Selected entries																
1	Aamo 5	72.50	61.17	8.50	9.34	7.33	3.00	3.58	2.52	12.11	17.34	45.84	102.50	17.65	2.09	5.41
2	Aamo 13	74.00	54.00	6.17	8.00	6.67	3.00	3.21	2.85	12.74	14.92	50.00	123.50	8.42	1.96	5.29
3	Aamo 126	75.50	58.33	7.84	8.42	7.17	2.84	3.34	1.97	13.30	16.84	50.00	121.00	9.30	1.97	5.40
4	Aamo 210	73.50	61.67	8.34	8.92	7.34	3.17	3.69	1.75	14.09	15.83	52.50	103.50	12.19	1.81	5.46
5	Aamo 258	75.00	53.17	8.50	8.42	6.67	2.84	3.11	2.52	12.22	15.75	43.33	112.00	10.61	1.90	5.40
6	RK 50	83.50	59.50	8.50	9.34	6.84	4.17	3.41	2.32	9.84	15.17	52.50	118.00	9.92	1.65	5.83
7	Sel 21	75.00	51.67	7.67	7.34	5.50	3.00	2.68	2.35	9.80	14.00	57.50	113.50	9.23	2.35	5.87
8	TNPSc 156	91.00	66.50	6.84	10.42	7.75	3.50	3.42	3.31	21.59	13.17	40.00	125.50	6.66	0.71	5.84
9	TNPSc 176	73.00	50.67	7.67	7.42	6.25	2.67	2.88	1.40	9.73	13.59	50.00	105.00	9.99	2.25	5.76
10	TNPSc 183	82.00	60.17	7.50	8.92	7.34	2.67	2.90	1.73	8.62	14.17	39.17	108.00	7.08	2.12	5.49
11	TNPSc 201	85.00	66.83	8.34	9.59	8.12	4.17	3.78	2.11	20.55	15.75	45.84	118.00	15.14	1.83	5.58
12	<i>Peruvaragu</i>	93.00	61.17	8.83	8.59	7.09	3.00	4.02	3.65	16.75	12.84	45.00	123.50	8.05	1.56	5.56
13	<i>Thittakudi</i>	95.50	72.00	8.50	8.87	6.67	3.17	3.65	2.11	12.74	15.83	48.33	119.50	6.32	1.07	5.97
Checks																
14	CO3 (Check)	84.50	62.17	8.50	9.25	7.00	2.67	2.85	2.44	6.45	15.34	55.00	118.00	4.79	1.54	5.91
15	<i>Adari</i> (check)	98.00	65.50	8.50	9.50	8.00	5.00	4.06	3.39	22.50	14.00	42.50	135.00	4.58	1.44	4.67

Table 18 continued...

Sl No	Genotypes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
		DF	PH	Pro T	Inflo L	RL	RN	CD	PF	WL	Int L	RA	DM	G Y	YP	T W
Lines included for comparison																
16	<i>Athipatti</i> (strong culm)	99.5 0	66.7 5	9.00	9.88	7.7 5	3.5 0	4.7 8	3.3 5	33.2 9	12.2 5	37.5 0	129.5 0	5.8 7	1.5 6	4.9 9
17	Aamo8 (weak culm)	64.0 0	41.0 0	7.50	7.75	5.7 5	2.2 5	2.2 0	0.8 3	4.53	11.2 5	20.0 0	104.0 0	8.2 6	1.6 8	3.9 7
18	Aamo10 (weak culm)	59.5 0	37.7 5	6.75	6.88	5.5 0	2.5 0	1.7 5	1.0 7	6.30	13.2 5	23.7 5	105.0 0	4.4 9	1.5 0	4.2 4
19	Aamo36 (weak culm)	63.5 0	41.0 0	7.50	7.13	6.6 3	2.2 5	2.2 2	0.7 1	5.62	11.2 5	33.7 5	111.0 0	5.9 0	1.5 7	4.2 6
20	Aamo398 (weak culm)	64.5 0	34.2 5	9.50	6.25	5.2 5	2.7 5	1.9 6	1.2 2	6.74	13.0 0	33.7 5	105.0 0	6.7 5	1.6 1	4.0 6
21	TNPsc106 (weak culm)	62.5 0	37.2 5	7.00	6.50	4.8 8	2.2 5	1.7 3	0.8 4	5.03	11.5 0	18.7 5	104.0 0	4.5 4	1.5 0	4.7 0
22	TNPsc170 (weak culm)	66.5 0	41.0 0	7.75	7.25	6.0 0	2.5 0	2.1 2	0.8 8	5.61	9.50	20.0 0	105.5 0	4.7 6	1.5 1	3.6 3
	Mean	77.7 7	54.7 0	7.96	8.36	6.7 0	3.0 4	3.0 6	2.0 6	12.2 8	13.9 3	41.1 4	114.1 1	8.2 0	1.6 9	5.1 5
	SE	1.46	3.07	1.06	0.65	0.6 1	0.4 2	0.2 7	0.6 3	1.51	1.21	5.97	5.00	2.4 5	0.1 7	0.2 4
	CD (0.05%)	3.03	6.39	NA	1.35	1.2 6	0.8 8	0.5 6	1.3 0	3.14	2.51	12.4 3	10.40	5.1 1	0.3 6	0.5 1

Abbreviations used: DF- Days to 50 per cent flowering (days); PH- Plant height (cm); ProT- Number of productive tillers (count); InfloL- Inflorescence length (cm); RN- Raceme number (count); RL- Raceme length (cm); CD- Culm diameter (mm); PF- Pulling force (N); WL- Culm weight per unit length (mg/cm); IntL- Internode length (cm); RA- Recovery angle after bending (degrees); DM- Days to maturity (days); GY- Grain yield per plant (g); YP- Yield per plot (Kg); TW- Test weight (g).

genotypes exceeded the check, significantly in raceme length. The general mean for this trait was 6.70 cm (Table 18).

4.3.1.6 Raceme number

The general mean observed for the genotypes studied was 3.04, with highest number (5.00) was registered by *Adari* and the least (2.25) by TNPsc 106, Aamo 8 and Aamo 36, the weak lines. Among the test entries, RK 50, TNPsc 201 and *Adari* had significantly greater number of racemes than the check, CO3 (Table 18).

4.3.1.7 Culm diameter

The line *Athipatti*, recorded the highest value (4.78 mm) where as TNPsc 106 recorded the least (1.73 mm) of all. Only *Athipatti* had significantly higher culm diameter than the check *Adari*. But the entries, Aamo 5, Aamo 210, TNPsc 201, *Peruvaragu* and *Thittakudi* were on par with the check, *Adari*, with respect to culm diameter (Table 18).

4.3.1.8 Pulling force

The range for this trait was 0.71 (Aamo 36) to 3.65 N (*Peruvaragu*), with a mean of 2.06 N. Though none of the lines excelled significantly, the test entries, viz., Aamo 5, Aamo 13, Aamo 258, RK 50, Sel 21, TNPsc 156, TNPsc 201, CO3, *Peruvaragu*, *Thittakudi* and *Athipatti* were on par with the check *Adari*, with respect to this trait (Table 18).

4.3.1.9 Culm weight per unit length

This trait ranged between 4.53 mg/cm (Aamo 8) to 33.29 mg/cm (*Athipatti*). The general mean recorded for this trait was 12.28 mg/cm. Only *Athipatti* had significantly better value than the check *Adari*, but TNPsc 156 and TNPsc 201 were on par with the check *Adari* with respect to this trait (Table 18).

4.3.1.10 Recovery angle after bending

The range registered for this trait among the 22 genotypes was 18.75° (TNPsc 106) to 57.50° (Sel 21) with an overall mean of 41.14°. The entries Sel 21 and CO3 had significantly better recovery than the check *Adari* (Table 18).

4.3.1.11 Internode length

The internode length ranged from 9.50 cm (TNPsc 170) to 17.34 cm (Aamo 5). The average recorded for this trait was 13.93 cm. The weak culm genotypes, *viz.*, Aamo 8, Aamo 36, TNPsc 170 had shorter internode than the check *Adari* (Table 18).

4.3.1.12 Days to maturity

The genotype Aamo 5 was the earliest to mature with 102.50 days duration and the check, *Adari* recorded a longest duration of 135 days among the entries studied. The mean for this trait was around 114.11 days. *Adari* and *Athipatti* were significantly late to mature than the check CO3 and three test entries, *viz.*, Aamo 5, Aamo 210, TNPsc 176 and five weak culm lines *viz.*, Aamo 8, Aamo 10, Aamo 398 and TNPsc 106, TNPsc 170 were significantly early to mature when compared with the check, CO3 (Table 18).

4.3.1.13 Grain yield per plant

Among the test entries, the highest yielder was Aamo 5 with grain yield of 17.65 g per plant and the poorest yielder being Aamo 10 (4.49 g). The average yield for all the genotypes was 8.20 g. The test entries, Aamo 5, Aamo 210, Aamo 258, RK 50, TNPsc 176, and TNPsc 201 were identified as higher yielders when compared with the check, CO3 (Table 18).

4.3.1.14 Grain yield per plot

The range recorded for this trait was 0.710 kg per plot (TNPsc 156) to 2.350 kg per plot (Sel 21) with an overall average of 1.690 kg per plot. The test entries Aamo 5, Aamo 13, Aamo 126, Sel 21, TNPsc 176 and TNPsc 183 were identified as higher yielders when compared with the check, CO3 (Table 18).

4.3.1.15 Test weight

The general mean for this trait was 5.15 g. Mean values registered a range of 3.63 g (TNPsc 170) to 5.97 g (*Thittakudi*). None of the genotypes had better test weight than the check CO3. But the test entries *viz.*, Aamo 5, Aamo 210, RK 50, Sel 21, TNPsc 156, TNPsc 176, TNPsc 183, TNPsc 201, *Peruvaragu* and *Thittakudi* were on par with the check CO3 with respect to the trait test weight (Table 18).

4.3.2 Frequency distribution of eight qualitative traits in 22 genotypes

The frequency distributions for eight qualitative traits which were observed (Table 19) in discrete classes are described below.

4.3.2.1 Plant type

Among the 22 entries, all were erect, except TNPsc 201, which is decumbent in growth habit (Table 19).

4.3.2.2 Ear exertion at dough stage

All the genotypes were with fully exerted ears, except TNPsc 170, which was partial in ear exertion (Table 19)

4.3.2.3 Spikelet arrangement on rachis

Most of the genotypes (11) had regular spikelet arrangement, 3 genotypes had irregular arrangement and the remaining (8) genotypes had variable spikelet arrangement (Table 19).

4.3.2.4 Degree of chlorosis

Among the 22 genotypes, only four exhibited severe chlorosis and the remaining were low (17 genotypes) to intermediate (1 genotype) with respect degree of chlorosis (Table 19).

4.3.2.5 Stay green nature at maturity

Stay green nature at maturity was present in all the lines except *Thittakudi* and CO3, and weak culm lines, Aamo 8, Aamo 10, TNPsc 106 and TNPsc 170 (Table 19).

4.3.2.6 Shattering of inflorescence

All the test entries had non-shattering inflorescence, but among the weak lines, Aamo 8, Aamo 10 and Aamo 398 had shattering of inflorescence (Table 19).

4.3.2.7 Uniformity in maturity

Out of the 22, ten genotypes viz., Aamo 5, Aamo 210, Aamo 258, Sel 21, TNPsc 176, TNPsc 183, Aamo 8, Aamo 398, TNPsc 106 and TNPsc 170 exhibited uniformity in maturity (Table 19).

Table 19. Performance of 22 germplasm accessions of kodo millet for scorable traits
[Kharif, (Aug.- Dec.), 2013]

Sl No	Genotypes	1	2	3	4	5	6	7	8
		PT	EE	SA	DC	SG	SI	UM	DL
Selected entries									
1	Aamo 5	1	1	3	3	1	0	1	5
2	Aamo 13	1	1	2	3	1	0	0	5
3	Aamo 126	1	1	3	3	1	0	0	3
4	Aamo 210	1	1	2	3	1	0	1	5
5	Aamo 258	1	1	3	7	1	0	1	5
6	RK 50	1	1	1	7	1	0	0	5
7	Sel 21	1	1	2	3	1	0	1	5
8	TNPSc 156	1	1	1	5	1	0	0	3
9	TNPSc 176	1	1	3	3	1	0	1	5
10	TNPSc 183	1	1	1	3	1	0	1	3
11	TNPSc 201	2	1	3	3	1	0	0	5
12	<i>Peruvaragu</i>	1	1	1	3	1	0	0	3
13	<i>Thittakudi</i>	1	1	1	7	0	0	0	3
Checks									
14	CO3	1	1	1	7	0	0	0	5
15	Adari	1	1	1	3	1	0	0	3
Lines included for comparison									
16	<i>Athipatti</i> (strong culm)	1	1	1	3	1	0	0	3
17	Aamo8 (weak culm)	1	1	3	3	0	1	1	7
18	Aamo10 (weak culm)	1	1	3	3	0	1	0	7
19	Aamo36 (weak culm)	1	1	3	3	1	0	0	7
20	Aamo398 (weak culm)	1	1	1	3	1	1	1	5
21	TNPsc106 (weak culm)	1	1	1	3	0	0	1	7
22	TNPsc170 (weak culm)	1	2	1	3	0	0	1	7

Abbreviations used: PT- Plant type; EE- Ear exertion at dough stage; SA- Spikelet arrangement on rachis; DC- Degree of chlorosis; SG- Stay green nature at maturity; SI- Shattering of inflorescence; UM- Uniformity in maturity; DL- Degree of lodging

4.3.2.8 Degree of lodging

Adari, *Thittakudi*, *Athipatti*, *Peruvaragu*, TNPsc 183, TNPsc 156 and Aamo 126 had low degree of lodging. Other entries had intermediate (10 genotypes) to severe (five genotypes) lodging scores (Table 19).

4.3.3 Mean performance of genotypes for biochemical traits

The result of ANOVA on six biochemical traits for the 22 genotypes (13 selected, two checks and seven genotypes extremes with respect to culm strength) is presented in table 20. Mean square of the traits, cellulose, hemicellulose and lignin were not significant among the 22 genotypes studied, but the genotypes were significantly different with respect to traits such as cellulose, hemicellulose and lignin per unit length of the culm. Hence, further analysis in these genotypes was done based on these three traits alone.

Mean performance of 22 germplasm accessions with respect to three biochemical traits are presented in table 21.

4.3.3.1 Cellulose per unit length of the culm

The range observed among the 22 genotypes was 0.70 (Aamo 36) to 6.42 mg/cm (*Athipatti*). The average for this trait was 2.05 mg/cm. Only *Athipatti* registered significantly higher value for this trait, others were on par with the check for culm strength, *Adari* (Table 21).

4.3.3.2 Hemicellulose per unit length of the culm

The highest hemicellulose content was recorded in the genotype, *Athipatti* (10.93 mg/cm) and the least content was recorded in TNPsc 106 (0.93 mg/cm) with the overall mean of 3.98 mg/cm. The genotypes, TNPsc 156, TNPsc 176 and *Athipatti* were on par with *Adari* in hemicellulose content. But none had significantly higher content than the check, *Adari* (Table 21).

4.3.3.3 Lignin per unit length of the culm

The overall mean observed was 2.47 mg/cm of dry culm. This trait ranged from 0.78 mg/cm (TNPsc 106) to 6.98 mg/cm (*Athipatti*). Though none had significantly

Table 20. ANOVA for six bio-chemical traits in 22 germplasm accessions of kodo millet [Kharif, (Aug.- Dec.), 2013]

Sl. No.	Source	Mean sum of squares		
		Replications	Treatments	Error
		(df=1)	(df=21)	(df=21)
1	Cellulose (%)	0.00	55.86	46.09
2	Hemicellulose (%)	270.57	52.58	47.54
3	Lignin (%)	158.95	23.91	40.71
4	Cellulose per unit length of the culm	0.31	2.87*	1.02
5	Hemicellulose per unit length of the culm	4.94	11.97**	1.61
6	Lignin per unit length of the culm	0.52	5.34**	1.42

*significant at 5% level; **significant at 1% level

Table 21. Mean performance for biochemical traits in 22 kodo millet germplasm accessions [Kharif, (Aug.- Dec.), 2013]

Sl. No.	Geneotypes	Cellu-lose (%)	Hemi-cellulose (%)	Lignin (%)	Cellulose per unit length of the culm (mg/cm)	Hemi-cellulose per unit length of the culm (mg/cm)	Lignin per unit length of the culm (mg/cm)
Selected lines							
1	Aamo 5	13.93	31.55	18.71	1.69	4.17	2.32
2	Aamo 13	14.57	25.98	17.64	1.83	3.10	2.18
3	Aamo 126	13.15	31.70	21.96	1.74	4.62	3.19
4	Aamo 210	13.31	37.78	16.58	1.85	5.49	2.41
5	Aamo 258	15.13	30.55	15.76	1.86	3.20	1.66
6	RK 50	27.57	23.99	16.51	2.66	2.33	1.64
7	Sel 21	17.78	26.97	19.91	1.75	2.57	1.90
8	TNPSc 156	16.18	30.75	22.98	3.29	5.82	4.28
9	TNPSc 176	19.58	22.54	22.78	1.83	2.64	2.73
10	TNPSc 183	25.01	24.86	20.87	2.13	2.36	1.81
11	TNPSc 201	14.39	30.22	19.82	2.93	6.26	4.06
12	<i>Peruvaragu</i>	20.33	26.87	19.62	3.32	5.69	4.02
13	<i>Thittakudi</i>	14.30	34.96	23.32	1.81	4.98	3.41
Checks							
14	CO3	26.14	28.86	14.53	1.71	2.13	1.04
15	<i>Adari</i>	7.80	38.96	23.84	1.77	8.41	5.15
Additional lines chosen for comparison							
16	<i>Athipatti</i> (strong culm)	22.09	30.93	20.91	6.41	10.93	6.98
17	Aamo8 (weak culm)	25.73	34.42	10.05	1.44	4.23	0.84
18	Aamo10 (weak culm)	14.91	33.66	14.20	1.02	2.64	1.11
19	Aamo36 (weak culm)	15.19	38.06	17.29	0.69	2.66	0.97
20	Aamo398 (weak culm)	17.94	31.71	17.27	1.01	1.29	0.91
21	TNPsc106 (weak culm)	19.97	25.11	20.43	0.93	0.92	0.78
22	TNPsc170 (weak culm)	24.36	20.00	16.25	1.37	1.06	0.87
Mean		18.15	30.02	18.69	2.05	3.98	2.47
SE		6.79	6.89	6.38	1.01	1.27	1.19
CD (0.05%)		NA	NA	NA	2.10	2.64	2.48

NA: Not applicable, since treatments are insignificant.

higher lignin than the check, *Adari*, the genotypes TNPsc 156, TNPsc 176, TNPsc 201, *Peruvaragu*, *Thittakudi* and *Athipatti* were at par with the check, with respect to this trait (Table 21).

4.3.4 Association studies

Estimation of phenotypic correlation coefficients and the direct and indirect effects of these traits on one another was studied using the mean data.

4.3.4.1 Correlation between degree of lodging and other culm strength related traits

The traits for this study included eight culm related traits *viz.*, cellulose per unit length of the culm, hemicellulose per unit length of the culm, lignin per unit length of the culm, pulling force, culm weight per unit length, culm diameter, recovery angle after bending, degree of lodging.

Six traits *viz.*, cellulose per unit length of the culm, hemicellulose per unit length of the culm, lignin per unit length of the culm, pulling force, culm weight per unit length, culm diameter had significant positive inter-correlation among themselves (Table 22).

The recovery angle after bending had significant positive correlation with pulling force (0.57) and culm diameter (0.58) alone (Table 22).

Degree of lodging had significant negative correlation with remaining seven traits (Table 22) *viz.*, cellulose per unit length of the culm (-0.56), hemicellulose per unit length of the culm (-0.63), lignin per unit length of the culm (-0.75), pulling force (-0.71), culm weight per unit length (-0.69), culm diameter (-0.78), recovery angle after bending (-0.58).

4.3.4.2 Path analysis

In this study, path analysis was carried out to understand the direct and indirect effects of associated traits with degree of lodging (Table 23), with degree of lodging as dependent variable and remaining seven traits as independent variables.

The maximum negative direct effect on degree of lodging was recorded by lignin content per unit length of the culm (-1.341, very high) followed by culm diameter (-0.314, high), pulling force (-0.137, low) and recovery angle after bending (-0.184, low).

Table 22. Correlation co-efficient between culm related traits using 22 genotypes of kodo millet [Kharif, (Aug.- Dec.), 2012]

Traits	CC	HC	LC	PF	WL	CD	RA
HC	0.76**						
LC	0.81**	0.93**					
PF	0.66**	0.65**	0.74**				
WL	0.85**	0.92**	0.97**	0.77**			
CD	0.76**	0.84**	0.87**	0.83**	0.87**		
RA	0.22	0.20	0.31	0.57**	0.29	0.58**	
DL	-0.56**	-0.63**	-0.75**	-0.71**	-0.69**	-0.78**	-0.58**

*significant at 5% level; **significant at 1% level

Table 23. Direct and indirect effects of different traits on pulling force [Kharif, (Aug.- Dec.), 2012]

Traits	Effect via							Correlation (r _p) with degree of lodging
	CC	HC	LC	PF	WL	CD	RA	
CC	0.059	0.241	-1.086	-0.091	0.596	-0.238	-0.040	-0.56**
HC	0.045	0.317	-1.247	-0.089	0.645	-0.264	-0.037	-0.63**
LC	0.048	0.295	-1.341	-0.101	0.680	-0.273	-0.057	-0.75**
PF	0.039	0.206	-0.992	-0.137	0.540	-0.260	-0.105	-0.71**
WL	0.050	0.291	-1.300	-0.106	0.701	-0.273	-0.053	-0.69**
CD	0.045	0.266	-1.166	-0.114	0.610	-0.314	-0.107	-0.78**
RA	0.013	0.063	-0.416	-0.078	0.203	-0.182	-0.184	-0.58**

Residual effect = 0.512

*significant at 5% level; **significant at 1% level

Abbreviations used: CC- Cellulose per unit length of the culm; HC- Hemicellulose per unit length of the culm; LC- Lignin per unit length of the culm; PF- Pulling force ;WL- Culm weight per unit length; CD- Culm diameter; RA- Recovery angle after bending; DL- Degree of lodging.

The maximum positive direct effect on degree of lodging was recorded by culm weight per unit length (0.701, high), followed by hemicellulose per unit length of the culm (0.317, high) and cellulose per unit length of the culm (0.059, negligible)

The indirect effect of cellulose per unit length of the culm (-1.086, very high), hemicellulose per unit length of the culm (-1.247, very high), pulling force (-0.992, high), culm weight per unit length (-1.300, very high), culm diameter (-1.166, very high) and recovery angle after bending (-0.416, high) were maximum through lignin content per unit length of the culm.

4.4 Assessment of yield and degree of lodging in six selected kodo millet genotypes [Summer (Jan. - May), 2014]

4.4.1 Yield assessment in six Kodo millet genotypes

Analysis of variance for yield per plot showed significant variation between genotypes and replications (Table 24). Among the test entries, the genotype Sel 21 recorded the highest yield (2.853 Kg/plot), followed by TNPsc 176 (2.789 Kg/plot) and TNPsc 183 (2.585 Kg/plot). Moreover, these three entries were significantly higher than the check, CO3 in yield performance. The weak culm line registered a yield, which was on par with the check, CO3. But *Adari*, the land race recorded significantly lower yield than the yield check CO3 (Table 25).

4.4.2 Degree of lodging

Among the three test entries the genotypes, Sel 21 and TNPsc 176 and the weak line Aamo 10 exhibited severe lodging (score 7) and the checks, CO3 and *Adari* showed moderate incidence of lodging (score 5). The test entry TNPsc 183 alone showed low degree of lodging (score 3) (Table 25).

4.5 Anatomy and elemental analysis for culm strength in select kodo millet genotypes [Summer (Jan. - May), 2014]

4.5.1 Anatomical observations

General observations for cellular details using light microscopy was carried out in all the six genotypes, *ie.*, three test entries, *viz.*, Sel 21, TNPsc 176, TNPsc 183, two checks, *viz.*, CO3 and *Adari* and one weak culm line Aamo 10 (Plates 13; 14).

Table 24. ANOVA for yield per plot in six germplasm accessions of kodo millet [Summer (Jan.-May), 2014]

Sl.No	Source	Degrees of freedom	Sum of squares	Mean sum of squares
1	Replications	3	0.253	0.084**
2	Treatments	5	5.418	1.084**
3	Error	15	0.598	0.04

*significant at 5% level; **significant at 1% level

Table 25. Mean performance for yield per plot and degree of lodging in six germplasm accessions of kodo millet [Summer (Jan.-May), 2014]

Sl. No	Genotypes	Yield (Kg/plot)	Degree of lodging (score)	Remarks
1	TNPsc 176	2.789	7	lodged severely during grain filling
2	TNPsc 183	2.585	3	low incidence of lodging in two out of four plots towards maturity
3	Sel 21	2.853	7	lodged severely during grain filling
4	Aamo 10 (weak culm line)	2.129	7	lodged severely after heading
5	Adari (Check)	1.434	5	moderately lodged during grain filling
6	CO3 (check)	1.966	5	moderately lodged during maturity
Mean		2.292		
SE		0.141		
CD (0.05%)		0.301		

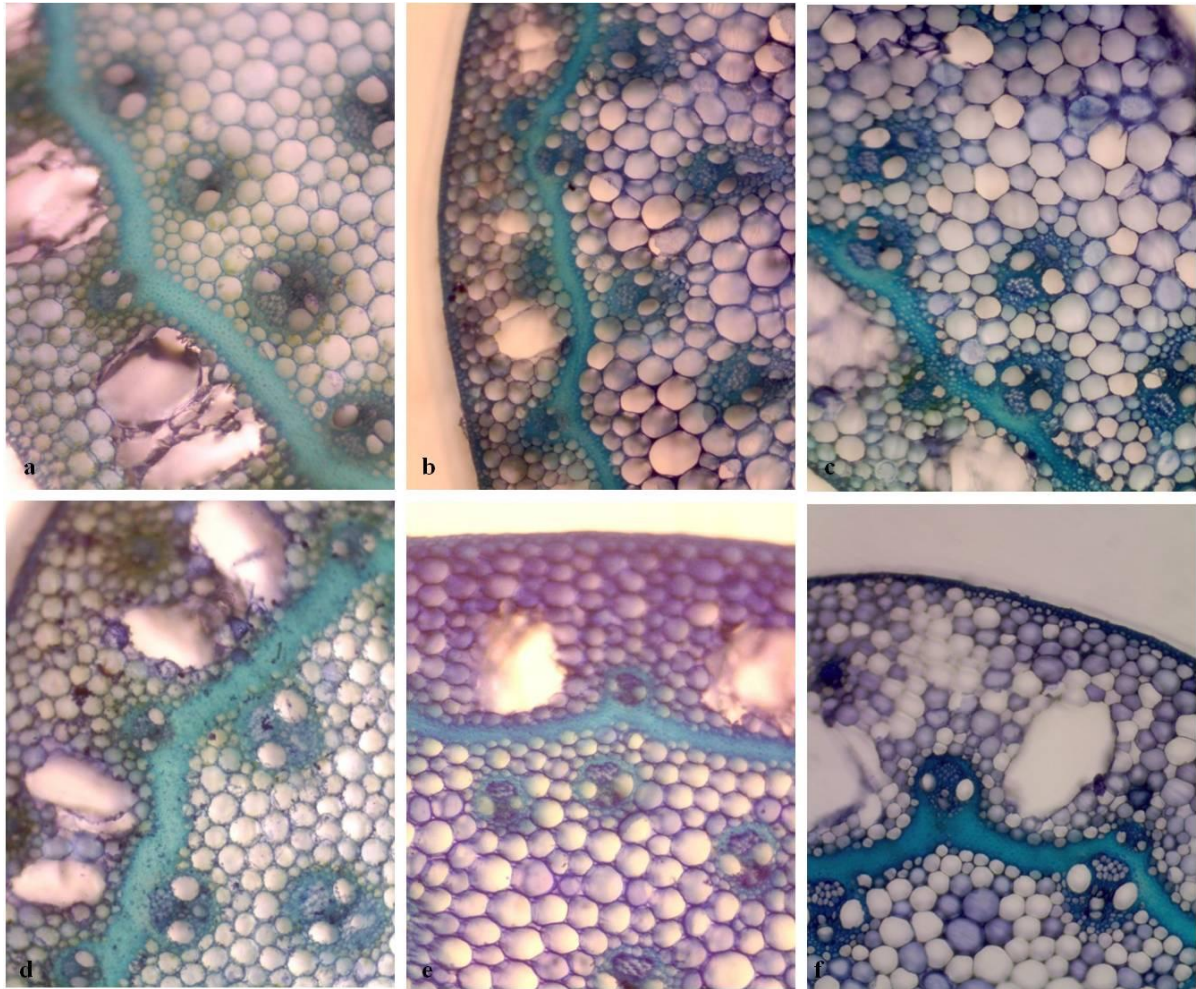


Plate 13. Cellular details revealed through light microscopy using TB O stain
a. *Adari*; **b.** *Aamo 10*; **c.** *CO3*; **d.** *TNPsc 176*; **e.** *TNPsc 183*; **f.** *Sel 21*. Thicker band of sclerenchyma arc was observed in genotypes, *Adari*, *TNPsc 176* and *Sel 21*, which contributes to culm strength (10x magnification)

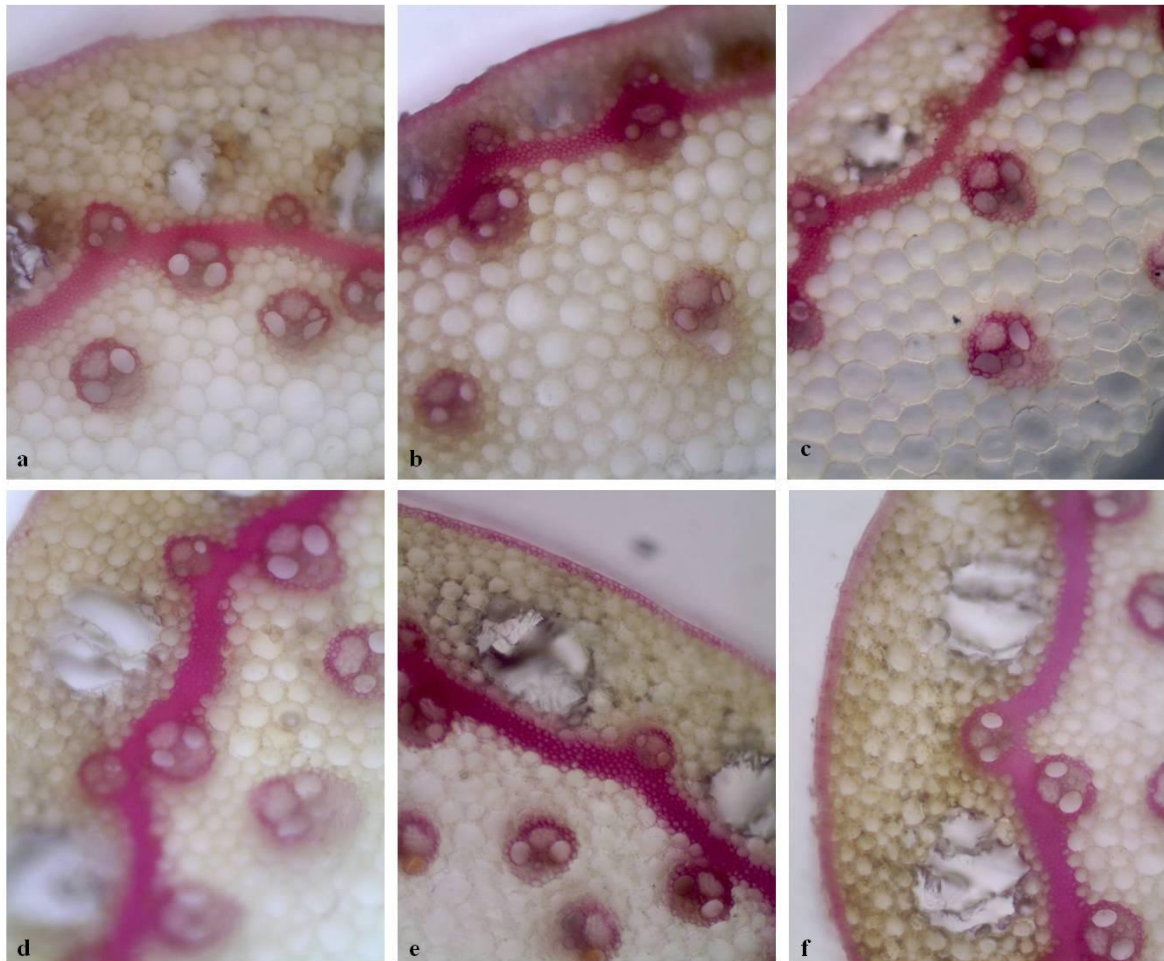


Plate 14. Lignified areas revealed through light microscopy using Phloroglucinol-HCl stain

a. *Adari*; **b.** *Aamo 10*; **c.** *CO3*; **d.** *TNPsc 176*; **e.** *TNPsc 183*; **f.** *Sel 21*. Stain absorption was more intense in *Sel 21* and *TNPsc 176* (10x magnification)

4.5.2 Mean performance of genotypes with respect to anatomical features

Ultrastructure of the culm cross section were studied under SEM and measurements were recorded on number of vascular bundles, culm thickness, sclerenchyma arc thickness, air space thickness, sclerenchyma cell wall thickness, parenchyma cell wall thickness and parenchyma cell diameter in four genotypes *viz.*, *Adari*, the strong culm check, Aamo 10, the weak line, Sel 21 and TNPsc 183, the two test entries (Plate 15).

Only four traits, *viz.*, number of vascular bundles, culm thickness, sclerenchyma arc thickness and air space thickness showed significant variation among treatments (Table 26).

Mean performance of the six genotypes for seven anatomical measurements are furnished in table 27. Four traits *viz.*, number of vascular bundles, culm thickness, sclerenchyma arc thickness and air space thickness, which had significant variation between treatments are explained hereunder.

4.5.2.1 Number of vascular bundles

Among the four genotypes studied, *Adari* had the highest number of vascular bundles (55.80), followed by Sel 21 (48.60), TNPsc 183 (44.40) and Aamo 10 (36.60). Aamo 10 had significantly lower number of vascular bundles than the rest of the genotypes and *Adari* had significantly higher number of vascular bundles than the rest (Table 27; Fig. 6a).

4.5.2.2 Culm thickness

The entries studied were significantly different from each other with respect to culm thickness. The highest value for culm thickness was recorded in Sel 21 (1283.40 μm), followed by *Adari* (958.30 μm), TNPsc 183 (782.82 μm) and Aamo 10 (560.30 μm) (Table 27; Fig. 6b; Plate 16).

4.5.2.3 Sclerenchyma arc thickness

The genotypes TNPsc 183 and Aamo 10 were similar in their expression (23.74 μm and 25.73 μm) with respect this trait. Likewise the genotypes Sel 21 and *Adari* were similar in their expression (39.02 and 43.37 μm) (Table 27; Fig. 6c; Plate 17).

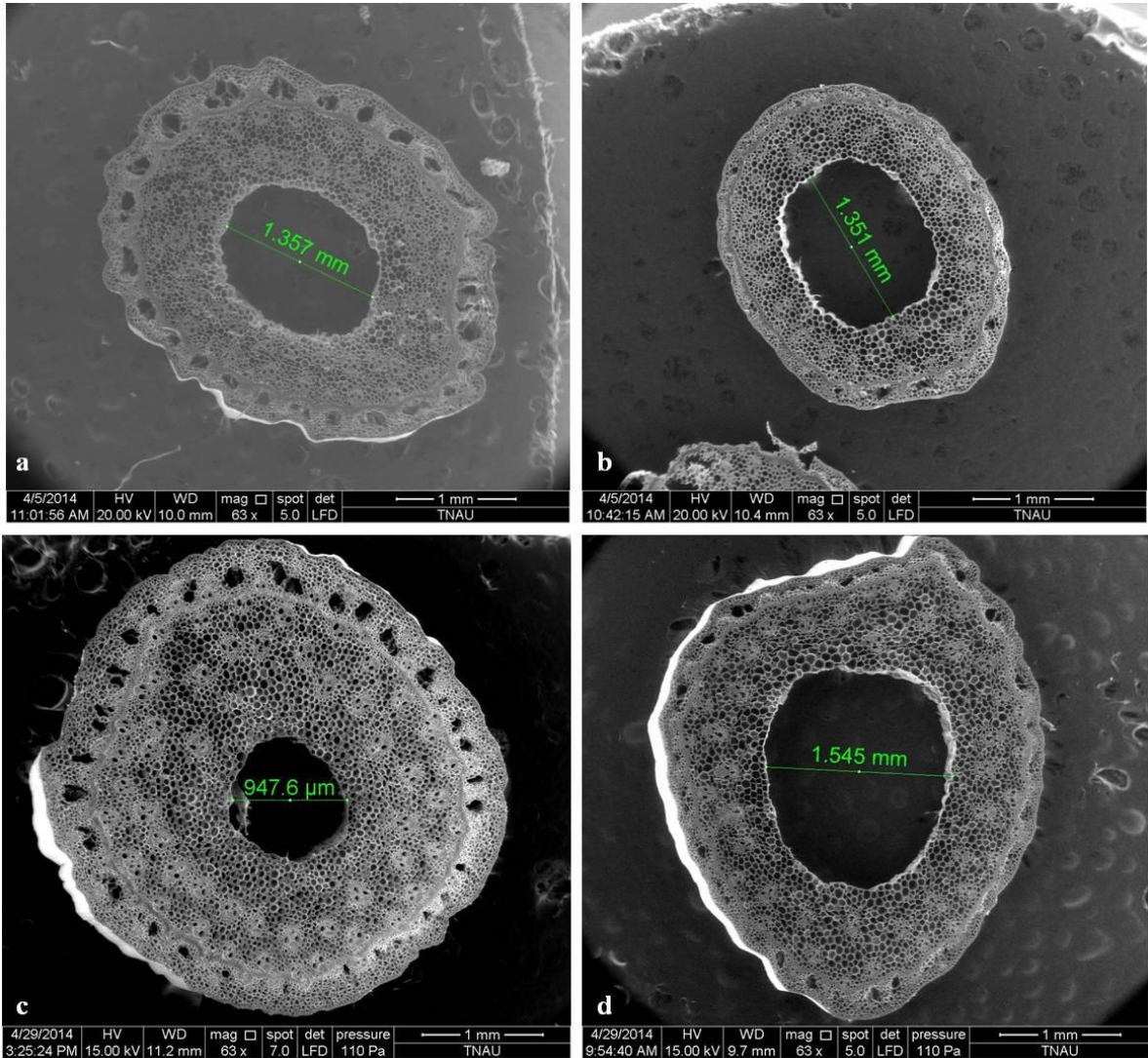


Plate 15. Cross section observed through SEM in select kodo millet genotypes
a Adari; **b**. Aamo 10; **c**. Sel 21; **d**. TNPsc 183 (63x magnification). Culm thickness was more in Sel 21

Table 26. ANOVA for traits relating to cell dimensions in four germplasm accessions of kodo millet

Sl. No	Source	df	Mean sum of squares						
			Number of vascular bundles	Culm thickness	Sclerenchyma arc thickness	Air space thickness	Cell wall thickness in sclerenchyma around vascular bundles	Cell wall thickness in parenchyma	Parenchyma cell diameter
1	Replications	4	28.075	331.124	18.777	416.388	0.227	0.098	75.555
2	Treatments	3	322.05**	465773.474**	470.622**	36575.033**	0.713	0.127	98.494
3	Error	12	26.508	2285.414	15.628	318.353	0.206	0.082	83.234

*significant at 5% level; **significant at 1% level

Table 27. Mean performance for traits relating to cell dimensions in four germplasm accessions of kodo millet

Sl. No	Genotypes	Number of vascular bundles (count)	Culm thickness (µm)	Sclerenchyma arc thickness (µm)	Air space thickness (µm)	Cell wall thickness in sclerenchyma around vascular bundles (µm)	Cell wall thickness in parenchyma (µm)	Parenchyma cell diameter (µm)
1	TNPsc 183	44.40	782.82	23.74	138.40	2.15	2.21	55.92
2	Sel 21	48.60	1283.40	39.02	291.68	2.69	2.33	47.63
3	<i>Adari</i>	55.80	958.30	43.37	245.20	2.07	2.39	56.76
4	Aamo 10	36.60	560.30	25.73	111.95	1.79	2.59	50.14
	Mean	46.35	896.21	32.97	196.81	2.17	2.38	52.61
	SE	3.26	30.24	2.50	11.28	0.29	0.18	5.77
	CD (0.05%)	7.10	65.88	5.45	24.59	NA	NA	NA

NA: Not applicable, since treatments are insignificant

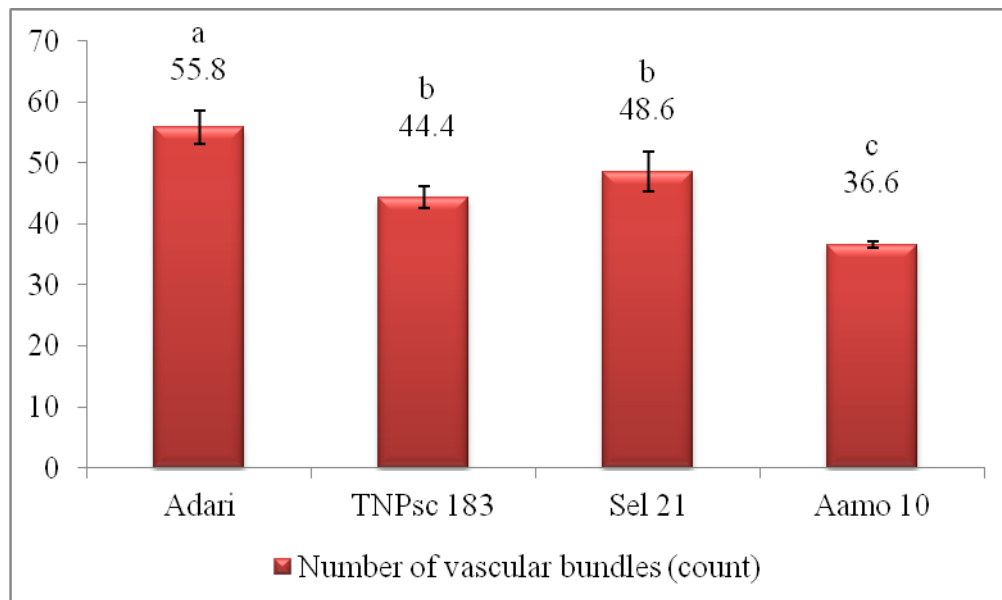


Figure 6a. Number of vascular bundles in select kodo millet genotypes (Error bars indicate standard error)
 TNPsc 183 and Sel 21 were identical with respect to the number of vascular bundles though vary in culm thickness

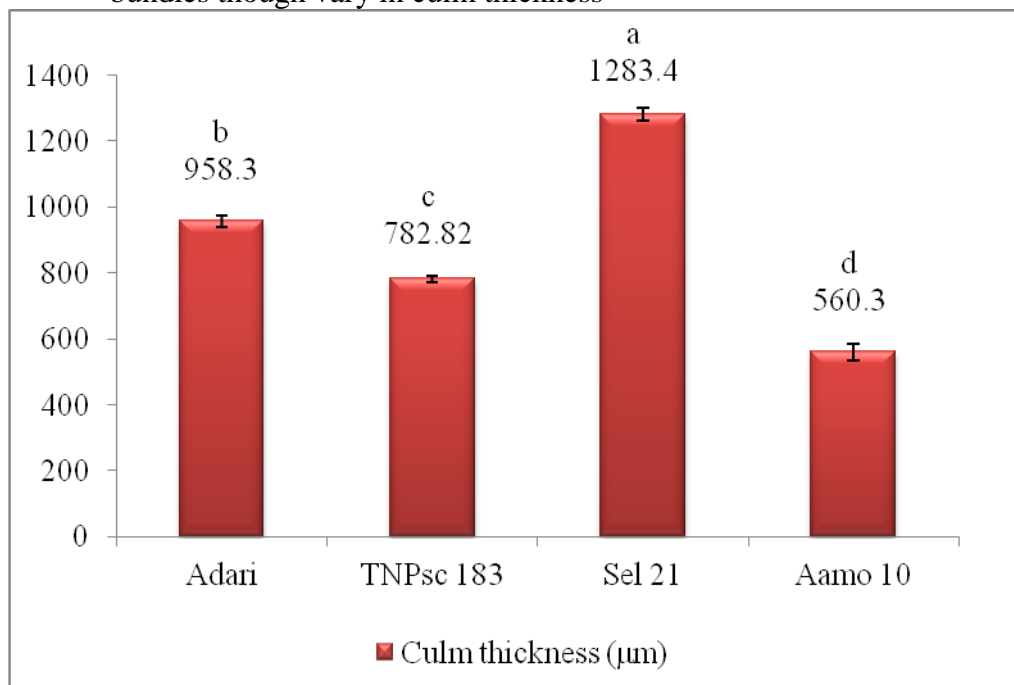


Figure 6b. Culm thickness in select kodo millet genotypes (Error bars indicate standard error)
 All the genotypes differ significantly with each other with respect to culm thickness

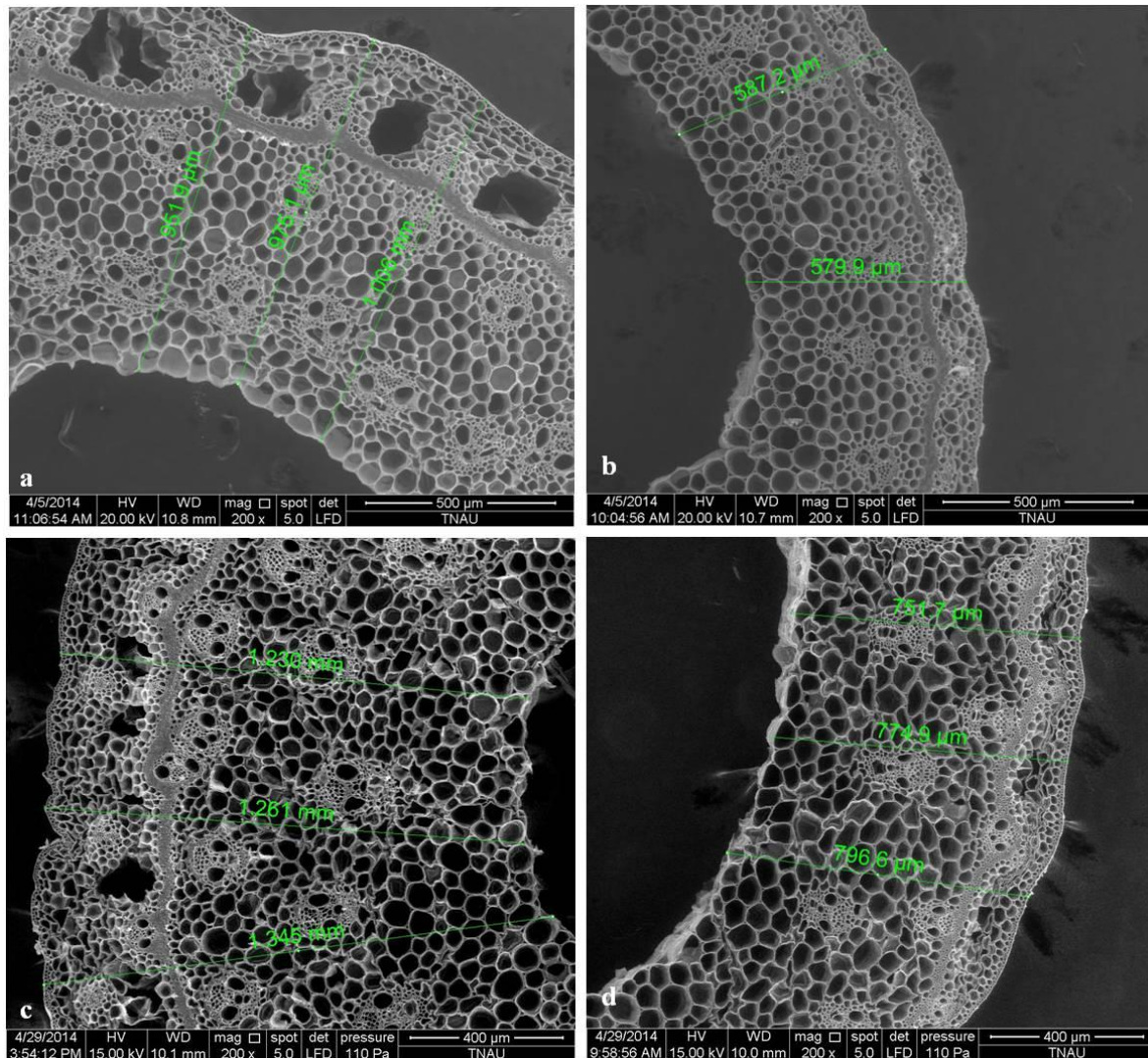


Plate 16. Variations in culm thickness in cross section of culms observed through SEM in select kodo millet genotypes

a. *Adari*; **b.** *Aamo 10*; **c.** *Sel 21*; **d.** *TNPsc 183* (200x magnification). *Sel 21* exhibiting the highest culm thickness. *Aamo 10* with least thickness

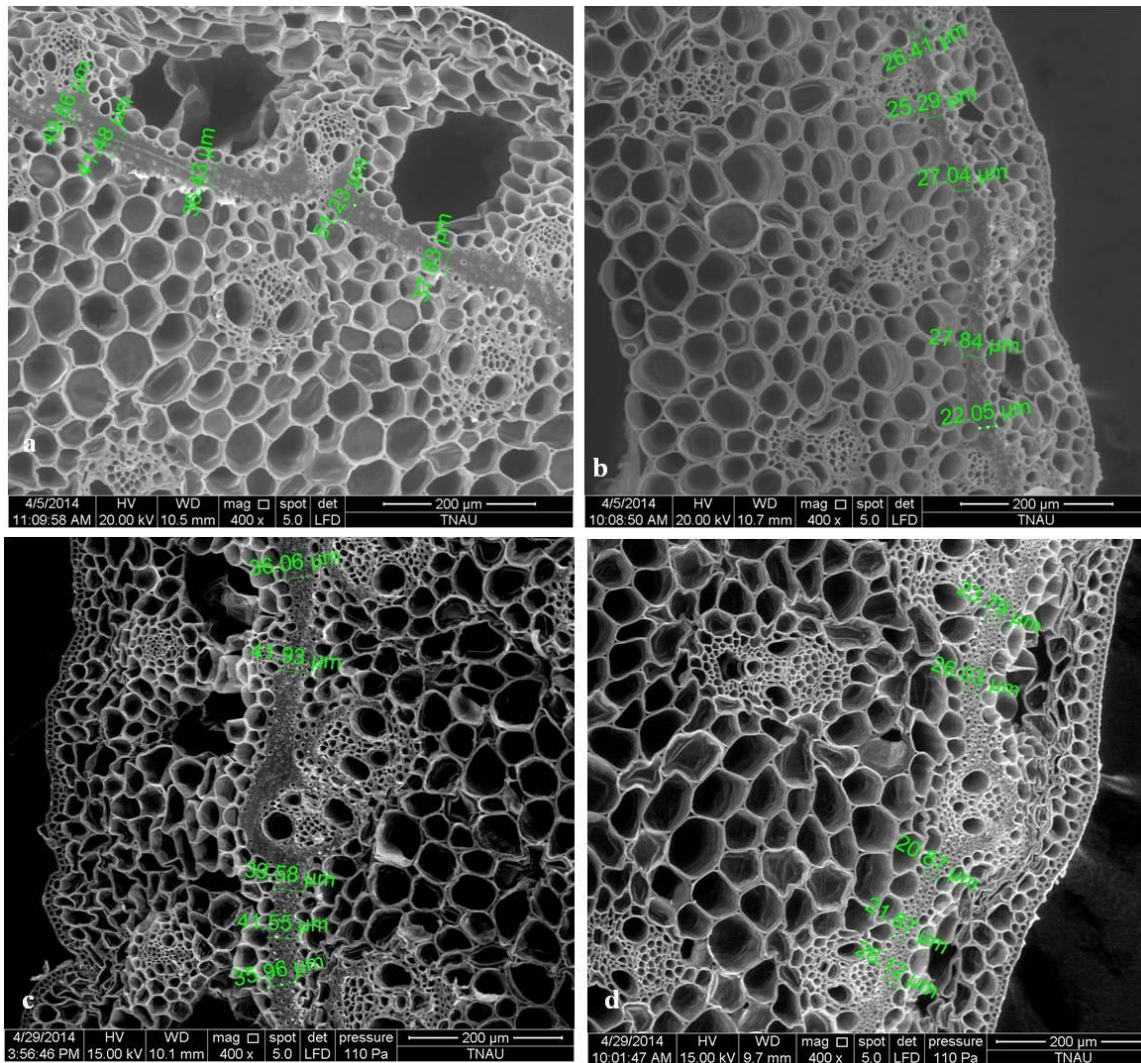


Plate 17. Variations in sclerenchyma arc thickness in cross section of culms observed through SEM in select kodo millet genotypes
a. *Adari*; **b.** *Aamo 10*; **c.** *Sel 21*; **d.** *TNPsc 183* (400x magnification). *Adari* and *Sel 21* were significantly thicker than the *Aamo 10* and *TNPsc 183*

4.5.2.4 Air space thickness

All the genotypes were significantly different from each other with respect to this trait. Maximum air space thickness was recorded by Sel 21 (291.68 μm), followed by *Adari* (245.20 μm), TNPsc 183 (138.40 μm) and Aamo 10 (111.95 μm) (Table 27, Fig 6d).

4.5.3 Mean performance of genotypes with respect to elemental composition in different tissues

The ANOVA for Silicon and Potassium content in different tissues showed difference between genotypes studied as well as between tissues (Table 28).

4.5.3.1 Silicon (Si)

The lines TNPsc 183 (0.270%), Sel 21 (0.305%), *Adari* (0.227%) were on par with respect to this trait. But the weak line Aamo 10 (0.069%) had significantly lower Si content. Between different tissues, there was no difference in Si content (Table 29; Fig. 7a).

The silicon content (%) in sclerenchyma in the arc, sclerenchyma around vascular bundles and parenchyma were on par with each other (Fig. 7c)

4.5.3.2 Potassium (K)

The test entries TNPsc 183 (1.063%) and Sel 21 (0.807%) were on par with each other and significantly lower than *Adari* and Aamo 10 with respect to relative signals for the element Potassium. *Adari* (2.092%) and Aamo 10 (2.604%) had higher values for this trait (Table 29; Fig. 7b).

The potassium content (%) in sclerenchyma in the arc, sclerenchyma around vascular bundles and parenchyma were distingly different from each other with a maximum (3.02%) recorded in parenchyma cell walls and minimum (1.25%) in cells walls in sclerenchyma arc (Fig. 7d).

4.6 Quantitative reverse transcription-polymerase chain reaction (qRT-PCR) of gene expression in select kodo millet genotypes

The rice, *FLEXIBLE CULM1* gene is associated with lignin biosynthesis. The relative gene expression of *FCI* (Fig. 8) was maximum in *Adari* (2.01) followed by Sel 21 (1.53), TNPsc 183 (1.44) and Aamo 10 (1.27).

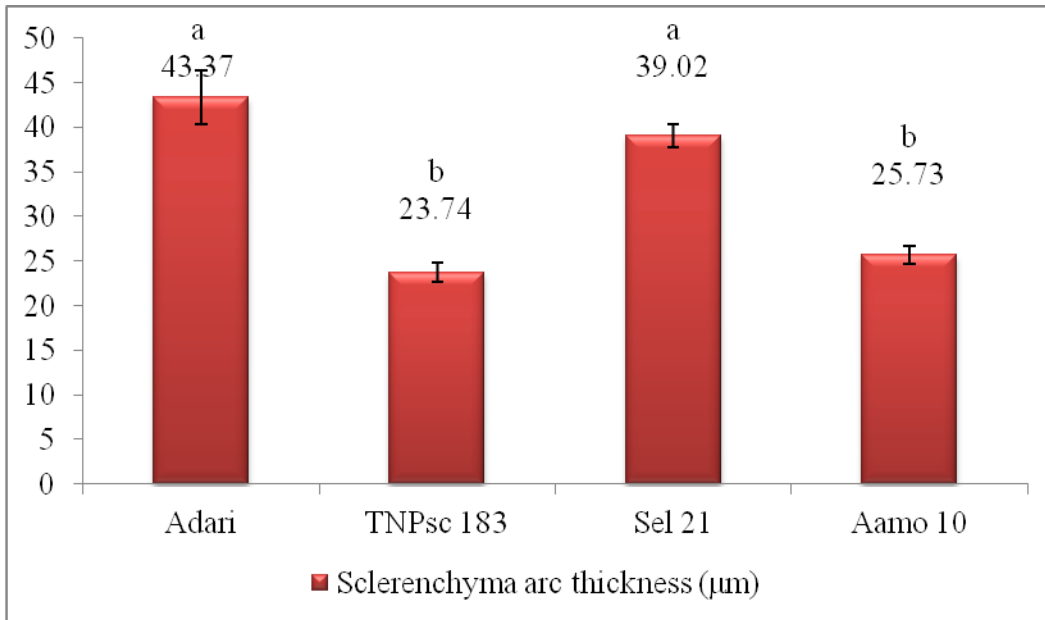


Figure 6c. Sclerenchyma arc thickness in select kodo millet genotypes (Error bars indicate standard error)
 Arc thickness was higher in *Adari* and *Sel 21* when compared with *TNPsc 183* and *Aamo 10*

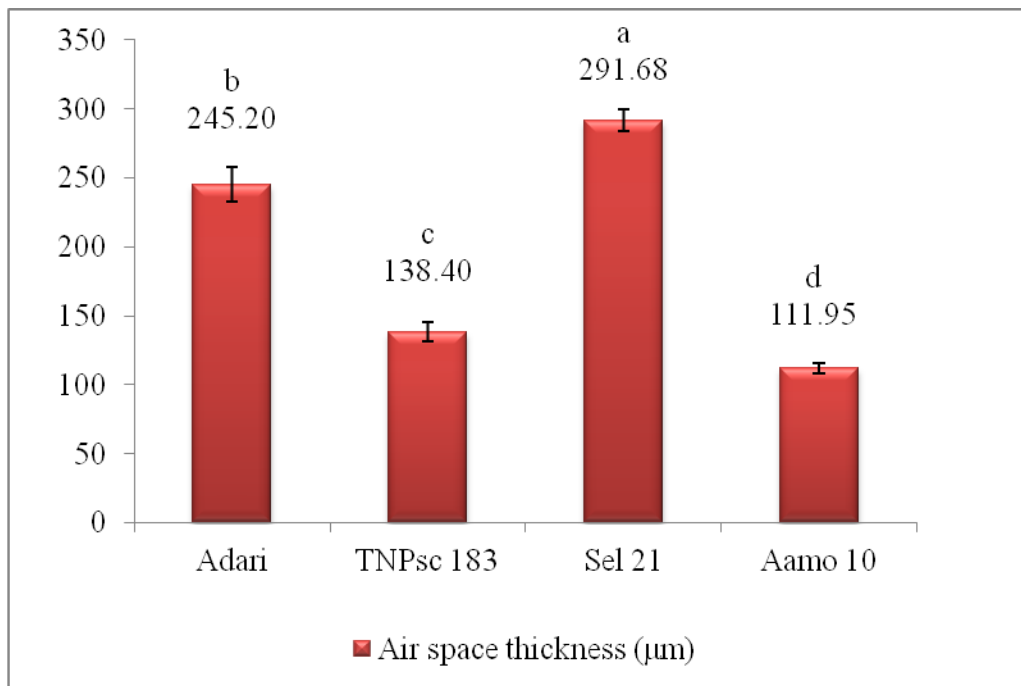


Figure 6d. Air space thickness in select kodo millet genotypes (Error bars indicate standard error)
 All the genotypes were different with respect to air space thickness

Table 28. ANOVA for elemental composition (%) using SEM-EDX in four germplasm accessions of kodo millet

Source of variation	Degrees of freedom	Mean sum of squares	
		Si	K
Replications	4	0.005	0.633
Treatments	11	0.087**	4.726**
Factor A (Genotypes)	3	0.163**	10.803**
Factor B (Tissues)	2	0.061*	2.798*
A X B	6	0.058**	2.33*
Error	44	0.017	0.803

*significant at 5% level; **significant at 1% level

Table 29. Mean performance for elemental composition (%) using SEM-EDX in four germplasm accessions of kodo millet

Sl. No.	Tissues	Relative Signal %					
		Element	TNPsc 183	TNPsc 262	<i>Adari</i>	Aamo 10	Mean±SE
1	Sclerenchyma arc	Si	0.438	0.280	0.158	0.052	0.232±0.03
		K	1.320	0.622	1.082	1.968	1.248±0.51
2	Parenchyma	Si	0.320	0.336	0.320	0.082	0.265±0.03
		K	1.350	0.850	3.914	5.982	3.024±0.51
3	Sclerenchyma around vascular bundles	Si	0.052	0.298	0.204	0.072	0.157±0.03
		K	0.520	0.948	3.368	3.134	1.993±0.51
Mean of Si (SE=0.05)			0.270	0.305	0.227	0.069	
Mean of K (SE=0.69)			1.063	0.807	2.092	2.604	

CD (0.05%) between genotypes (Si) = 0.095; CD (0.05%) between tissues (Si) = 0.082

CD (0.05%) between genotypes (K) = 0.659; CD (0.05%) between tissues (K) = 0.571

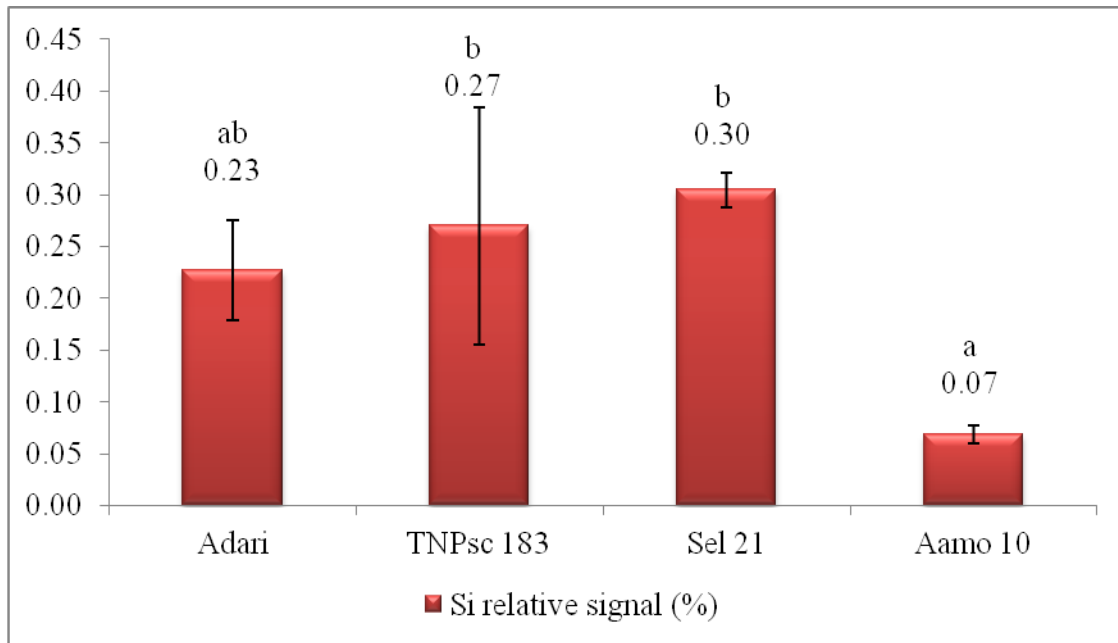


Figure 7a. Silicon relative signal in (%) in select kodo millet genotypes (Error bars indicate standard error)
Silicon content was higher in TNPsc 183 and Sel 21

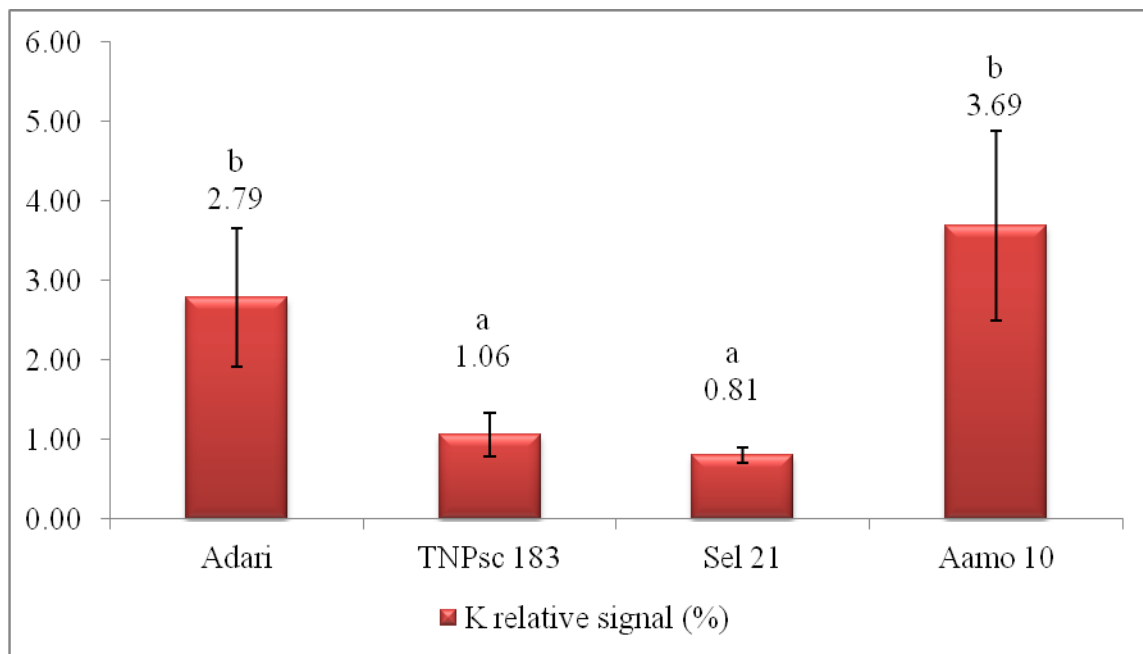


Figure 7b. Potassium relative signal in (%) in select kodo millet genotypes (Error bars indicate standard error)

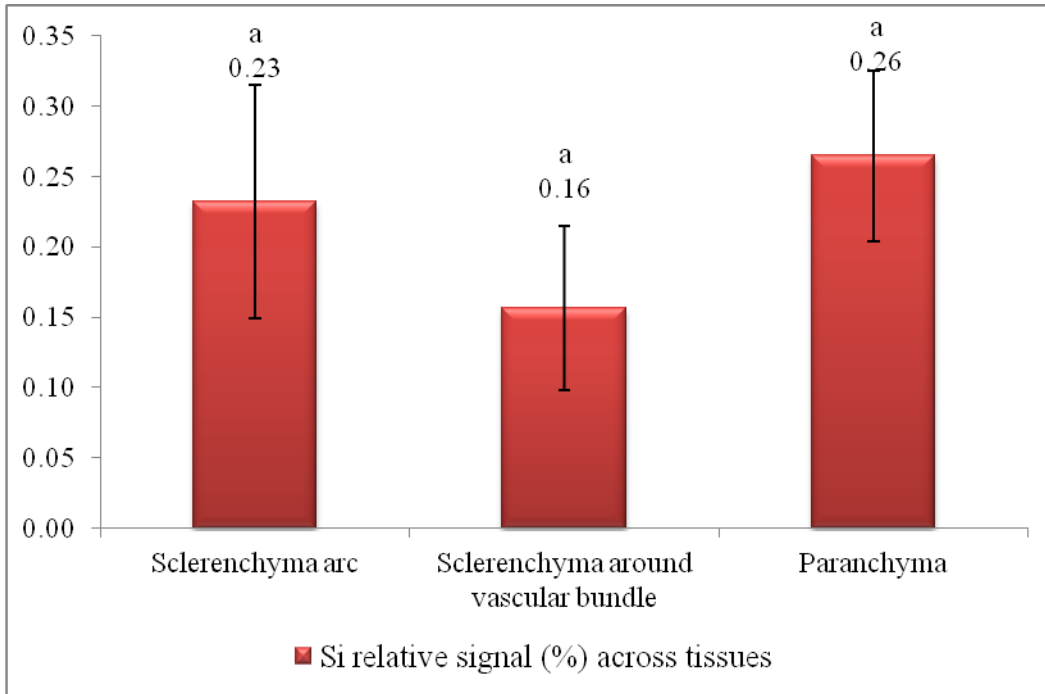


Figure 7c. Silicon relative signal (%) in different tissues in kodo millet genotypes
 (Error bars indicate standard error)
 Silicon content was identical across tissues

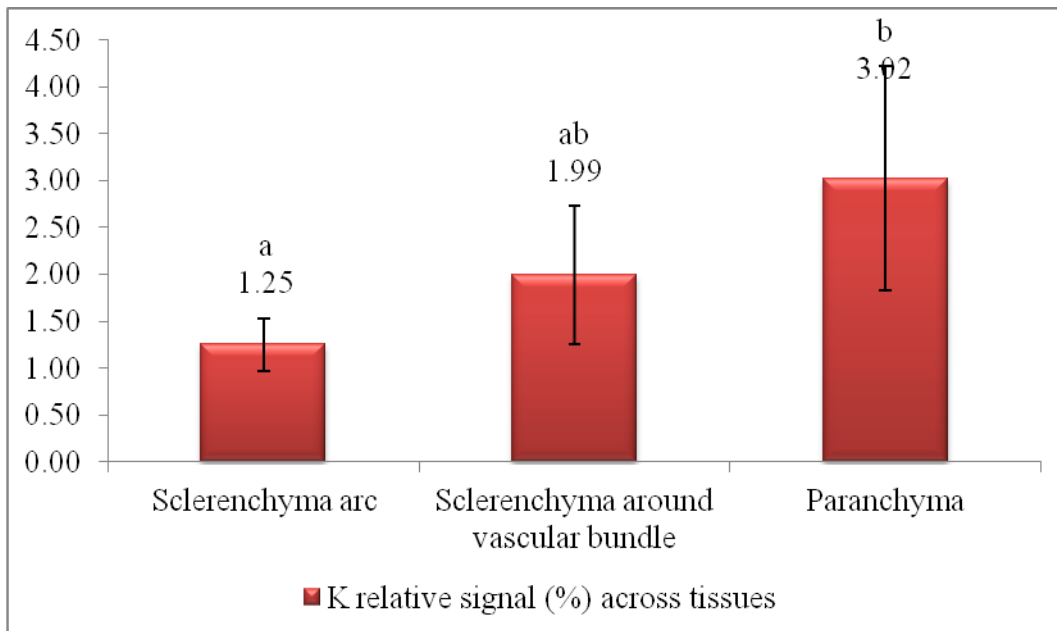


Figure 7d. Potassium relative signal (%) in different tissues in kodo millet genotypes
 (Error bars indicate standard error)
 Potassium content was more in parenchyma than sclerenchyma

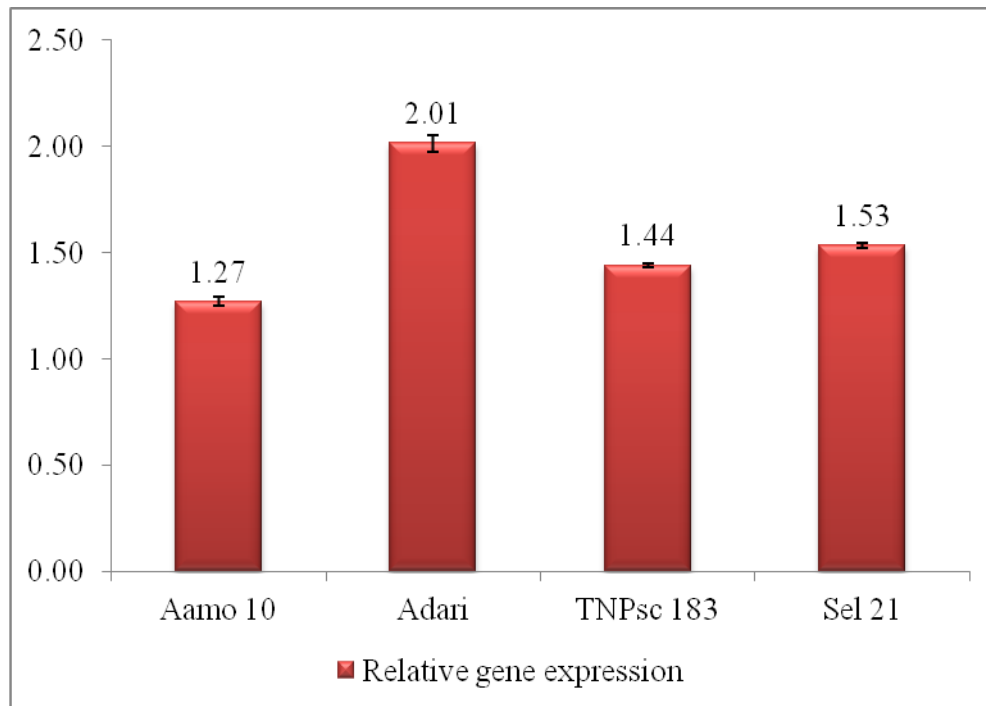


Figure 8. *FLEXIBLE CULMI* expression obtained from qRT-PCR in select kodo millet genotypes (Error bars indicate standard error)

FLEXIBLE CULMI, gene associated with lignin biosynthetic pathway registered high level of expression in *Adari*, the strong culm line and low level of expression in Aamo 10, the weak culm line. The test entries TNPsc 183 and Sel 21 registered optimal expression levels.

Discussion

CHAPTER V

DISCUSSION

Today the food grain production is largely reliant on two crops, rice and wheat. In this context, the potentials of indigenous crop plants including millets are being gradually revisited for its genetic constitution which is becoming increasingly relevant in the changing agricultural scenario. The present challenges to the agriculture production are largely due to climatic and edaphic adversaries. And, any native crop species that holds comparative advantages under these challenging environments need to be focused on (Padulosi *et al.*, 2009). One such group that appears to be prospective in complementing the shortfall in agriculture production comprises the small millets, as they still contribute to the regional food security of the dry and marginal lands, where major cereal crops such as rice and wheat fails to yield.

The small millet group is represented by six species, namely, finger millet, kodo millet, foxtail millet, little millet, proso millet and barnyard millet. Among them, kodo millet (*Paspalum scrobiculatum* L) is indigenous to India (de Wet *et al.*, 1983). Kodo millet thrives well in arid climate with minimal water requirement compared to other cereal members as well as least affected by pest and diseases. Besides being a staple for poor and marginal agricultural regions, kodo millet is gaining a new role as a health food for its superior nutritive value, the presence of more dietary fibre, anti-oxidant nature (Chandrasekara and Shahidi, 2010) and anti-diabetic principles (Hegde *et al.*, 2005). More said, being a C4 plant, it is gaining attention due to its suitability to changing agro-climatic conditions as most of the arable land (69%) in India is arid and dry. Hence, any technological intervention in this crop could have a major boost in the cultivation area, production and productivity.

Kodo millet is predominantly grown as a pure crop and yields high net returns as compared to other dry land crops owing to its high unit area productivity and market price of the produce in addition to its fodder value. In Tamil Nadu, due to scarcity of labour, farmers are into the practice of using paddy combine harvesters to reap the crop. However, the spreading plant habit and poor culm strength of land races including high yielding varieties leads to heavy lodging at the time of maturity and result in grain loss

when harvested mechanically. Given the context, genotypes that are non-lodging with sufficient culm strength may be suitable for mechanical harvesting. Hence, identifying kodo millet genotypes which can stand erect, non-lodging with sufficient culm strength and uniform in maturity has been the endeavor of this research.

5.1 Phenotypic variations for culm strength in the kodo millet germplasm

A total of 297 kodo millet germplasm accessions, including land races collected from different parts of Tamil Nadu and from other parts of the country, maintained at Department of Millets, Tamil Nadu Agricultural University, were used in the present investigation. For initial screening and evaluation, all the genotypes were raised in Augmented Block design during *kharif* 2012. The released variety CO3 and *Adari*, a land race, were taken as a check for high yield and culm strength respectively.

All the germplasm accessions were phenotyped based on 13 morpho-metric traits, *viz.*, days to 50 per cent flowering (days), plant height (cm), number of productive tillers (count), inflorescence length (cm), raceme number (count), raceme length (cm), culm diameter (mm), pulling force (PF), internode length (cm); recovery angle after bending (degrees), days to maturity (days), grain yield per plant (g), test weight (g) and four scorable traits *viz.*, plant type, ear exertion at dough stage, spikelet arrangement on rachis and degree of lodging. The traits, plant height (Kelbert *et al.*, 2004), culm diameter (Kashiwagi *et al.*, 2008), internode length (Aliaga *et al.*, 1986), pulling force and recovery ability after bending (Torro *et al.*, 2011) were chosen for the study, based on the published literatures in rice and wheat.

Analysis of variance exhibited variation between genotypes for all the morphometric traits studied which is a prerequisite for any breeding program based on selection. Subsequent analysis included correlation and path analyses to identify traits related to culm strength which is quantifiably represented by pulling force. Since, in ABD, genotypes were not replicated except controls, phenotypic correlation coefficients between pairs of traits were calculated. The traits such as inflorescence length, raceme number, raceme length, and test weight were excluded from correlation analysis to avoid ambiguity as this correlation study is to understand the traits related to culm strength. In this study, the sample size inclusive of checks is 299. Hence, the correlation can be

relied upon (Schonbrodt and Perugini, 2013). A plant to be suitable to mechanized harvesting, it should have sufficient culm strength, with good yield. Hence, the results of the intercorrelation among traits are discussed hereunder with special focus on these three traits.

5.1.1 Association studies

5.1.1.1 Traits related to culm strength

The pulling force, which quantifiably represents culm strength exhibited high positive correlation with culm diameter, plant height, number of productive tillers per plant, grain yield per plant and days to 50 per cent flowering. But, pulling force had insignificant correlation with internode length, recovery ability after bending and degree of lodging (Table 7).

The correlation of pulling force with other traits indicated that the lines with high culm strength were taller with thicker culm, more productive tillers and yield, but were late in flowering. This result corroborated with the findings of Kashiwagi *et al.* (2008), who reported that the stem diameter was positively correlated with pushing resistance in rice. The insignificant correlation of pulling force with recovery angle after bending is in accordance with the earlier findings of Torro *et al.* (2011) in rice, who reported that culm strength and elasticity were two independent traits. The correlation between degree of lodging and recovery angle is negative, *i.e.*, the plants which recovered better seldom lodged. Hence, to assess lodging resistance alone, recovery angle would be the ideal trait for screening as reported by Torro *et al.* (2011). But to identify non-lodging genotypes with good culm strength to suit mechanical harvesting, simultaneous selection for high pulling force and recovery angle have to be performed as evinced from this study.

5.1.1.2 Traits related to lodging

Plant elasticity represented by recovery angle after bending had negative correlation with internode length and degree of lodging. The correlation between internode length and degree of lodging was positive and highly significant. Recovery angle after bending had high negative correlation with degree of lodging, plant height, internode length and grain yield per plant. This reveals that lines which are tall with more grain weight and longer basal inter-nodal length will have poor recovery after bending. Degree of lodging was

negatively associated with days to flowering and days to maturity. This suggests that short duration lines are more prone to lodging which may be due to shorter time window available for strengthening and lignification of the culm which is needed for culm strength (Li *et al.*, 2000). Moreover, correlation between degree of lodging and plant height was positive indicating that the taller plants are more susceptible to lodging. Similar reports of association of lodging with longer stems are available in wheat (Crook and Ennos, 1994) and, rice (Chang and Vergara, 1972). Crook and Ennos (1994) reported that lodging resistance was associated with shorter stems because taller stem increased the force of gravity on to the plant. Moreover, tall plants with sufficient culm strength normally stand erect, but any adverse environmental conditions like high plant density, heavy rain or wind lodges the plant and its recovery to original position demands more force. Hence, the tall plants fail to recover after lodging. The lack of correlation between degree of lodging and culm diameter in the study reinstates the above statement that both weaker and strong culms may tend to lodge, the former for its inherent weakness in culm and inability to stand erect and the latter because of its inability to recover upon lodging. So, an ideal plant type has to be medium tall with sufficient culm strength which can stand while being cut by the blades of the mechanical harvester and recover better upon lodging due to adverse environmental conditions (Table 7).

Internode length was positively correlated with degree of lodging which means shorter the internode length, lesser will be the degree of lodging. This corroborated with the findings of Jezowski *et al.* (1987) that the lower internodes of lodging resistant F₁ hybrids from crosses between lodging resistant and lodging susceptible barley cultivars were shorter and thicker at maturity than the lower internodes of their lodging susceptible siblings. In the present study, correlation of internode length with recovery angle after bending was negative and that with culm diameter was not significant. This is in conformity with the earlier work of Aliaga *et al.* (1986), who reported that culm elasticity correlated with internode length, but not with culm thickness in rice. Internode length was found to be negatively correlated with days to 50 per cent flowering and days to maturity. This suggests that the short duration lines are with longer basal internodes thus giving them the elasticity to bend (Table 7).

5.1.1.3 Traits related to grain yield

Grain yield per plant had positive correlation with plant height, number of productive tillers per plant, culm diameter, internode length, degree of lodging and pulling force, and negative association with recovery ability after bending (Table 7). This is the most important trait which should also be given due consideration, because increased return is the ultimate goal of any study. Though the grain yield increases with plant height, it becomes static beyond a height of 60 cm (Fig. 9). Likewise if the pulling force and culm diameter increases beyond 6 N (Fig. 10) and 4 mm (Fig. 11), respectively, there is no corresponding increase in the grain yield. This could be because of diversion of source to strengthen the culm at the cost of yield. Moreover, the grain yield decreases with days to maturity. Hence an ideal cultivar should be medium tall and medium duration with optimal pulling force and culm diameter.

5.1.1.4 Path analysis reveals culm diameter influences culm strength

In this study, path analysis was carried out to find out true relationships between the associated traits with pulling force (Table 8). Path analysis revealed that culm diameter (high) had the highest positive direct effect on pulling force followed by plant height (moderate) and recovery angle after bending (moderate). Hence, the culm diameter, which registered high positive direct effect on pulling force, is the best selection index for assessing culm strength. Following this, plant height and recovery angle after bending also have the direct positive association with pulling force. So, to identify genotypes with good culm strength in kodo millet, culm diameter, plant height and recovery angle after bending could be the candidate traits based on which selection could be imposed.

Though the correlation and path analyses on these 299 genotypes aimed to identify traits related to mechanical strength of culm, features related to high yield and lodging resistance were also included to arrive at a plant type suitable for machine harvest without compromising on yield. This could be the reason for high residual effect (Table 8). Moreover, apart from the morphological traits studied, polysaccharides (Cellulose, Hemicellulose), lignin (Li *et al.*, 2003) and other elemental (Silicon, Potassium) composition (Datta and Mikkelsen, 1985; Idris *et al.*, 1975; Ma and Yamaji, 2006) of the culm may have a

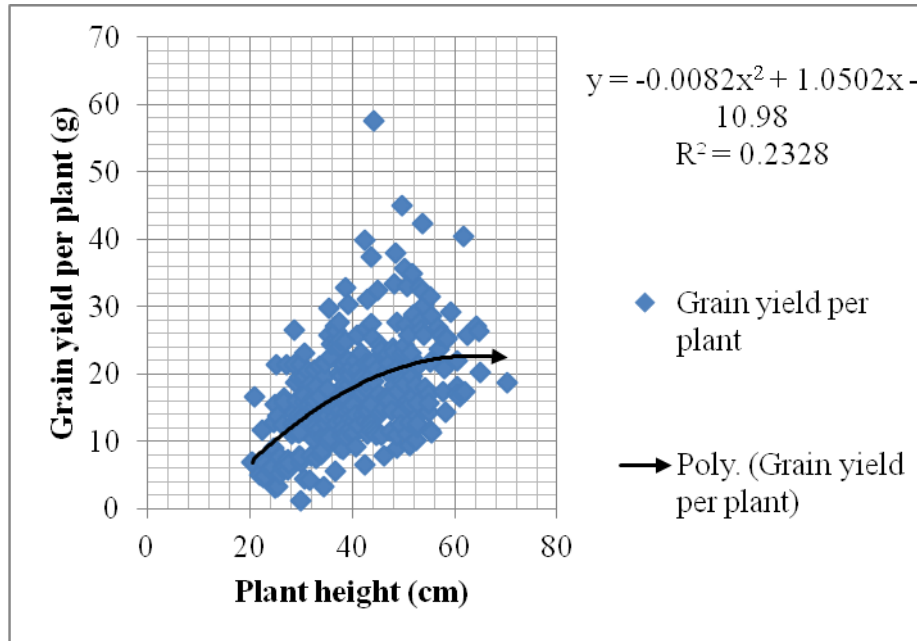


Figure 9. The polynomial regression of plant height with grain yield per plant. The grain yield reaches a static beyond 60cm height.

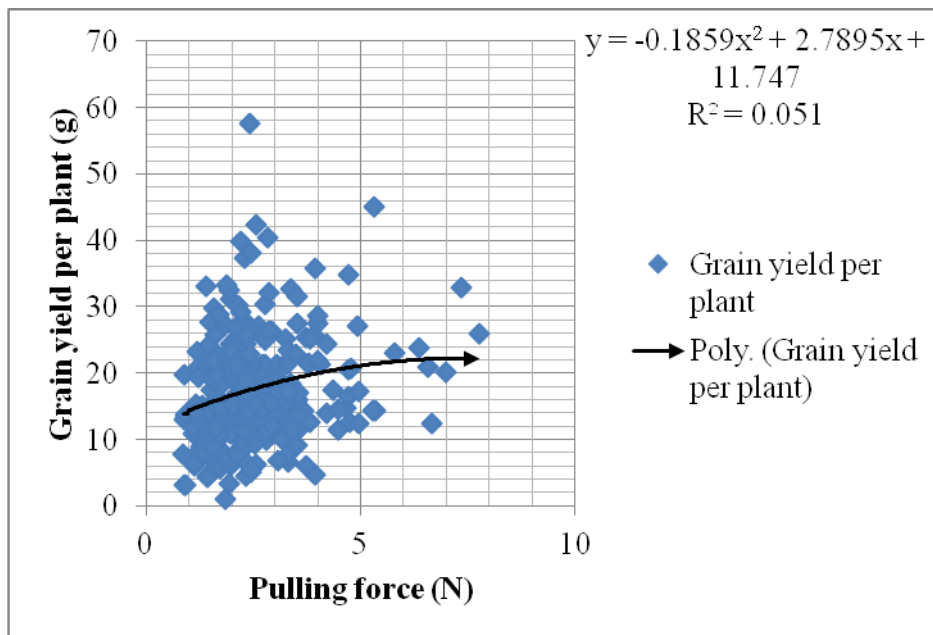


Figure 10. The polynomial regression of pulling force on grain yield per plant. Beyond a pulling force of 6 Newton there is no corresponding increase in grain yield.

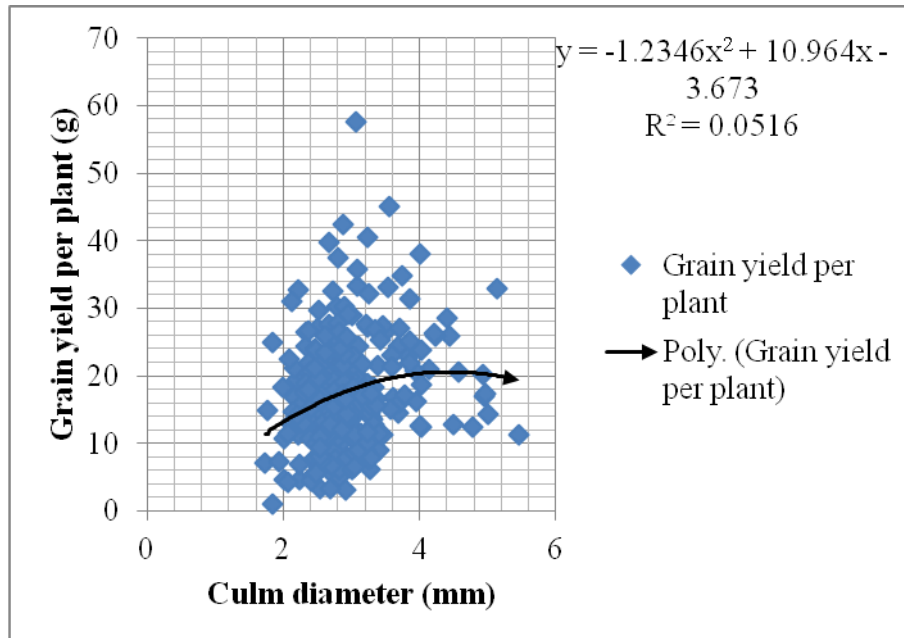


Figure 11. The polynomial regression of culm diameter with grain yield per plant.
Culm diameter more than 4 mm has no positive effect on grain yield per plant.

direct bearing on culm strength. Non-inclusion of these traits may also be the one of the important reason for high residual effect.

Thus the genotypes which had higher culm diameter, pulling force, recovery angle after bending were alone forwarded for the next season. Strict selection could not be exercised, as the mean for different traits were obtained from non-replicated and single season trial. But, genotypes which were less than 40 cm height were not selected, as dwarf genotypes may not be suitable for machine feeding.

5.2 Variability and diversity studies and screening for culm strength

A total of 30 genotypes were selected based on these selection criteria and were then evaluated in the subsequent season (Summer, 2013) in replicated trial in RBD. Other than these 30 genotypes, CO3 and *Adari* local were included as checks. Apart from the above, nine lines which were lodged severely during the previous season were also included for better understanding of culm strength contributing traits.

All the 41 genotypes were phenotyped for 14 morphometric traits *viz.*, days to 50 per cent flowering (days), plant height (cm), number of productive tillers, inflorescence length (cm), raceme number (count), raceme length (cm), culm diameter (mm), pulling force (N), culm weight per unit length (mg/cm), internode length (cm), recovery angle after bending (degrees), days to maturity (days), grain yield per plant (g); test weight (g) and eight scorable traits *viz.*, plant type, ear exertion at dough stage, spikelet arrangement on rachis, degree of chlorosis, stay green nature at maturity, shattering of inflorescence, uniformity in maturity, degree of lodging. The metric trait culm weight per unit length and scorable traits, degree of chlorosis, stay green nature at maturity, shattering of inflorescence, uniformity in maturity, were included for strengthening the understanding of culm traits and suitability to machine harvest.

Analysis of variance recorded significant variation between genotypes studied for all the morphometric traits studied except for the trait number of productive tillers per plant. Hence further analyses such as variability and diversity studies were carried out on these 41 genotypes excluding the trait number of productive tillers per plant.

Variability studies aimed at understanding the variability present in the genotypes, the scope for improvement through selection and the genetics of the traits.

5.2.1 Variance components

The extent of variability in respect of range, phenotypic and genotypic coefficients of variation, is presented in table 12. In the present study, the traits, days to 50 per cent flowering, culm diameter, culm weight per unit length and days to maturity showed narrow differences between phenotypic and genotypic coefficients of variation indicating low effect of environment and greater role of genetic factors on the expression of these traits. Hence, culm diameter and culm weight per unit length are more reliable to be used as selection criteria. Similar findings of less influence of environment on stem weight per cm and stem diameter was reported by Zuber *et al.* (1999) in wheat. The rest of the traits (plant height, inflorescence length, raceme length, number of racemes, internode length, pulling force, recovery angle after bending, grain yield per plant and test weight) exhibited wide differences suggesting their vulnerability to environment. Hence, these traits could be used as selection criteria based on studies conducted on different environments only.

Since the phenotypic and genotypic variances are associated with units, the co-efficient of variation was worked out for valid comparisons between traits. High GCV and PCV estimates were recorded for pulling force, weight per unit length of the culm and grain yield per plant. Wide variability ensures more scope for improvement in these traits. Amawate (1965), Salpute (1966), Tiwari and Janoria (1967), Amawate and Dabral (1970), Choudhary and Singh (1970) and Rao (1991) also observed wide range of variability for morphological characters and grain yield in kodo millet among land races of Madhya Pradesh.

Low GCV and PCV estimates were recorded for inflorescence length, days to maturity and test weight among the genotypes studied. Similarly, Verma and Satpute (1969) and Ahluwalia *et al.* (1970) also reported that the magnitude of PCV was high for grain yield per plant whereas it was low for developmental traits like days to flowering and days to maturity in kodo millet. Subramanian *et al.* (2010) also reported high GCV and PCV for seed yield per plant and low GCV and PCV for test weight in kodo millet.

5.2.2 Heritability in broad sense and genetic advance

In plant breeding, the magnitude of heritability of the trait of interest determines the success of the breeding program as it has a close bearing on the response to selection. The genotypes under study showed low level of heritability for the trait internode length. The highest heritability was recorded for culm weight per unit length followed by grain yield per plant, days to 50 per cent flowering, culm diameter, pulling force and days to maturity indicating high influence of genetic components. The remaining traits, plant height, inflorescence length, raceme length, raceme number, internode length, recovery ability after bending and test weight, exhibited moderate level of heritability (Table 12). Similar reports of high level of heritable variation are available for days to flowering (Verma and Satpute, 1969 and Ahluwalia *et al.*, 1970) and low level of heritability for panicle length (Dhagat *et al.*, 1971) in kodo millet. Dhagat (1978), Parihar (1985), Rao and Tiwari (1988), Sharma (1988), and Kandaswamy *et al.* (1990) reported high heritability for grain yield and low heritability for plant height and days to flowering.

Heritability gives the information on the magnitude of inheritance of quantitative traits, while genetic advance will be helpful in formulating suitable selection procedures. Therefore, heritability estimates in conjunction with the predicted genetic advance will be more reliable (Johnson *et al.*, 1955). In the present study, the maximum genetic advance was recorded for weight per unit length of the culm, followed by grain yield per plant, pulling force and culm diameter (Table 12). These four traits also registered high heritability, which indicated that these traits were governed predominantly by additive genes and selection could be effective for improving them. Hence there is more scope for improvement in yield and culm attributes from the genotypes studied. Subramanian *et al.* (2010) also observed high heritability coupled with high genetic advance for the trait, seed yield per plant in kodo millet genotypes studied by them.

5.2.3 Euclidean distances and clustering among genotypes

The summary of distance coefficients between pairs of genotypes using Euclidean geometry is shown in figure 3. The mean phenotypic distance among genotypes was 4.89, suggesting low phenotypic diversity. This could be due to composition of the genotypes included for this analysis. Thirty genotypes were selections based on high culm strength

and nine were weak culm genotypes chosen for comparison. The lower genetic distance among the genotypes within each group might have lowered the overall average genetic distance. Another reason could be the presence of autogamy on account of highly cleistogamous flowers (Rangaswami-Ayyangar and Panduranga-Rao, 1934) and occasional apospory (Narayanaswami, 1954) reported in this crop. The lowest distance coefficient was exhibited by Aamo 13 and Aamo 126 pair, followed by TNPSc 217 and TNPSc 215 pair, and Aamo 5 and Aamo 126 pair. These pairs of genotypes showed similarity in almost all quantitative traits measured. Hence, Aamo 13, Aamo 126 and Aamo 5 might have originated from the same ancestor. Similarly, the accessions TNPSc 217 and TNPSc 215 could also be having close genetic lineage, which was reflected in the distance coefficient value. The distance coefficient values were relatively high for TNPSc 170, a weak culm line, with other strong culm lines, such as *Peruvaragu*, *Adari* and *Athipatti*. Clustering based on genetic distances yielded five clusters (Fig. 4).

Cluster means for the 13 morphological traits presented in table 15, revealed that most of the strong culm genotypes were grouped in clusters I and II as indicated by the higher mean values for these traits. The highest cluster mean for grain yield was recorded by Cluster I. Cluster compositions (Table 14), indicated that the land races, accommodated in the clusters II and III, are highly deviated from the lines grouped in clusters IV and V, which included weak culm lines with poor plant height, culm diameter, pulling force, weight per unit length and short duration. Shorter duration might be the reason for poor culm strength of genotypes in clusters IV and V. Hence, it could be interpreted that tall stature, thick culms and long duration were the most obvious traits of land races compared to other germplasm accessions. This might be because of the fact that land races resulted from continuous selection by farmers, with emphasis on visible traits, such as plant height, duration, inflorescence length, raceme length, test weight and thick culms.

The released variety CO3 was grouped in Cluster I, which accommodated half of the germplasm accessions (20) studied. This cluster is an important one as the lines in this cluster had culm strength comparable to that of land races (in clusters II and III) but better yielders with medium duration. Hence, any further improvement in yield could be achieved by including lines like, Aamo 210, Aamo 258 and Sel 21 in cluster I either through selection or recombination breeding. Likewise, any improvement in culm

strength could be achieved by using lines in Cluster II. Earliness and short stature were the typical features of clusters IV and V. Hence, the genotypes in these clusters are a good source for exploiting earliness and dwarf stature in this crop. In addition to these, the lines in cluster V were good yielders, too.

5.2.4 Principal component analysis (PCA)

Grouping of the traits through PCA was carried out to examine the percentage contribution of each trait to total genetic variation. The PCA revealed that the first four principal components accounted for 39, 16, 9, and 9 percent of total variability, respectively and cumulatively accounted for 73 per cent (Table 16). The first principal component correlated with days to 50 per cent flowering, plant height and pulling force. The traits such as inflorescence length, raceme number, raceme length and culm diameter, contributed more to the second component. Only grain yield per plant made a substantial contribution to the third component. The fourth component was loaded by weight per unit length of the culm and test weight. Investigation by Dhagat and Singh (1983) in a population consisting of 96 genetic stocks of kodo millet *via* Mahalanobis's D^2 and canonical analyses revealed that vegetative characters, days to 50 per cent flowering, days to maturity, plant height and straw yield, were important factors for differentiation among the germplasm accessions from different geographical regions. In this study also, days to 50 per cent flowering and plant height made substantial contribution to genetic variation than any other traits.

The scatter plot of the first Principal Components Axis (PC1) against the second Principal Component Axis (PC2) revealed that all the 41 kodo millet accessions could be grouped into five distinct PCA clusters, which were in conformity with the clusters obtained through hierarchical cluster analysis, except for the land race *Pacheri*, which got grouped in III PCA cluster instead of I as in the case of UPGMA method (Fig. 5).

Thus, principal component and cluster analyses complemented each other with almost similar pattern in cluster composition. The land races in clusters II and III had almost similar attributes except yield per plant. The traits related to duration, inflorescence and culm attributes were similar in both clusters. Hence, any improvement in culm strength could be made using the lines in these clusters. The clusters IV and V, in

contrast, had weak culm lines with short stature and short duration. The cluster I, had the maximum number of genotypes, with medium duration and intermediate culm strength, but with highest grain yield among the clusters. This group could be well explored for further selection and recombination breeding to overcome the low yield in kodo millet. Despite the limitation in estimating total genetic variation, this study indicated that morphological traits were useful for preliminary evaluation and can be used as a general approach for assessing genetic diversity among morphologically distinguishable kodo millet genotypes.

5.3 Association studies with biochemical traits and identification of superior lines suitable for machine harvest

Based on the mean performance in metric traits and qualitative traits, 15 genotypes were selected and forwarded to next level of evaluation and investigation. For comparison, seven genotypes which were extreme in their phenotype (one thick culm line, but low yielder and six thin culm lines which lodged severely in previous two seasons) were also included. All the 22 genotypes were evaluated for 15 morphometric and eight qualitative traits. The analysis of variance for all the 15 traits showed significant variations for all the traits except for number of productive tillers per plant.

In addition to morphological traits, biochemical analysis on cellulose, hemicellulose and lignin content were also carried out. Analysis of variance for biochemical traits had recorded non significant variations between genotypes for cellulose, hemicellulose and lignin content expressed in percentage in dry weight of the culm. But there were significant variations with respect to cellulose, hemicellulose and lignin content estimated per unit length of the culm as revealed in Barley by Kokubo *et al.* (1989) and wheat by Kong *et al.* (2013). Hence for further analyses and evaluation, biochemical content per unit length of the culm was considered.

Of the 13 test entries, selected based on culm strength the genotypes *viz.*, RK 50, Aamo 258, Sel 21, Aamo 13, Aamo 5, Aamo 210 and TNPsc 176 showed intermediate degree of lodging (score 5). This implicates that mere improvement for culm strength will not yield the desired non lodging phenotype. Hence, correlation studies with 22 genotypes were carried out involving biochemical traits and other culm related traits *viz.*,

cellulose, hemicellulose and lignin content per unit length of the culm, and pulling force, culm weight per unit length, culm diameter, recovery angle after bending and degree of lodging. To identify the traits which had direct effect on degree of lodging; path analysis was carried out keeping the remaining seven traits as independent.

5.3.1 Traits identified as correlated with degree of lodging

Six traits *viz.*, cellulose per unit length of the culm, hemicellulose per unit length of the culm, lignin per unit length of the culm, pulling force, culm weight per unit length, culm diameter had significant positive inter-correlation among themselves (Table 22). But the recovery angle after bending had significant positive correlation with pulling force and culm diameter (Table 22).

Degree of lodging had significant negative correlation with all the remaining seven traits (Table 22) *viz.*, cellulose per unit length of the culm, hemicellulose per unit length of the culm, lignin per unit length of the culm, pulling force, culm weight per unit length, culm diameter, and recovery angle after bending. Hence any improvement in lodging resistance could be achieved through improvement in these seven traits. This was in accordance with Wang and Li (1996), who reported that the accumulation of dry matter, especially the poly-saccharides, cellulose and hemicellulose in wheat culm resulted in lodging resistance.

5.3.2 Path analysis reveals lignin content and culm diameter influence degree of lodging

In this study path analysis was carried out to understand the direct and indirect effects of associated traits with degree of lodging (Table 23). The maximum negative direct effect on degree of lodging was recorded by lignin content per unit length of the culm (very high) (Fig. 12) followed by culm diameter (high). The negative effect implicates that higher the lignin content and culm diameter the lesser will be lodging.

The maximum positive direct effect on degree of lodging was recorded by culm weight per unit length (high), followed by hemicellulose per unit length of the culm (high). This means that these traits affect lodging resistance, but associated with lodging resistance through improvement in lignin content (indirect effect). Likewise, the traits cellulose per unit length of the culm, pulling force, culm diameter and recovery angle

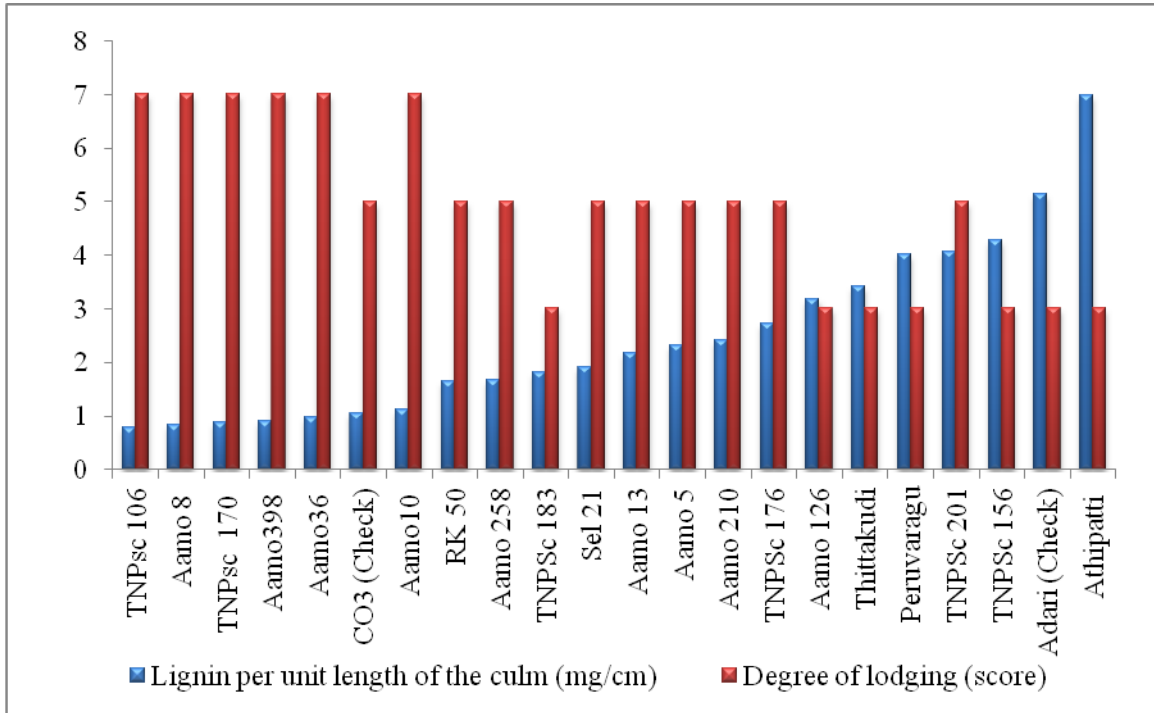


Figure 12. Degree of lodging vs. lignin content per unit length of the culm in 22 genotypes of kodo millet [Kharif, (Aug. - Dec.), 2013]

Genotypes sorted in ascending order with respect to lignin content (per unit length of the culm). High degree of lodging (score 7) registered in genotypes with low lignin content. Most of the genotypes on the higher side of lignin content exhibited low degree of lodging (score 3)

after bending were associated with lodging resistance through improvement in lignin content per unit length of the culm (indirect effects high).

Similar association of high lignin content with lodging resistance is reported in wheat (Ma, 2009 and 2010b) and pea (Banniza *et al.*, 2005). In rice, Ookawa and Ishihara (1993) reported that lignin played an important role in lodging resistance, but they suggested that its distribution and density were more important than its concentration.

Among the 13 test entries, Aamo 5, Aamo 210, Aamo 258, Sel 21, TNPsc 176 and TNPsc 183 were superior, with respect to culm and yield related traits. All these lines surpassed the check CO 3 in terms of yield (Fig.13). These six genotypes could be suggested as suitable to machine harvesting. Even among these six selected entries TNPsc 183 was the superior as it showed low degree of lodging in all the seasons. Aamo 13, Aamo 126, RK 50, TNPsc 201 and *Peruvaragu* were high yielders but not suitable for machine harvest as they were not uniform in their maturity. Considering the yield (per plot) alone the genotypes Sel 21, TNPsc 176 and TNPsc 183 excelled other genotypes (Fig. 13). Hence these three genotypes could be suggested for improving the yield level in kodo millet.

5.4 Characterization of select genotypes at anatomical and molecular level

The genotypes which were on the higher side of lignin content per unit length of the culm recorded lower yield. Hence, genotypes, Sel 21, TNPsc 176 and TNPsc 183 (Plates 18, 19,20,21,22 and 23) were selected based on culm diameter, lignin content and yield (Fig. 13). These three lines also had uniform maturity, absence of chlorosis and no shattering (Table 19). These three test entries were raised along with the two checks, CO3 and *Adari*. A thin culm line Aamo 10 was also included for comparison.

5.4.1 Anatomical studies

The stem is one of the most important plant organs playing a key function in transportation, storage and mechanical support. The transverse section of internodes from the center to the outer layer is mainly composed of different tissues: pith, parenchyma, vascular bundles, sclerenchyma and epidermis. Vascular bundles are distributed within the transverse section in two circles: (1) small vascular bundles close to the epidermis

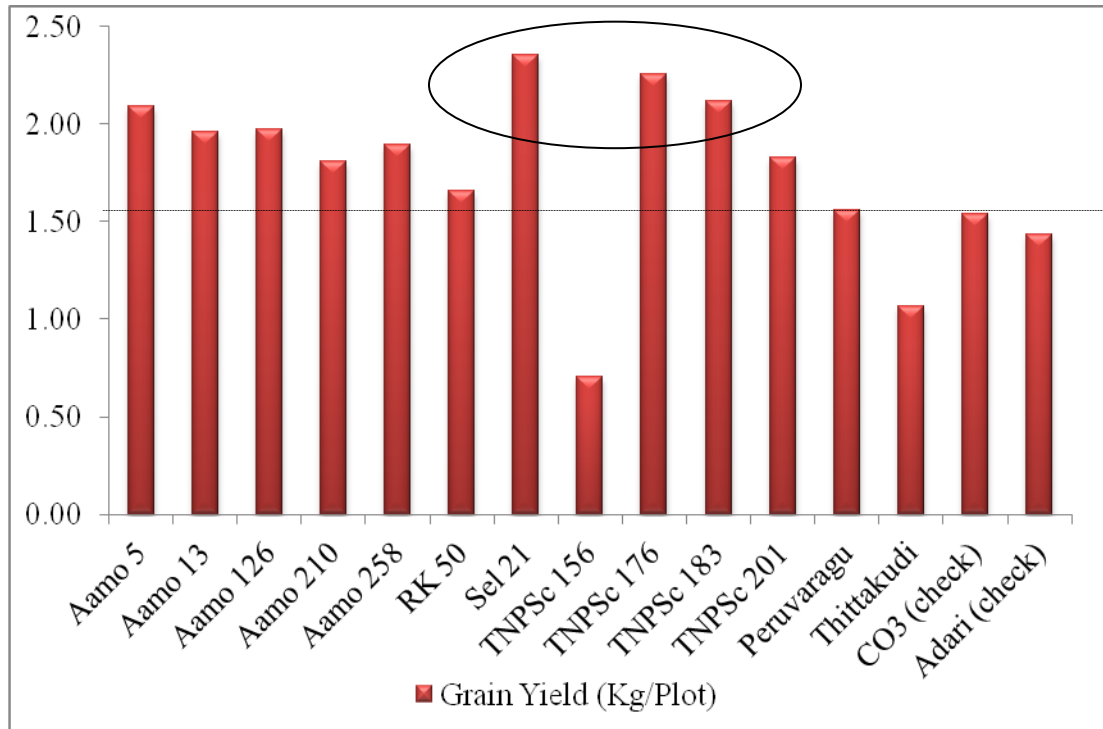


Figure 13. Average yield per plot of 13 kodo millet test entries with two checks [Kharif, (Aug. - Dec.), 2013]

Sel 21, TNPsc 176 and TNPsc 183 (encircled) recorded higher yield than other genotypes including CO3, the yield check. The horizontal line shows the yield level in CO3.



Plate 18. Kodo millet genotypes chosen for evaluation in summer (Jan.-May), 2014
a. Adari; b. CO3; c. Aamo 10; d. TNPsc 176; e. Sel 21; f. TNPsc 183



Plate 19. Inflorescences of kodo millet genotypes chosen for evaluation in summer (Jan.-May), 2014

a. Adari; b. CO3; c. Aamo 10; d. Sel 21; e. TNPsc 176; f. TNPsc 183



Plate 20. Variations in inflorescence morphology in kodo millet genotypes chosen for evaluation in summer (Jan.-May), 2014

The land race *Adari* presents itself with more number of racemes and highest inflorescence length in all. Among the three test entries, TNPsc 183 had the longest inflorescence



Plate 21. Variations in raceme type in kodo millet genotypes chosen for evaluation in summer (Jan.-May), 2014

Adari, CO3 and TNPsc 183 had longer raceme, with regular spikelet arrangement; Sel 21, TNPsc 176 and Aamo 10 had shorter raceme with irregular spikelet arrangement



Plate 22. Differences in culm diameter and culm thickness in kodo millet genotypes

Sel 21 had the highest culm thickness. Aamo 10 had the minimum culm thickness and diameter

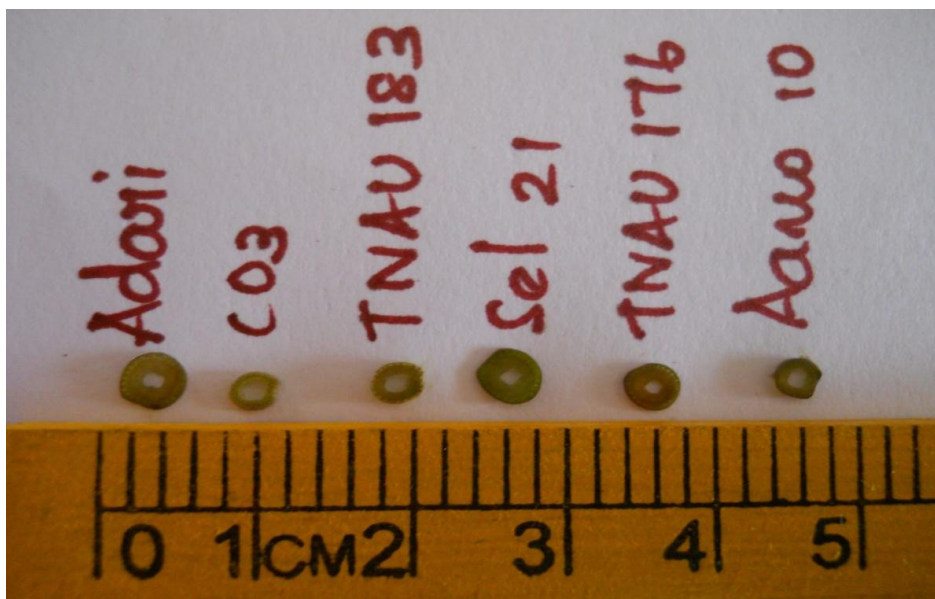


Plate 23. Differences in culm diameter and culm thickness in kodo millet genotypes

Sel 21 and Adari had the maximum culm diameter and thickness followed by TNPsc 176

embedded in sclerenchyma, which consists of fiber cells and (2) large vascular bundles, which are included in the parenchyma. The structure and composition of cell walls of different types of tissues are different to suit the functions, they are intended to perform. For example, parenchyma cells, which consist of primary walls, provide the main structural support in growing regions of the plant body. Sclerenchyma cells, which have both primary walls and thick secondary walls, provide the major mechanical support in non-elongating regions of the plant body (Carpita and McCann, 2000).

For studying the anatomical features, histo-chemicals, like TBO and Phloroglucinol-HCl were used. The TBO was used to reveal cellular details (Plate 13) and phloroglucinol-HCl, staining used to understand the localization and distribution of lignin. Lignifications were seen in sclerenchyma arc and in the fiber cells around the vascular bundles. As revealed through light microscope, (Plate 14), the width of the sclerenchyma arc was higher in two test entries Sel 21, TNPsc 176 and in *Adari*. But the intensity of the staining is more in TNPsc 183; this could be the reason for its low degree of lodging in all the four seasons.

To complement the observations made in light microscopy, SEM was also used to study the anatomical features underlying the variations, in select kodo millet genotypes. The SEM is a surface-based technique to reveal structure down to nanometer resolution. For anatomical study using SEM and EDX only four genotypes were chosen. They were *Adari*, the check for culm strength, Aamo 10, the weak line which constantly lodged in all the seasons, Sel 21, which proved to be with high yield, uniform maturity, no shattering, high culm diameter and pulling force, medium culm weight, higher seed weight, but with intermediate degree of lodging and TNPsc 183, which exhibited low degree of lodging throughout all the seasons, with high culm strength, but medium level of test weight and yield, were chosen.

Among the test entries only TNPsc 183 showed low degree of lodging. Sel 21 had the maximum culm thickness and sclerenchyma arc thickness but lodged. But the number of vascular bundles in Sel 21 and TNPsc 183 were on par. This means the additional thickness (Fig.6b) found in Sel 21 was not supported by the increased number of vascular bundles in Sel 21 (Fig. 6a). This means more vascular bundles per unit area in TNPsc 183

may be the reason for its lodging resistance. This again proves that rather than culm thickness, the traits, culm diameter and number of vascular bundles prove to be associated with non-lodging. A study by Wang *et al.* (2006) indicated that number of vascular bundles were an important factor affecting mechanical strength in wheat. In this study also though the culm diameters were similar in both Sel 21 and TNPsc 183, the lodging of Sel 21 could be attributed to the absence of proportional increase in vascular bundles with increased in culm thickness.

5.4.2 Silicon and Potassium composition using SEM-EDX

Silicon relative signal (%) using SEM-EDX in select kodo millet genotypes (Fig. 7a) revealed that the silicon content is more in genotypes with higher culm diameter, *viz.*, Sel 21, TNPsc 183 and *Adari*. But it is very low in the thin culm line Aamo 10. This could be one reason for the severe lodging observed in Aamo 10. Unlike other thick culm genotypes, which show lodging during grain filling, Aamo 10 showed severe lodging during flowering in all the seasons. Hence, it can be concluded that silicon content also plays an important role in strengthening the culm. Similar reports of association of silicon content with mechanical strength were available in rice (Zhang *et al.*, 2010a; Hasan *et al.*, 1993) and wheat (Gartner *et al.*, 1984)

Potassium relative signal (%) did not differentiate the lodging and non lodging line, because its content is lower in TNPsc 183 and Sel 21 and higher in *Adari* and Aamo 10. Hence, potassium content is not associated with lodging behavior in these four genotypes studied (Fig. 7b). This corroborates with the findings of Zuber and Loesch (1966) who reported that the correlation between potassium content and culm related traits such as crushing strength, ring thickness and weight of 2-inch section were negative and statistically not significant in maize. They observed that potassium content in lodging resistant maize lines were lower than lodging susceptible lines.

Silicon content (%) across cell types (sclerenchyma cells in the arc and around vascular bundles and parenchyma cells) did not differ significantly. It is almost similar in all the genotypes studied (Fig. 8c). But potassium content (%) in sclerenchyma cells in the arc and parenchyma cells differ significantly. Potassium content is higher in parenchyma cells, and lesser in sclerenchyma cells, indicating that potassium content is more in non-lignified

cells (Fig. 7d). Similar report of high potassium accumulation in axial parenchyma inferences was made in black heartwood (*Cryptomeria japonica*) using SEM-EDX (Matsunaga *et al.*, 2006). This indicates, in non-lignified cells like parenchyma, high potassium accumulation adds to its strength.

5.4.3 Expression studies of *FLEXIBLE CULM1* gene in kodo millet

Li *et al.* (2009a) identified a *flexible culm1* (*fc1*) mutant by screening a rice T-DNA insertion mutant library. This mutant exhibited an abnormal development phenotype, including late heading time, semi-dwarf habit, and flexible culm. The wild type, *FCI* is reported to encode a cinnamyl-alcohol dehydrogenase and mainly expressed in the sclerenchyma cells of the secondary cell wall and vascular bundle regions. Hence, a deficiency of *FCI* in the *fc1* mutant caused a reduction in cell wall thickness, as well as a decrease in lignin in these types of cells. Histological and biochemical analyses by them revealed that the p-hydroxyphenyl and guaiacyl monomers in *fc1* cell wall were reduced greatly. And, suggests that *FCI* plays an important role in the biosynthesis of lignin and the control of culm strength in rice.

In the present study, *FCI* gene was chosen on a priori basis. Quantification of *FCI* gene expression in select kodo millet genotypes revealed higher value for *Adari* (2.01) in relation to the weak culm line, *Aamo 10* (1.27). This suggests the association of *FCI* gene with high and low lignin content in *Adari*, 5.15 mg/cm and *Aamo 10*, 1.11 mg/cm, respectively. The test entries TNPsc 183 and Sel 21 had optimum *FCI* expression levels of 1.44 and 1.53, respectively (Fig. 8). The high regression value between gene expression level and lignin content per unit length of the culm (Fig. 14) confirms the major role played by *FCI* (*CAD7*) in lignin accumulation and its contribution to culm strength in kodo millet. *FCI* gene expression levels could be explored for its use as a marker in identifying high lignin and in turn high culm strength lines in other cereal members as well. A few other genes (*BRITTLE CULM1*, *BRITTLE CULM6*, *SHINE*, etc.) have been reported in literatures to contribute for culm strength. However, they could not be analyzed in kodo millet for the lack of sequence information and in turn the difficulty in finding perfectly conserved motifs to enable designing primers from orthologous sequences.

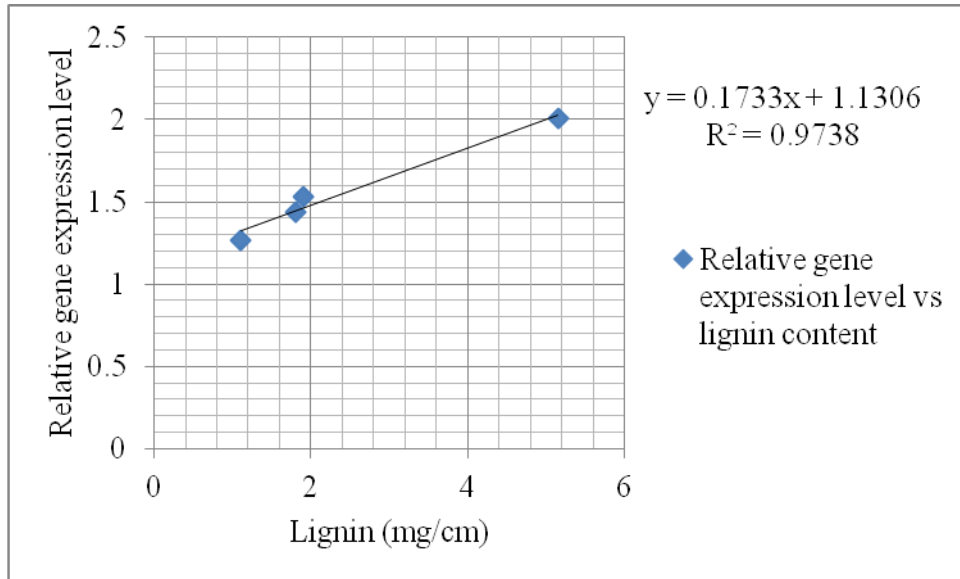


Figure 14. The linear regression of lignin content per unit length of the culm with relative *FCI* gene expression.

The high regression value confirms the association of *FCI* gene in lignin accumulation in kodo millet.

5.4.4 Evaluation of select three genotypes based on yield and lodging

Yield evaluation in three test entries, revealed that Sel 21 had the maximum yield followed by TNPsc 176 and TNPsc 183 (Table 25). The weak culm line Aamo 10 lodged severely with the onset of flowering. The contributory factors could be poor culm diameter, lower lignin content per unit length of the culm, poor number of vascular bundles and poor Si content. The test entry, Sel 21, had severe incidence of lodging during grain filling though they had high culm diameter, culm thickness, and more number of vascular bundles. The lodging observed in Sel 21 and TNPsc 176 might be due to the heavier upper part which in turn might be due to high grain yield and test weight. The TNPsc 183 showed low degree of lodging which could be attributed to high culm strength, more number of vascular bundles, high silicon content comparatively lesser upper part weight due to lesser yield and test weight than that of Sel 21 and TNPsc 176 (Plate 24). Hence, for a plant to remain non-lodging, there must be a balance between the upper and lower part of the plant. The proportionality between the physical strength in basal internodes and the weight of the upper part determines the vulnerability of a given cultivar to lodging (Zhang *et al.*, 2013). Accordingly, lodging occurred owing to loss of balance between the weight of upper part and the sturdiness of the basal culm (Dunn and Briggs, 1989; Li *et al.*, 2009b; Quang Duy *et al.*, 2004).

Considering the above features, TNPsc 183, which had low incidence of lodging, tall stature, uniform maturity, non shattering of seeds, low degree of chlorosis and better yield was identified as the genotype suitable for machine harvest. The genotypes, Sel 21 and TNPsc 176 were the high yielders and can be suggested for improving the yield in kodo millet.

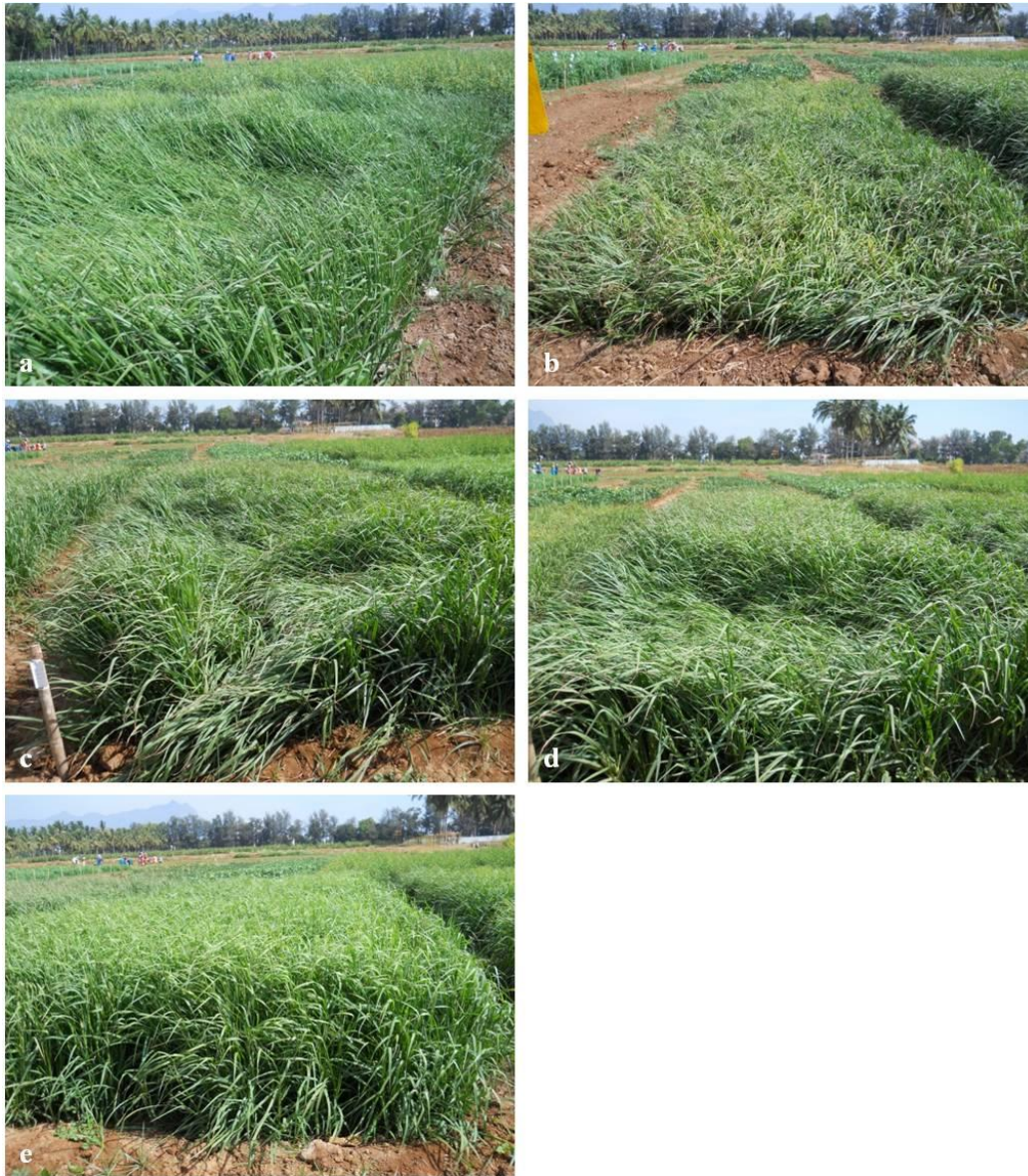


Plate 24. Degree of lodging in kodo millet genotypes chosen for evaluation in summer (Jan.-May), 2014

a. *Adari*; **b.** *Aamo 10*; **c.** *Sel 21*; **d.** *TNPsc 176*; **e.** *TNPsc 183*. Among these genotypes, *TNPsc 183* showing low degree of lodging compared to others

Summary

CHAPTER VI

SUMMARY

This study was carried out to identify high yielding kodo millet genotypes with strong culm suitable for mechanical harvesting. The study involved characterization of kodo millet germplasm with respect to traits contributing to culm strength and yield, identification of high yielding genotypes with strong culm and assessment of their performance over seasons. Further, factors contributing to culm strength at physical, biochemical, anatomical and molecular levels were also studied. The results of the study are summarized hereunder.

- A collection of 297 kodo millet germplasm accessions were evaluated for culm strength under an augmented design, with two checks (CO3, for yield and *Adari*, for culm strength). Correlation and path analyses in these lines indicated that culm diameter, plant height and recovery angle after bending could be the candidate traits based on which selection could be imposed to identify genotypes with good culm strength in kodo millet
- Based on these selection criteria, 41 accessions including the two checks were selected and forwarded to study the consistency of performance with respect to culm strength and yield.
- The extent of variability, proportion of heritable variation and the expected gain out of selection were assessed using the 41 genotypes.
- Variability studies suggested high level of genotypic and phenotypic variations for traits such as pulling force, culm weight per unit length and grain yield per plant indicating more scope for improving the culm strength as well as yield.
- High heritability for traits such as days to 50 per cent flowering, culm diameter, culm weight per unit length, grain yield per plant, pulling force and days to maturity shows that these traits are least affected by environment and hence more reliable for selection and further genetic improvement of the respective traits.

- Culm weight per unit length, grain yield per plant, pulling force and culm diameter exhibited high heritability along with high genetic advance, indicating preponderance of additive gene action for the expression of these four traits.
- Cluster and principal component analyses resulted in five clusters with almost similar pattern in cluster composition. In PCA, the first principal component, which accounted for 39 per cent of total variability highly correlated with days to 50 per cent flowering, plant height and pulling force.
- From the 41 lines, 13 were identified as having good culm strength and yield and those 13 were raised along with two checks, one strong culm line but with poor yield and six weak culm lines.
- Correlation and path analyses showed that only lignin per unit length of the culm had very high negative direct as well as indirect effects on degree of lodging, followed by culm diameter. Hence any improvement in lodging could be achieved through improving the lignin content and culm diameter.
- Among the 13 test entries, Aamo 5, Aamo 210, Aamo 258, Sel 21, TNPsc 176 and TNPsc 183 were superior, with respect to culm and yield related traits and hence these could be suggested as suitable to machine harvesting.
- From these six genotypes, only three high yielding genotypes (TNPsc 176, TNPsc 183 and Sel 21) were selected and forwarded for further evaluation based on yield, degree of lodging, anatomical and molecular characteristics.
- Silicon relative signal (%) using SEM-EDX proved that silicon content contributes for culm strength. Potassium relative signal (%) did not differentiate the lodging and non lodging lines and hence, not associated with lodging behavior atleast in these four genotypes studied. Moreover, K content is higher in non-lignified parenchyma cell walls.
- Expression studies by qRT-PCR with *FLEXIBLE CULM1* gene, involved in lignin biosynthetic pathway confirmed the role of *FC1* gene in lignin biosynthesis. The highest expression recorded in strong culm check *Adari* and the lowest expression in Aamo 10 proved the role of lignin in culm strength in kodo millet. The test entries TNPsc 183 and Sel 21 had medium levels of expression.

- Yield evaluation in three test entries, revealed that Sel 21 had the maximum yield followed by TNPsc 176 and TNPsc 183. The test entries, Sel 21 and TNPsc 176, had severe incidence of lodging during grain filling which might be due to heavier grains, since their test weight was more than TNPsc 183. On the other hand TNPsc 183 had low incidence of lodging. Hence, among the three genotypes, TNPsc 183 was identified to be an ideal line to suit machine harvest.

References

REFERENCES

- Ahluwalia, M., Singh, Diljit, Solaman, C. and Naqvi, S. 1970. Studies on variability in kodo millet (*Paspalum scrobiculatum* L.). **Andhra Agric. J.**, **17**: 58-60.
- Aliaga, J.R., Culianez, F. and Primo, E. 1986. Estudio descriptivo de variedades de arroz. Caja de Ahorros de Valencia. **Valencia**. 102 pp.
- Al-Qaudhy, W, Morris, R. and Mumm, R.F. 1988. Chromosome locations of genes for traits associated with lodging on winter wheat. **Crop Sci.**, **28**: 631-635.
- Amawate, J.S. 1965. Variability and correlation of important agronomic characters with yield in *Paspalum scrobiculatum* L., **M.Sc. (Agric.) Thesis**, JNKVV, Jabalpur, Madhya Pradesh, India.
- Amawate, J.S. and Dabral, K.C. 1970. Variability in kodo millet (*Paspalum scrobiculatum* L.). **Indian J. Agric. Sci.**, **40**: 616-619.
- Ambavaram, M.M.R., Krishnan, A., Trijatmiko, K.R. and Pereira, A. 2011. Coordinated activation of cellulose and repression of lignin biosynthesis pathways in rice. **Plant Physiol.**, **155**: 916-931.
- Aohara, T., Kotake, T., Kaneko, Y., Takatsuji, H., Tsumuraya, Y. and Kawasaki, S. 2009. Rice *BRITTLE CULM 5* (BRITTLE NODE) is involved in secondary cell wall formation in the sclerenchyma tissue of nodes. **Plant Cell Physiol.**, **50**: 1886-1897.
- Appenzeller, L., Doblin, M., Barreiro, R., Wang, H., Niu, X., Kollipara, K., Carrigan, L., Tomes, D., Chapman, M. and Dhugga, K.S. 2004. Cellulose synthesis in maize: isolation and expression analysis of the cellulose synthase (*CesA*) gene family. **Cellulose**, **11**: 287-299.
- Atkins, I.M. 1938. Relation of certain plant characters to strength of straw and lodging in winter wheat. **J. Agri. Res.**, **56**: 99-120.
- Banniza, S., Hashemi, P., Warkentin, T.D., Vandenberg, A. and Davis, A.R. 2005. The relationships among lodging, stem anatomy, degree of lignification, and resistance to mycosphaerella blight in field pea (*Pisum sativum*). **Canadian J. Bot.**, **83**: 954-967.

- Bernards, M.A. and Lewis, N.G. 1998. The macromolecular aromatic domain in suberized tissue: a changing paradigm. **Phytochem.**, **47**: 915-933.
- Berry, P.M., Griffin, J.M., Sylvester-Bradley, R., Scott, R.K., Spink, J.H., Baker, C.J. and Clare, R.W. 2000. Controlling plant form through husbandry to minimize lodging in wheat. **Field Crops Res.**, **67**: 51-58.
- Bhiah, K.M., Guppy, C., Lockwood, P. and Jessop, R. 2010. Effect of potassium on rice lodging under high nitrogen nutrition. Proceed. 19th World Congress of soil sci., soil solutions for a changing world, pp. 136-139.
- Bondale, K.V. 1994. Status of small millets in India. Paper presented in National seminar on ragi and small millets, held during January 6-7, 1994 at IGKV, Raipur India.
- Burton, G.W. 1952. Quantitative inheritance in grasses. **Proc. 6th Int. Grassland Cong.**, **1**: 277-283. Carpita, N. and McCann, M. (2000). The cell wall. In *Biochemistry and Molecular Biology of Plants*, B.B. Buchanan, W. Gruissem, and R.L. Jones, eds (Rockville, MD: American Society of Plant Physiologists), pp. 52-108.
- Carpita, N. and McCann, M. 2000. The cell wall. In *Biochemistry and molecular biology of plants*, B.B. Buchanan, G. Wilhelm, and R.L. Jones, eds (Rockville, IL: American Society of Plant Physiologists), pp. 52-108.
- Cenci, C.A., Grando, S. and Ceccarelli, S. 1984. Stem anatomy in barley (*Hordeum vulgare* L.) **Canadian J. Bot.**, **62**: 2023-2027.
- Chandrasekara, A. and Shahidi, F. 2010. Content of insoluble bound phenolics in millets and their contribution to antioxidant capacity. **J. Agric. Food Chem.**, **58**: 6706-6714.
- Chang, T.T. 1964. Varietal differences in lodging resistance. *Intl. Rice Comm. Newsl. Intl. Rice Res. Inst.*, Los Banos, Philippines. **13**: 1-11.
- Chang, T.T. and Vergara, B.S. 1972. Ecological and genetic information on adaptability and yielding ability in tropical rice varieties. In *International Rice Research Institute. Rice Breeding*, Los Banos, Philippines, pp. 431-453,
- Chaturvedi, G.S., Misra, C.H., Singh, C.N., Pandey, C.B., Yadav, V.P., Singh, A.K., Divivedi, J. L., Singh, B.B. and Singh, R.K. 1995. Physiological flash flooding. **Intl. Rice Res., Inst.**, Los Banos: 79-96.

- Ching, A., Dhugga, K.S., Appenzeller, L., Meeley, R., Bourett, T.M., Howard, R.J. and Rafalski, A. 2006. *Brittle stalk 2* encodes a putative glycosyl phosphatidylinositol-anchored protein that affects mechanical strength of maize tissues by altering the composition and structure of secondary cell walls. **Planta**, **224**: 1174-1184.
- Choudhary, L.B. and Singh, R. 1970. Variety x environment interaction in kodo millet under two levels of fertility. **Indian J. Agric. Sci.**, **40**: 977-983.
- Chuanren, D., Bochu, W., Pingqing, W., Daohong, W. and Shaoxi, C. 2004. Relationship between the minute structure and the lodging resistance of rice stems. **Colloids Surf. B. Biointerfaces**, **35**:155-158.
- Crook, M.J. and Ennos, A.R. 1993. The mechanics of root lodging in winter wheat (*Triticum aestivum* L.). **J. Exp. Bot.**, **44**: 1219-1224.
- Crook, M.J. and Ennos, A.R. 1994. Stem and root characteristics associated with lodging resistance in four winter wheat cultivars. **J. Agric. Sci.**, **123**: 167-174.
- Das Gupta, S. 1960. Point biserial correlation and its generalization. **Psychometrika**. **25**: 393-408.
- Dash, S.K., Pradhan, S.K., Mall, A.K., Singh, O.N. and Rao, G.J.N. 2011. Screening of potential donors for development of new plant types in irrigated rice (*Oryza sativa*) in Eastern India. **J. Crop and Weed**, **7**: 74-79.
- Datta, S.K. and Mikkelsen, D.S. 1985. Potassium nutrition of rice. In: Munson RD (Eds). Potassium in agriculture. ASA Madison WI.
- Davidson, J. and Phillips, M. 1930. Lignin as a possible factor in lodging of cereals. **Science**, **72**: 401- 402.
- de Wet, J.M.J., Prasada Rao, K.E., Mengesha, M.H. and Brink, D.E. 1983. Diversity in kodo millet, *Paspalum scrobiculatum*. *Economic Bot.*, **37**: 159-163.
- Dewey, D.R. and Lu, K.H. 1959. A correlation and path coefficient analysis of components of crested wheat grass seed production. **Agron. J.**, **51**: 515-518.

- Dhagat, N.K. 1978. Variability in inter-relationship and genetic divergence in *Paspalum scrobiculatum* (L.). **Ph.D. thesis**, Jawaharlal Nehru Krishi Vishwa Vidyalaya (JNKVV), Jabalpur, Madhya Pradesh, India.
- Dhagat, N.K. and Singh, S.P. 1983. Genetic Divergence in kodo millet. *Indian J. Genet. Plant Breed.*, 43: 168-172.
- Dhagat, N.K., Joshi, R.C., and Sharma, D. 1971. Correlation and heritability in kodo millet (*Paspalum scrobiculatum* L.). **Indian J. Agric. Sci.**, 41: 682-684.
- Dunn, G.J., and Briggs, K.G. 1989. Variation in culm anatomy among barley cultivars differing in lodging resistance. **Canadian J. Bot.**, 67:1838-1843.
- Easson, D.L., White E.M. and Pickles, S.J. 1993. The effects of weather, seed rate and cultivar on lodging and yield in winter wheat. **The J. Agric. Sci.**, 121: 145-156.
- Evans, L.S., Kahn-Jetter, Z., Marks, C. and Harmoney, K.R. 2007. **J. Torrey Bot. Soc.**, 134: 458- 467
- Falconer, D.S. 1981. Introduction to quantitative genetics. 3rd Ed. Logman Scientific & Technical, Logman House, Burnt Mill, Harlow, Essex, England.
- Federer, W.T. 1956. Augmented designs. **Hawaiian Planters Rec.**, 55: 191-208.
- Federer, W.T. 1961. Augmented designs with one way elimination of heterogeneity. **Biometrics**, 17: 447-473.
- Flintham, J.E., Borner, A., Worland, A.J. and Gale, M.D. 1997. Optimizing wheat grain yield: effects of *Rht* (gibberellin-insensitive) dwarfing genes. **The J. Agric. Sci.**, 128(1): 11-25.
- Fouere, A., Pellerin, S. and Duparque, A. 1995. A portable electronic device for evaluating root lodging resistance in maize. **Agron. J.**, 87: 1020-1024.
- Frei, M. 2013. Lignin: Characterization of a Multifaceted Crop Component. **The Scientific World J.**, 25 pp.
- Gahan, P.B. 1984. Plant histochemistry and cytochemistry - An introduction. Academic Press, London, 301pp.

- Gartner S., Charlot C. and Paris-Pireyre, N. 1984. Microanalyse de la silice et resistance à la verse mécanique du blé tender. **Physiol. Veg.**, **22**: 811-820.
- Goering, H. K. and Van Soest, P.J. 1975. Forage fiber analyses (apparatus, reagents, procedures, and some applications). Agric. Handbook No. 379. ARS USDA, Washington, DC.
- Hasan, S., Shimojo, M. and Goto, I. 1993. Chemical components influencing lodging resistance of rice plant and its straw digestibility *in vitro*. **AJAS.**, **6**: 41- 44.
- Hegde, P.S. and Chandra, T.S. 2005. ESR spectroscopic study reveals higher free radical quenching potential in kodo millet (*Paspalum scrobiculatum*) compared to other millets. **Food Chem.**, **92**: 177-82.
- Hegde, P.S., Rajasekaran, N.S. and Chandra, T.S. 2005. Effects of the antioxidant properties of millet species on oxidative stress and glycemic status in alloxan-induced rats. **Nutrition Res.**, **25**: 1109-1120.
- Hirano, K., Kotake, T., Kamihara, K., Tsuna, K., Aohara, T., Kaneko, Y., Takatsuji, H., Tsumuraya, Y. and Kawasaki, S. 2010. Rice *BRITTLE CULM3 (BC3)* encodes a classical dynamin *OsDRP2B* essential for proper secondary cell wall synthesis. **Planta**, **232**: 95-108.
- Idris M.D., Hossain M.M. and Choudhury F.A. 1975. The effect of silicon on lodging of rice in presence of added nitrogen. **Plant Soil**, **43**: 691-695.
- Inoue, M., Gao, Z.S. and Cai, H.W. 2004. QTL analysis of lodging resistance and related traits in Italian ryegrass (*Lolium multiflorum* Lam.). **Theor. Appl. Genet.**, **109**: 1576 -1585.
- Janoria, M. P. 1963. Variability and classification of *Paspalum scrobiculatum* (L.) of Madhya Pradesh. **M.Sc. (Agric.) Thesis**, JNKVV, Jabalpur, Madhya Pradesh, India.
- Jellum, M.D. 1962. Relationships between lodging resistance and certain stem characters in oats. **Crop Sci.**, **2**: 263–267.

- Jezowski, S., Surma, M. and Adamski, T. 1987. Diallel analysis of characteristics determining lodging resistance of barley (*Hordeum vulgare* L). An estimate of parental forms and F₁ hybrids regarding morphological and physical characters of the stem. **Genetica Polonica**, **28**: 333-348.
- Johnson, H.W., Robinson, H.F. and Comstock, R.E. 1955. Estimate of genetic and environmental variability in soybeans. **Agron. J.**, **47**: 314-318.
- Jones, L., Ennos, A.R. and Turner, S.R. 2001. Cloning and characterization of irregular xylem4 (*irx4*): a severely lignin-deficient mutant of *Arabidopsis*. **Plant J.**, **26**: 205-216.
- Kandaswamy, G., Ramamoorthy, N. and Manoharan, V. 1990. Genetic variability in kodo millet (*Paspalum scrobiculatum* L.). **Madras Agric. J.**, **77**: 9-12.
- Kashiwagi, T. and Ishimaru, K. 2004. Identification and functional analysis of a locus for improvement of lodging resistance in rice. **Plant Physiol.**, **134**: 676-683.
- Kashiwagi, T., Sasaki, H. and Ishimaru, K. 2005. Factors Responsible for decreasing sturdiness of the lower part in lodging of rice (*Oryza sativa* L.), **Plant Prod. Sci.**, **8**: 166-172.
- Kashiwagi, T., Togawa, E., Hirotsu, N. and Ishimaru, K. 2008. Improvement of lodging resistance with QTLs for stem diameter in rice (*Oryza sativa* L.). **Theor. Appl. Genet.**, **117**: 749-757.
- Kelbert, A.J., Spaner, D., Briggs, K.G. and King, J.R. 2004. The association of culm anatomy with lodging susceptibility in modern spring wheat genotypes. **Euphytica**, **136**: 211-221.
- Keller, M., Karutz, C.H., Schmid, J.E., Stamo, P., Winzeler, M., Keller, B. and Messmer, M.M. 1999. Quantitative trait loci for lodging resistance in a segregating wheat × spelt population. **Theor. Appl. Genet.**, **98**: 1171-1182.
- Khush, G.S. 1999. Green revolution: preparing for the 21st century. **Genome**, **42**: 646-655.

- Kokubo, A., Kuraishi, S. and Sakurai, N. 1989. Culm strength of barley: correlation among maximum bending stress, cell wall dimensions, and cellulose content. **Plant Physiol.**, **91**: 876-882.
- Kokubo, A., Sakurai, N., Kuraishi, S. and Takeda, K. 1991. Culm brittleness of barley (*Hordeum vulgare* L.) mutants is caused by smaller number of cellulose molecules in cell wall. **Plant Physiol.**, **97**: 509-514.
- Kong, E., Liu, D., Guo, X., Yang, W., Sun, J., Li, X., Zhan, K., Cui, D., Lin, J. and Zhang, A. 2013. Anatomical and chemical characteristics associated with lodging resistance in wheat. **The Crop J.**, **1**: 43-49.
- Kono, M. 1995. Physiological aspects of lodging. In Matsuo, T., Kumazawa, K., Ishii, R., Ishihara, K., Hirata, H. eds, Science of the rice plant, vol 2, Physiology, Food and Agriculture Policy Research Center, Tokyo, pp 971-982.
- Kono, M. and Takahashi, J. 1961a. Studies on the relationships between breaking strength and chemical components of paddy stem. **J. Soil. Sci. Manure**, **32**: 149-152.
- Kono, M. and Takahashi, J. 1961b. Studies on the relationship between breaking strength and holo-cellulose content of paddy stem. **J. Soil. Sci. Manure**, **32**: 461-465.
- Kotake, T., Aohara, T., Hirano, K., Sato, A., Kaneko, Y., Tsumuraya, Y., Takatsuji, H. and Kawasaki, S. 2011. Rice *Brittle culm 6* encodes a dominant-negative form of CesA protein that perturbs cellulose synthesis in secondary cell walls. **J. Exp. Bot.**, doi:10.1093/jxb/erq395
- Langseth, W. and Stabbetorp, H. 1996. The effect of lodging and time of harvest on deoxynivalenol contamination in barley and oats. **J. Phytopathol.**, **144**: 241-245.
- Lenka, D. and B. Mishra. 1973. Path coefficient analysis of yield in rice varieties. **Ind. J. Agric. Sci.**, **43**: 376-337.
- Li, Y.H. 1979. The morphological and anatomical structure of cereals. Shanghai sci. tech. press, pp. 203-294 (in Chinese with English abstract).
- Li, S., Fu, S., Zeng, Q., Zhao. and Zhou, B. 1994. Structural advantages of natural composites-Bamboo. **Biometrics**, **2**: 15-32.

- Li, Q.Q. 1998. Creation, evaluation and utilization of winter wheat germplasm. Shangdong Science and Technology Press, Jinan, China: 203-219.
- Li, H.B., Bai, K.Z., Kuang, T.Y., Hu, Y.X., Jia, X. and Lin, J.X. 2000. Structural characteristics of thicker culms in the high-yield wheat cultivars. **Acta Bot. Sinica**, **42**: 1258-1262.
- Li, Y., Qian, Q, Zhou, Y., Yan, M., Sun, L., Zhang, M., Fu, Z., Wang, Y., Han, B., Pang, X., Chen, M. and Li, J. 2003. *BRITTLE CULM1*, which encodes a COBRA-like protein, affects the mechanical properties of rice plants. **Plant Cell**, **15**: 2020–2031.
- Li, X., Yang, Y., Yao, J., Chen, G., Li, X., Zhang, O. and Wu, C. 2009a. *FLEXIBLE CULM1* encoding a cinnamyl-alcohol dehydrogenase controls culm mechanical strength in rice **Plant Mol. Biol.**, **69**: 685-697.
- Li, H. J., Zhang, X. J., Li, W. J., Xu, Z. J., Xu, H.. 2009b. Lodging resistance in japonica rice varieties with different panicle types. *Chinese J. Rice Sci.*, **23**: 191-196.
- Lush, J.L. 1940. Inter size correlation and regression of offspring on dams as a method of estimating heritability of characters. **Proc. Am. Soci. Animal Prod.**, **33**: 293-301.
- Ma, Q.H., Xu, Y., Lin, Z.B. and He, P. 2002. Cloning of cDNA encoding *COMT* from wheat which is differentially expressed in lodging-sensitive and -resistant cultivars. **J. Exp. Bot.**, **53**: 2281-2282.
- Ma, J.F. and Yamaji, N. 2006. Silicon uptake and accumulation in higher plants. **Trends in Plant Sci.**, **11**: 392-397.
- Ma, Q.H. 2009. The expression of caffeic acid 3-O-methyltransferase in two wheat genotypes differing in lodging resistance. **J. Exp. Bot.**, **60**: 2763-2771.
- Ma, J.F. 2010a. Silicon transporters in higher plants in book title: MIPs and Their role in the exchange of metalloids. series title: **Adv. Exp. Med. Biol.**, **679**: 99-109. Edited by: Thomas P. Jahn and Gerd P. Bienert Publisher Springer New York Print.
- Ma, Q.H. 2010b. Functional analysis of a cinnamyl alcohol dehydrogenase involved in lignin biosynthesis in wheat. **J. Exp. Bot.**, **61**: 2735-2744.

- Mallesh, N.G. and Hadimani, N.A. 1994. Nutritional and technological characteristics of small millets and preparation of value-added products from them. In. Riley, K.W., Gupta, S.C., Seetharman, A. and Mushonga, J.N. eds. *Advances in small millets*. New York: International Science Publisher. pp 271-287.
- Matsubayashi, M., Ito, R., Nomoto, T., Takase, T. and Yamada, N. 1967. Theory and practice of growing rice. **Fuji Publ. Co. Ltd., Tokyo**. 527 pp.
- Matsunaga, H., Shiotari, R., Matsumura, J., Oda, K., Utsumi, Y. and Yamamoto, K. 2006. Potassium distribution in black heartwood of sugi (*Cryptomeria japonica*) I: Localization in axial parenchyma cells. **J. Wood Sci.**, **52**: 95-100
- Mewa, D., Belay, G. and Bekele, E. 2013. Variability and trait association in culm and grain yield characteristics of recombinant inbred lines of *Eragrostis tef* × *Eragrostis pilosa*. **African J. Agric. Res.**, **8**: 2376-2384.
- Min, D.H., 2001. Studies on the lodging resistance with its subtraits of different height wheat varieties and correlation between plant height and yield. **J. Triticeae Crops**, **21**: 76-79 (in Chinese with English abstract).
- Monties B. 1989. Lignins. In: Harborne JB, *Methods in plant biochemistry*. vol. I: Plant phenolics. London: Academic Press, pp 113-157.
- Narayaswami, S. 1954. The structure and development of the caryopsis in some Indian millets III: *Paspalum scrobiculatum* L. **Bulletin of Torrey Botanical Club**, **81**: 288-299.
- Norden, A.J. and Frey, K. J. 1970. Factors associated with lodging resistance in oats. **Agron. J.**, **51**: 561-568.
- O'Brien, T.P., Feder, N. and McCully, M.E. 1964. Polychromatic staining of plant cell walls by toluidine blue O. *Protoplasma*, **59**: 367-373.
- Okada, M., Yamakawa, Y., Fujii, K., Nishiyama, H., Motomura, H., Kai, S., and Imai, T. 1967. On the new varieties of paddy rice, 'Hoyoku', 'Kokumasari' and 'Shiranui', **Bull. Kyushu Agr. Exp. Sta.**, **12**: 187-224.

- Ooawa, T., Todokoroand, Y., Ishihara, K. 1993. Changes in physical and chemical characteristics of culm associated with lodging resistance in paddy rice under different growth conditions and varietal differences of their changes. *Japanese J. Crop Sci.*, 62: 525-533.
- Ookawa, T. and Ishihara, K. 1992. Varietal difference of physical characteristics of the culm related to lodging resistance in paddy rice. *Japanese J. Crop Sci.*, 61: 419-425.
- Ookawa, T. and Ishihara, K. 1993. Varietal difference of the cell wall components affecting the bending stress of the culm in relation to the lodging resistance in paddy rice. *Japanese J. Crop Sci.*, 62: 378-384.
- Ookawa, T. Hobo, T., Yano, M., Murata, K., Ando, T., Miura, H., Asano, K., Ochiai, Y., Ikeda, M., Nishitani, R., Ebitani, T., Ozaki, H., Angeles, E.R., Hirasawa, T. and Matsuoka, M. 2010. New approach for rice improvement using a pleiotropic QTL gene for lodging resistance and yield. ***Nature Comm.***, 1:132.
- Padulosi, S., Bhag Mal, Bala Ravi, S., Gowda, J., Gowda, K.T.K., Shanthakumar, G., Yenagi, N. and Dutta, M. 2009. Food security and climate change: Role of plant genetic resources of minor millets. ***Indian J. Plant Genet. Resour.***, 22: 1-16.
- Panse, V.G. and Sukhatme, P.V. 1967. Statistical methods for agricultural workers. (2nd Edn.), ICAR Publication, New Delhi. pp 152-161.
- Parihar, S.S. 1985. Genetic variability, association analysis and genetic divergence in Kodo millet (*Paspalum scrobiculatum* L.). ***M.Sc. (Agric.) Thesis***, JNKVV, Jabalpur, Madhya Pradesh, India.
- Perrier, X., Flori, A. and Bonnot, F. 2003. Data analysis methods. In: Hamon, P., Seguin, M., Perrier, X., Glaszmann, J. C. Ed., Genetic diversity of cultivated tropical plants. Enfield Science Publishers. Montpellier. pp 43 - 76.
- Pinthus, M.J. 1967. Spread of the root system as an indicator for evaluating lodging resistance of wheat. ***Crop Sci.***, 7: 107-111.
- Pinthus, M.J. 1973. Lodging in wheat, barley and oats: the phenomenon, its causes and preventive measures. ***Adv. Agron.***, 25: 209-263.

- Quang Duy, P., Abe, A., Hirano, M., Sagawa, S. and Kuroda, E. 2004. Analysis of lodging-resistant characteristics of different rice genotypes grown under the standard and nitrogen-free basal dressing accompanied with sparse planting density practices. **Plant Prod. Sci.** **7**: 243-251.
- Ramaiah, K. and Mudaliar, S.D. 1934. Lodging of straw and its inheritance in rice (*Oryza sativa*). **Indian J. Agric. Sci.**, **4**: 880–894.
- Rangaswami Ayyangar, G.N. and Panduranga Rao, V. 1934. Studies in *Paspalum scrobiculatum* L., the Kodo millet. **Madras Agric. J.**, **22**: 419-425.
- Rao, S.S. 1991. Genetic variability in minor millets. **Indian J. Agric. Sci.**, **61**: 322-323.
- Rao, S.S. and Tiwari, R.D. 1988. Variability and correlation studies in kodo millet. **Millets Newsl.**, **7**: 53-54.
- Rao, Y.C., Yang, Y.L., Xin, D.D., Li, X.J., Zhai, K.E., Ma, B.J., Pan, J.W., Qian, Q. and Zeng, D.L. 2013. Characterization and cloning of a brittle culm mutant (*bc88*) in rice (*Oryza sativa* L.). **Chinese Sci. Bulletin Crop Genet.**, **58**: 3000-3006.
- Salpute, R.G. 1966. Correlation studies on yield attributes in *Paspalum scrobiculatum* L., **M.Sc. (Agric.) Thesis**, JNKVV, Jabalpur, Madhya Pradesh, India.
- Sambrook, J., Fritsch, E.F., and Maniatis, T. 1989. Molecular cloning: A laboratory Manual. II edn. Cold Spring Harbour Laboratory. Press, Cold Spring Harbour, N.Y.
- Sarker, Z.I., Shamsuddin, A.K.M., Rahman. and Ara, R. 2007. Genotypic and phenotypic correlation and path analysis for lodging resistance traits in bread wheat (*Triticum aestivum* L.). **Bangladesh J. Genet Pl. Breed.**, **20**: 51-57.
- Sato, K. 1959. Studies on the starch contained in the tissues of rice plant (IV). Starch content in the culm related to lodging. **Proc. Crop Sci. Soc. Japan**, **26**: 19.
- Sattler, S.E., Funnell-Harris, D.L. and Pedersen, J.F. 2010. Brown midrib mutations and their importance to the utilization of maize, sorghum, and pearl millet lignocellulosic tissues. **Plant Sci.**, **178**: 229-238.
- Schönbrodt, F.D. and Perugini, M. 2013. At what sample size do correlations stabilize? **J. Res. Personality**, **47**: 609-612.

- Scudamore, K.A. 2000. "Mycotoxins in stored grain". Crop management into the millennium. Home-Grown Cereal Authority, London, pp. 181-189.
- Sharma, S.K. 1988. Assessment of genetic variability for some quantitative and physiological traits in kodo millet. **M.Sc. (Agric.) Thesis**, JNKVV, Jabalpur, Madhya Pradesh, India.
- Sindhu, A., Langewisch, T., Olek, A., Multani, D.S., McCann, M.C., Vermerris, W., Carpita, N.C. and Johal, G. 2007. Maize *Brittle stalk2* encodes a COBRA-like protein expressed in early organ development but required for tissue flexibility at maturity. **Plant Physiol.**, **145**: 1444-1459.
- Singh, S.V. 1994. Seed improvements in small millets and role of minikit programme in their development. Paper presented in National seminar on ragi and small millets, held during January 6-7, 1994 at IGKV, Raipur India.
- Sivasubramanian, S. and P. Madhavamenon. 1973. Genetic analysis of quantitative characters in rice through diallel crosses. **Madras Agric. J.**, **60**: 1097-1102.
- Snedecor, G.N. and Cochran, W.G. 1967. Statistical Methods, Oxford IBH, New Delhi. 338 p.
- Stapper, M. and Fischer, R.A. 1990. Genotype, sowing date and plant spacing influence on high-yielding irrigated wheat in Southern New South Wales. II: Growth, yield and nitrogen use. **Aust. J. Agric. Res.**, **41**: 1021-1041.
- Subramanian, A., Nirmalakumari, A. and Veerabhadhiran, P. 2010. Trait based selection of superior kodo millet (*Paspalum scrobiculatum* L.) genotypes. **Electronic J. Pl. Breed.**, **1**: 852-855.
- Takahashi, E., 1995. Uptake mode and physiological functions of silica. In Matsuo, T., Kumazawa, K., Ishii, R., Ishihara, K. and Hirata, H. (eds). Science of the rice plant 2: Physiology, Food and Agriculture Policy Research Center, Tokyo, pp.1-111.
- Tanaka, K., Murata, K., Yamazaki, M., Onosato, K., Miyao, A., Hirochika, H. 2003. Three distinct rice cellulose synthase catalytic subunit genes required for cellulose synthesis in the secondary wall. **Plant Physiol.**, **133**: 73-83.

- Taylor, N.G., Scheible, W.R., Cutler, S., Somerville, C.R. and Turner, S.R. 1999. The irregular xylem 3 locus of *Arabidopsis* encodes a cellulose synthase required for secondary cell wall synthesis. **Plant Cell**, **11**: 769-779.
- Terashima, K., Akita, S. and Sakai, N. 1992. Eco-physiological characteristics related with lodging tolerance of rice in direct sowing cultivation. *Japanese J. Crop Sci.*, **61**: 380-387.
- Terashima, K., Ogata, T. and Akita, S. 1994. Eco-physiological characteristics related with lodging tolerance of rice in direct sowing cultivation. II. Root growth characteristics of tolerant cultivars to root lodging. *Japanese J. Crop Sci.*, **63**: 34-41.
- Terashima, K., Ogata, T., Akita, S. 1995. Eco-physiological characteristics related with lodging tolerance of rice in direct sowing cultivation. III. Relationship between the characteristics of root distribution in the soil and lodging tolerance. *Japanese J. Crop Sci.*, **63**: 243-250.
- Tinarelli, A., 1988. *Il riso*, 2nd ed. Edagricole. Bologna. 744 p. [In Italian].
- Tiwari, D.K. and Janoria, M.P. 1967. Breeding potentialities of *Paspalum scrobiculatum* (L). **JNKVV Res. J.**, **1**: 88-99.
- Tomita, M. 2009. Introgression of Green Revolution *sd1* gene into isogenic genome of rice super cultivar Koshihikari to create novel semidwarf cultivar 'Hikarishinseiki' (Koshihikari-sd1). **Field Crops Res.****114**: 173-181.
- Torro, I., Breto, P. and Garcia-Yzaguirre, A. 2011. Short communication. Assessing lodging resistance in rice: A comparison of two indirect testing methods. **Spanish J. Agric. Res.**, **9**: 1245-1248.
- Travis, A. J., Murison, S.D., Hirst, D.J., Walker, K.C. and Chesson, A. 1996. Comparison of the anatomy and degradability of straw from varieties of wheat and barley that differ in susceptibility to lodging. **Journal of Agric. Sci.**, **127**: 01-10.
- Van Soest, P. J. 1987. Soluble carbohydrates and the non-fiber components of feeds. **Large Auim. Vet.**, **42**: 44.
- Verma, S.N.P. and Salpute, R.G. 1969. Heritability studies in kodo millet (*Paspalum scrobiculatum* L.). **JNKVV Res. J.**, **3**: 77-79.

- Verma, S.N.P. and Singh, R.P. 1982. Association analysis in three maturing groups of kodo millet. **Indian J. Agric. Sci.**, **52**: 488-491.
- Vijayalakshmi, B., Vijay, D., Raju, P.R.K. and Satyanarayana, P.V. 2008. Genetic divergence of qualitative and quantitative characters in low land rice germplasm. **Crop Res.**, **36**: 212-214.
- Wang, Y. and Li, C.H. 1996. Advances in the study of wheat lodging resistance. **J. Shangdong Agric. Univ.**, **27**: 503-508 (in Chinese with English abstract).
- Wang, Y., Li, S.S., Li, A.F., Li, X.B., Wang, H.G. and Li, Q.Q. 2000. Evaluation of lodging resistance and relation and path analysis of lodging resistance traits in wheat. **Acta Bot. Boreali-Occident Sin.**, **20**:79-85.
- Wang, J., Zhu, J., Lin, Q., Li, X., Teng, N., Li, Z., Li, B., Zhang, A. and Lin, J. 2006. Effects of stem structure and cell wall components on bending strength in wheat. **Chinese Sci. Bulletin**, **51**: 815-823.
- Wang, J., Zhu, J., Huang, R. and Yang, Y. 2012. Investigation of cell wall composition related to stem lodging resistance in wheat (*Triticum aestivum* L. by FTIR spectroscopy). **Plant Signaling & Behavior**, **7**: 856-863.
- Weber, C.R. and Fehr, W.R. 1966. Seed yield losses from lodging and combine harvesting in soybeans. **Agron. J.**, **58**: 287-289.
- Won, J.G., Hirahara, Y., Yoshida, T. and Imabayashi, S. 1998. Selection of rice lines using SPGP seedling method for direct seeding. **Plant Prod. Sci.**, **1**: 280-285.
- Wu, L.L., Liu, Z.L., Wang, J.M., Zhou, C.Y. and Chen, K.M. 2011. Morphological, anatomical, and physiological characteristics involved in development of the large culm trait in rice. **Australian J. Crop Sci.**, **5**: 1356
- Wu, B., Zhang, B., Dai, Y., Zhang, L., Shang-Guan, K., Peng, Y., Zhou, Y. and Zhu, Z. 2012. *Brittle Culm15* encodes a membrane-associated chitinase-like protein required for cellulose biosynthesis in rice. **Plant Physiol.**, **159**: 1440-1452.
- Xiao, S.H., Zhang, X.Y., Yan, C.S., Zhang, W.X., Hai, L. and Guo, H.J. 2002. Determination of resistance to lodging by stem strength in wheat. **Agric. Sci. China**, **35**: 7-11.

- Yadava, H. S. 1997. Retrospects and prospects of kodo millet in Indian agriculture. National seminar on small millets: Current research trends and future priorities as food feed and in processing for value addition. pp 23-24, TNAU, Coimbatore (TN).
- Yadava, H.S. and Jain, A.K. 2006. Advances in kodo millet research. Directorate of Information and Publications of Agriculture, Indian Council of Agricultural Research.
- Yan, C., Yan, S., Zeng, X., Zhang, Z. and Gu, M. 2007. Fine Mapping and Isolation of *Bc7(t)*, Allelic to *OsCesA4*. **J. Genet. Genom.**, **34**: 1019-1027.
- Yao, J., Ma, H., Zhang, P., Ren, L., Yang, X., Yao, G., Zhang, P. and Zhou, M. 2011. Inheritance of stem strength and its correlations with culm morphological traits in wheat (*Triticum aestivum* L.). **Can. J. Plant Sci.**, **91**: 1065-1070.
- Yasin, M., Verma, S.N.P., Jain, A.K. and Gupta, J.C. 1988. Variability and correlation studies in kodo millet. **Millet News.**, **7**: 52.
- Yu, S.B., Xu, W.J., Vijayakumar, C.H.M., Ali, J., Fu, B.Y., Xu, J.L., Jiang, Y.Z., Marghirang, R., Domingo, J., Aquino, C., Virmani, S.S. and Li, Z.K. 2003. Molecular diversity and multilocus organization of the parental lines used in the International rice molecular breeding program. **Theor. Appl. Genet.**, **108**: 131-140.
- Zhang, F., Jin, Z., Ma, G., Shang, W., Liu, H., Xu, M. and Liu, Y. 2010a. Relationship between lodging resistance and chemical contents in culms and sheaths of Japonica rice during grain filling. **Rice Sci.**, **17**: 311-318.
- Zhang, M., Zhang, B., Qian, Q., Yu, Y., Li, R., Zhang, J., Liu, X., Zeng, D., Li, J. and Zhou, Y. 2010b. *Brittle Culm 12*, a dual-targeting kinesin-4 protein, controls cell-cycle progression and wall properties in rice. **Plant J.**, **63**: 312-328.
- Zhang, W., Li, G., Yang, Y., Li, Q., Zhang, J., Liu, J., Wang, S., Tang, S. and Ding, Y. 2013. Effects of nitrogen application rate and ratio on lodging resistance of super rice with different genotypes. *J. Integrative Agric.*, Doi: 10.1016/S2095-3119(13)60388-3

- Zhu, L.,i, Shi, G.X., Li, Z.S., Kuang, T.Y., Li, B., Wei, Q.K., Bai, K.Z., Hu, Y.X. and Lin, J.X. 2004. Anatomical and chemical features of high-yield wheat cultivar with reference to its parents. **Acta Botanica Sinica**, **46** (5): 565-572.
- Zuber, M.S. and Loesch, P.J. 1966. Total ash and Potassium content of stalks as related to stalk strength in Corn (*Zea mays* L.). **Agron. J.**, **58**: 426-428.
- Zuber, U., Winzeler, H., Messmer, M.M., Keller, M., Keller, B., Schmid, J.E. and Stamp, P. 1999. Morphological traits associated with lodging resistance of spring wheat (*Triticum aestivum* L.). **J. Agron. Crop Sci.**, **182**: 17-24.

Publications



Research Article

Selection criteria for culm strength in Kodo millet (*Paspalum scrobiculatum*L.) to suit mechanical harvesting

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(Received: 13 May 2014; Accepted:08 Jul 2014)

Abstract

Kodo millet is an important small millet crop suited to dry land and subsistence farming in India. Due to scarcity of agricultural manpower, farm mechanization turns out to be inevitable to sustain the cultivation of millets. To facilitate mechanical harvesting the plant type should be non-lodging and sturdy. Hence, a thorough understanding of the basis for mechanical strength in kodo millet is a pre-requisite for identifying genotypes ideal for mechanical harvesting. Towards this objective, 297 kodo millet germplasm accessions were phenotyped for culm strength and other related traits. The study indicated that culm strength, measured in terms of pulling force, was positively correlated with culm diameter and plant height. But, it was independent of traits such as basal internode length, recovery angle after bending and degree of lodging. Thus, to identify non-lodging lines with sufficient culm strength to suit mechanical harvest, simultaneous selection for higher pulling force and recovery angle after bending, along with higher culm diameter and plant height appears to be relevant to identify genotypes suitable for mechanical harvest. Path analysis revealed that culm diameter is having the highest positive direct effect on pulling force followed by plant height and recovery angle after bending.

Key words:

Kodo millet, correlation, culm strength, pulling force, mechanical harvesting

Introduction:

Kodo millet (*Paspalum scrobiculatum* L), is a tropical small millet indigenous to India (de Wet *et al.*, 1983) and grown for its grain and fodder. It is a traditional, long duration, hardy and drought resistant crop cultivated in about 9 lakh hectares in India with an annual production of 3.11 lakh tonnes (Bondale 1994; Singh 1994). The area under kodo millet cultivation is witnessing a declining trend in the post-green revolution period due to predominance of the major cereals such as rice and wheat. However, an intensified drive to increase the acreage of small millets is important because millets still contribute to the regional food security of the dry and marginal lands, where major cereal crops fail to yield. Nowadays, thrust to grow millets is given due to their nutritional superiority as compared to the major cereals. Kodo millet has been reported to have higher free radical quenching potential when compared to other millets (Hegde and Chandra, 2005). Besides, it provides low priced protein, minerals and vitamins in form of sustainable food (Yadava and Jain, 2006). Growing health consciousness among the consumers also creates demand for this type of nutri-cereals which are anti-diabetic and anti-oxidant in nature (Chandrasekara and and Shahidi, 2010). Hence, technological intervention in this crop is essential to boost the production on a profitable scale.

Kodo millet is predominantly grown as a pure crop and yields high net returns as compared to other dry land crops owing to its high unit area

productivity and market price of the produce in addition to its fodder value. Due to scarcity of labour-, farmers use paddy combine harvesters to reap the crop. However, the spreading plant habit and poor culm strength of land races and high yielding varieties at present, leads to heavy lodging at maturity and therefore grain loss is more if harvested mechanically. Given the context, genotypes that are non-lodging with sufficient culm strength may be effectively cut by the blades of the mechanical harvester. Direct evaluation of each genotype for suitability to mechanical harvesting by raising them in large plots is not practicable. Hence, identifying traits that contribute for culm strength in kodo millet forms the premise of this investigation. Correlation gives an idea about the related traits which can be studied to identify superior lines with respect to specific phenome. Path analysis helps to partition the correlation coefficients into direct and indirect effects and provide a clear understanding of true relationships between associated traits. Therefore, this work attempts to identify the morphological traits contributing to culm strength in kodo millet and their nature of association.

Material and methods

A total of 297 germplasm accessions of kodo millet maintained by Small millets unit of Department of Millets were raised along with two checks, CO3 (a high yielding variety released by Tamil Nadu Agricultural University, India) and *Adari* (a local land race under cultivation in southern parts of India). The experiment was laid

out during *kharif* (Aug – Dec), 2012 at Department of Millets, Centre for Plant Breeding and Genetics, Tamil Nadu Agricultural University (TNAU), in Augmented Block Design (Federer, 1956, 1961) with four blocks and the checks placed randomly in each block. Each accession was raised in a single row with 45 x 10 cm spacing and all the recommended package of practices was adopted. Observations were recorded on ten morphological traits as described in Table 1.

Analysis of variance for augmented block design was done using the online statistical tool available at Indian Agricultural Statistics Research Institute (IASRI) website (<http://www.iasri.res.in/spadweb/default.aspx>). Pearson's linear correlation coefficient (r) was calculated for all the combinations of traits. Since the trait 'degree of lodging' was recorded as a categorical data, Point biserial correlation coefficient (r_{pb}) (<http://vassarstats.net/pbcorr.html>) Das Gupta (1960) was used to estimate the correlation between degree of lodging and other continuous data. Microsoft office excel 2007 was used to draw the polynomial regression curves. Correlation coefficients were partitioned into direct and indirect effects using path analysis suggested by Dewey and Lu (1959). Schonbrodt and Perugini (2013) reported that the sample size should approach 250 for stable estimates of correlation. In this study, the sample size is 299 including checks hence the correlation can be highly relied upon.

Results and Discussion

The analysis of variance was observed to be significant for all the traits studied (Table 2). The range of variability present for each trait in the germplasm is given in Table 3. The pulling force, which quantifiably represents the culm strength, exhibited the high positive correlation with culm diameter (0.544), plant height (0.379), number of productive tillers per plant (0.324), grain yield per plant (0.219) and days to 50% flowering (0.155). But, pulling force has no correlation with internode length, recovery ability after bending and degree of lodging. Plant elasticity represented by recovery angle after bending had negative correlation with internode length (-0.392) and degree of lodging (-0.470). The correlation between internode length and degree of lodging is positive and highly significant (0.490). Grain yield per plant had positive correlation with plant height (0.465), number of productive tillers per plant (0.191), culm diameter (0.206), internode length (0.250), degree of lodging (0.240) and pulling force (0.219), and negative association with recovery ability after bending (-0.316) (Table 4).

In this study, path analysis was carried out to find out the direct and indirect effects of associated traits with pulling force (Table 5). Path analysis revealed that culm diameter (0.378) had highest

positive direct effect on pulling force followed by plant height (0.292) and recovery angle after bending (0.255).

Traits related to Culm strength: The correlation of pulling force with other traits indicated that the lines with high culm strength are taller with thicker culm, more productive tillers and yield, but are late in flowering. This result corroborated with the findings of Kashiwagi et al. (2008), who reported that the stem diameter was positively correlated with pushing resistance in rice. The insignificant correlation of pulling force with recovery angle after bending is in accordance with the earlier findings of Torro et al. (2011) in rice, who reported that culm strength and elasticity are two independent traits. The correlation between degree of lodging and recovery angle is negative, i.e., the plants which recovered better seldom lodged. Hence, to assess lodging resistance alone recovery angle would be the ideal trait for screening as reported by Torro et al. (2011). But to identify non-lodging genotypes with good culm strength to suit mechanical harvesting, simultaneous selection for high pulling force and recovery angle has to be done as evidenced from this study.

Traits related to lodging

Recovery angle after bending had high negative correlation with degree of lodging, plant height, internode length and grain yield per plant. This reveals that lines which are tall with more grain weight and longer basal internodal length will have poor recovery after bending. Degree of lodging was negatively associated with days to flowering and days to maturity. This suggests that short duration lines are more prone to lodging which may be due to shorter time window available for strengthening and lignification of culm which is needed for culm strength (Li *et al.*, 2000). But the correlation between degree of lodging and plant height is positive. This could be due to the fact that, lodging happens in short duration lines because of its poor culm strength, which is decided by its genetic makeup. On contrary, the tall plants with sufficient culm strength normally stand erect, but any adverse environmental conditions like high plant density, heavy rain or wind lodges the plant, the recovery to original position demands more force. Hence the tall plants fail to recover after lodging. Moreover, there is no correlation between degree of lodging and culm diameter, which reinstates the above statement that both weaker and strong culms may tend to lodge, the former for its inherent weakness in culm and inability to stand erect and the latter because of its inability to recover upon lodging. So the ideal plant type has to be medium tall with sufficient culm strength which can stand while being cut by the blades of the mechanical harvester and can also recover better if lodged because of any adverse environmental conditions.

Internode length was positively correlated with plant height and degree of lodging and negatively with recovery angle after bending, but its correlation with culm diameter is insignificant. This confirms the earlier work of Aliaga *et al.* (1986), who reported that the culm elasticity is correlated with internode length, but not with culm thickness in rice. Internode length also is negatively correlated with days to 50% flowering and days to maturity. This suggests that the short duration lines are with longer basal internodes giving them the elasticity to bend.

Traits related to grain yield

Grain yield per plant is the most important trait which should also be given due consideration, because increased return is the ultimate goal of any study. Though the grain yield increases with plant height, it becomes static beyond a height of 60 cm (Fig. 1). Likewise if the pulling force and culm diameter increases beyond 6 Newton (Fig. 2) and 4 mm (Fig. 3), respectively, there is no corresponding increase in the grain yield, this could be because of diversion of source to strengthen the culm at the cost of yield. Moreover, the grain yield decreases with days to maturity. Hence an ideal cultivar should be medium tall and medium duration with optimal pulling force and culm diameter.

In *path analysis*, the culm diameter, which registered high positive direct effect on pulling force, is the best selection index for assessing culm strength. Following this, plant height and recovery angle after bending also have the true positive association with pulling force. So, to identify genotypes with good culm strength in kodo millet, culm diameter, plant height and recovery angle after bending could be the candidate traits upon which selection has to be imposed.

Though this study aims to identify traits related to mechanical strength, traits related to yield and lodging resistance were also included to arrive at a plant type suitable for machine harvest without compromising on yield. This could be the reason for high *residual effect* (Table 5). Moreover, apart from the morphological traits studied, polysaccharides (Cellulose, Hemi-cellulose), lignin (Li *et al.*, 2003) and other elemental (Silicon, Potassium) composition (Datta and Mikkelsen, 1985; Idris *et al.*, 1975; Ma and Yamaji, 2006) of the culm may have a direct bearing on culm strength. Hence these traits need to be explored in kodo millet for better understanding of the culm strength.

Thus, culm diameter which recorded the highest estimate of correlation and direct effect with pulling force is the most important morphological trait to assess the culm strength followed by plant height and recovery angle after bending. Selection

strictly for maximum culm strength may be only at the cost of yield. Therefore, as a trade off, selection of medium tall and medium duration with sufficient pulling force, culm diameter and recovery angle after bending should be considered to identify lines to suit mechanical harvest. However, further study of the culm biochemistry and anatomy can throw more light on the features that could impart high strength to the culm of kodo millet.

Acknowledgement: Funding by UGC, New Delhi to undertake the above research is gratefully acknowledged.

References

- Aliaga, J.R., Culianez, F. and Primo, E. 1986. Estudio descriptivo de variedades de arroz. Caja de Ahorros de Valencia. Valencia. 102.
- Bondale, K.V. 1994. Status of small millets in India. Paper presented in *National Seminar on Ragi and Small Millets*, held during January 6-7, 1994 at IGKV, Raipur India.
- Chandrasekara, A. and Shahidi, F. 2010. Content of Insoluble Bound Phenolics in Millets and Their Contribution to Antioxidant Capacity. *J. Agril. and Food Chemistry*, **58**: 6706-6714.
- Das Gupta S. 1960. Point biserial correlation and its generalization. *Psychometrika*, **25**: 393-408.
- Datta, S.K. and Mikkelsen, D.S. 1985. Potassium nutrition of rice. In: Munson, R.D., ed. Potassium in agriculture. ASA, Madison, WI.
- de Wet, J.M.J., Prasada Rao, K.E., Mengesha, M.H. and Brink, D.E. 1983. Diversity in Kodo Millet, *Paspalum scrobiculatum*. *Economic Bot.*, **37**: 159-163.
- Dewey, D.R. and Lu, K.H. 1959. A correlation and path coefficient analysis of components crested wheat grass and seed production. *Agron. J.*, **52**: 515-518.
- Federer, W. T. 1956. Augmented designs. *Hawaiian Planters Record*. **55**:191-208.
- Federer, W.T. 1961. Augmented designs with one way elimination of heterogeneity. *Biometrics*, **17**: 447-473.
- Hegde, P.S. and Chandra, T.S. 2005. ESR spectroscopic study reveals higher free radical quenching potential in kodo millet (*Paspalum scrobiculatum*) compared to other millets. *Food Chemistry*, **92**: 177-82.
- Idris, M.D., Hossain, M.M. and Choudhury, F.A. 1975. The effect of silicon on lodging of rice in presence of added nitrogen. *Plant Soil* **43**: 691-695.
- Kashiwagi, T., Eiji Togawa, Naoki Hirotsu and Ishimaru, K. 2008. Improvement of lodging resistance with QTLs for stem diameter in rice (*Oryza sativa* L.). *Theor. Appl. Genet.*, **117**: 749-757.
- Li H.B., Bai, K.Z., Kuang, T.Y., Hu, Y.X., Jia, X., Lin, J.X. 2000. Structural characteristics of thicker-culms in the high-yield wheat cultivars. *Acta Bot Sin.* **42**: 1258-1262.
- Li Y., Qian, Q., Zhou, Y., Yan, M., Sun, L., Zhang, M., Fu, Z., Wang, Y., Han, B., Pang, X., Chen, M. and Li, J. 2003. BRITTLE CULM1, which encodes a COBRA-like protein, affects the



- mechanical properties of rice plants. *Plant Cell* **15**: 2020-2031.
- Ma, J.F. and Yamaji, N. 2006. Silicon uptake and accumulation in higher plants. *Trends in Plant Sci.*, **11**:392-397.
- Schönbrodt, F.D. and Perugini, M. 2013. At what sample size do correlations stabilize? *J. Res. in Personality*, **47**: 609-612.
- Singh, S.V. 1994. Seed improvements in small millets and role of minikit programme in their development. Paper presented in *National Seminar on Ragi and Small Millets*, held during January 6-7, 1994 at IGKV, Raipur India.
- Torro, I., Breto, P. and Garcia-Yzaguirre, A. 2011. Short communication. Assessing lodging resistance in rice: A comparison of two indirect testing methods. *Spanish J. Agrl. Res., North America*, **9**: 1245-1248.
- Yadava, H.S., Jain, A.K., 2006. Advances in kodo millet research ISBN:81-7164-062-1, Directorate of Information and Publications of Agriculture, Indian Council of Agricultural Research, 2006 Indian Council of Agricultural research.



Table 1. Description of the traits studied

Sl. No	Trait	Description
1	Days to 50% flowering (DF)	Counted as days from sowing to 50% of the plants in flowering
2	Plant height (cm) (PH)	Taken from base of the plant to tip of the panicle in the main tiller
3	Number of productive tillers (Ptil)	Total number of productive tillers in the plant
4	Culm diameter (mm) (CD)	The machine cut the plant at the base of the plant approximately at a height of 20 cm. Hence, the culm diameter is measured at middle of the second internode from the base of the plant on the main tiller, using digital vernier caliper. The cross section of the internode is elliptical, hence, the shorter diameter is measured. (Fig 4)
5	Internode length (cm) (IntL)	The length of the second internode from the base of the plant on the main tiller is measured
6	Pulling Force (Newton) (PF)	The force required to bend the whole plant to an angle of 45° from the ground level measured using force gauge (Torro <i>et al.</i> , 2011) after setting the gauge to read the force at peak-hold. This setting will give the maximum force required from erect position to 45° angle. (Fig 5)
7	Recovery angle after bending (degrees) (RA)	The angle of recovery made by the plant after bending it to the ground level by holding the tip of the plant (this is a slight modification of the trait described by Torro <i>et al.</i> (2011) in rice). Instead of tiller angle difference, the angle of recovery is taken into account here. To read the angle, a protractor is kept behind the plant. (Fig 6)
8	Days to maturity (DM)	Number of days taken from sowing to physiological maturity of 50% of the main tillers
9	Grain yield per plant (gm) (GY)	Mean yield of three random plants.
10	Degree of lodging (DL)	0: non lodging; 1: lodging

*Except days to 50% flowering all the traits have been measured at physiological maturity of the crop.

Table 2. Anova for Augmented Block Design I

Sl.No	Source	Mean sum of squares				
		Treatments (df =298)	Checks (df =1)	Test entries (df =296)	CheckxTest entries (df =1)	Error (df =6)
1	Days to flowering	83.05*	465.12**	77.42*	1369.13**	14.79
2	Plant height	110.62**	269.93**	101.79**	2565.39**	11.86
3	Numbers of productive tillers	6.77**	8.14*	6.78**	2.25	0.87
4	Culm diameter	0.37*	0.75*	0.34*	7.65**	0.07
5	Basal internode length	5.77*	2.21	5.79*	3.13	0.84
6	Pulling force	1.30**	0.35	1.30**	2.35**	0.08
7	Recovery angle after bending	121.23*	153.12*	120.95*	172.79*	18.40
8	Days to maturity	91.05**	512.00**	84.31**	1664.03**	7.90
9	Grain yield per plant	62.72*	49.21	61.86*	331.04**	10.01

Table 3: Variability observed in the genotypes

Sl.No	Traits	Min	Max	Mean±SE
1	Days to flowering (days)	64.00	127.00	78.21±3.85
2	Plant height (cm)	20.33	70.20	40.83±3.44
3	Numbers of productive tillers	3.33	20.00	7.12±0.94
4	Culm diameter (mm)	1.72	5.45	2.92±0.28
5	Internode length (cm)	3.25	19.50	9.38±0.92
6	Recovery angle after bending (degrees)	20.00	80.00	54.89±4.29
7	Days to maturity (days)	102.00	172.00	121.23±2.81
8	Grain yield per plant (gm)	1.11	57.55	17.39±3.16
9	Pulling force (Newton)	0.86	7.78	2.54±0.30



Table 4. Simple correlation coefficients between pulling force and other traits

Characters	Days to flowering (days)	Plant height (cm)	Numbers of productive tillers	Culm diameter (mm)	Internode length (cm)	Recovery angle after bending (degrees)	DL	Days to maturity (days)	Grain yield per plant (gm)
Plant height (cm)	0.088								
Numbers of productive tillers	-0.113	0.416**							
Culm diameter (mm)	0.351**	0.521**	0.257**						
Internode length (cm)	-0.359**	0.556**	0.369**	-0.024					
Recovery angle after bending (degrees)	0.095	-0.432**	-0.176**	-0.135*	-0.392**				
DL	-0.250**	0.390**	0.100*	0.010	0.490**	-0.470**			
Days to maturity (days)	0.857**	-0.09	-0.320**	0.193**	-0.453**	0.160**	-		
Grain yield per plant (gm)	-0.107	0.465**	0.191**	0.206**	0.250**	-0.316**	0.290**	-0.190**	
Pulling force (Newton)	0.155*	0.379**	0.324**	0.544**	-0.026	0.103	0.240**	0.036	0.219**

*significant at 5% level; **significant at 1% level

Table 5. Direct and indirect effects of different traits on pulling force

Traits	DF	PH	Ptil	CD	IntL	RA	DL	DM	GY	Correlation with pulling force
DF	-0.054	0.026	-0.021	0.133	0.061	0.024	0.017	-0.020	-0.011	0.155*
PH	-0.005	0.292	0.078	0.197	-0.094	-0.110	-0.026	0.002	0.046	0.379**
Ptil	0.006	0.121	0.188	0.097	-0.063	-0.045	-0.007	0.007	0.019	0.324**
CD	-0.019	0.152	0.048	0.378	0.004	-0.034	-0.001	-0.004	0.020	0.544**
IntL	0.019	0.162	0.069	-0.009	-0.170	-0.100	-0.033	0.010	0.025	-0.026
RA	-0.005	-0.126	-0.033	-0.051	0.067	0.255	0.032	-0.004	-0.031	0.103
DL	0.013	0.114	0.019	0.004	-0.083	-0.120	-0.067	0.007	0.024	-0.068
DM	-0.046	-0.026	-0.060	0.073	0.077	0.041	0.019	-0.023	-0.019	-0.090
GY	0.006	0.136	0.036	0.078	-0.042	-0.081	-0.016	0.004	0.099	0.219**

Residual effect = 0.759

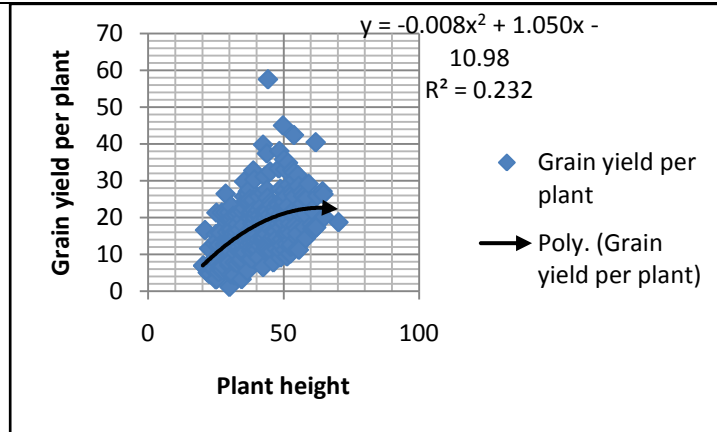


Figure 1: The polynomial regression of plant height with grain yield per plant
(Note: The grain yield reaches a static beyond 60cm height)

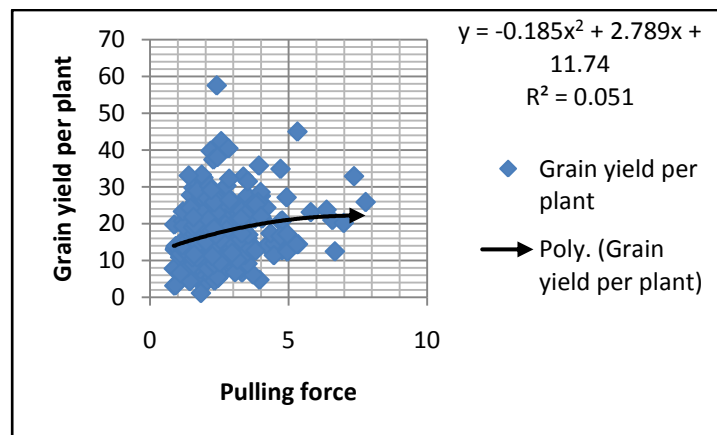


Figure 2: The polynomial regression of pulling force on grain yield per plant (Note: Beyond a pulling force of 6 newton there is no corresponding increase in grain yield)

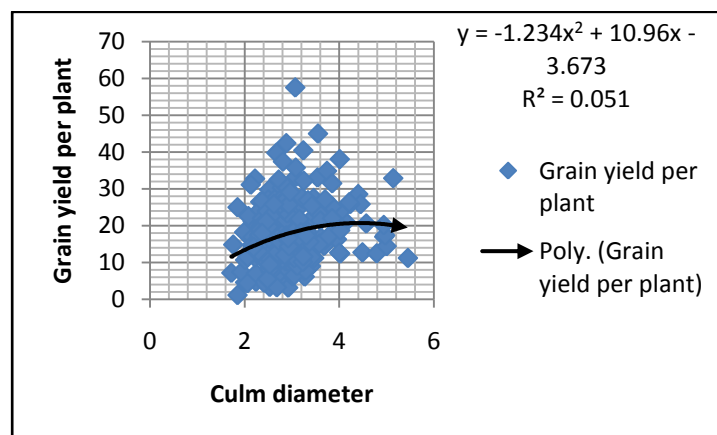


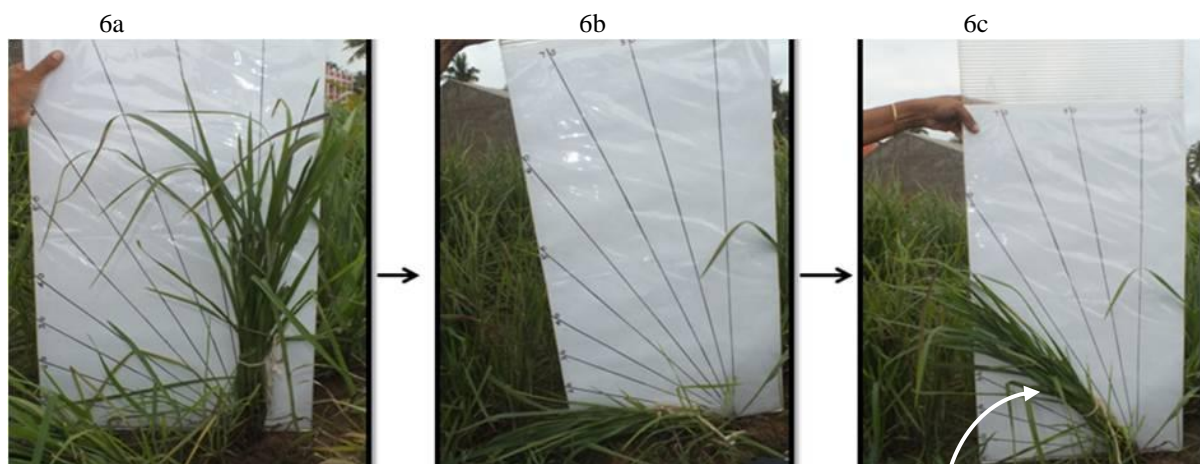
Figure 3: The polynomial regression of culm diameter with grain yield per plant (Note: Culm diameter more than 4mm has no positive effect on grain yield per plant)



Figure 4. Culm diameter measurement using vernier caliper



Figure 5. Pulling force measured force gauge.



**Figure 6. Measurement of recovery angle after bending,
6a: Plant at normal position, 6b: Plant bend to the ground level gently by holding the tip of the plant, 6c: The plant slowly released. Now the angle made by the plant from the ground level is measured.**

Genetic Analysis of Yield and Culm Strength Related Traits in Kodo millet (*Paspalum scrobiculatum* L.)

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ABSTRACT

Forty one Kodo millet germplasm accessions were evaluated to assess the magnitude of genetic variability, heritability and genetic advance for yield and culm strength related traits. The estimates of GCV and PCV were high for pulling force, culm weight per unit length, and grain yield per plant suggesting the presence of sufficient variability for these traits. Inter-node length was highly influenced by environment as evidenced by low level of heritability. High heritable variation was recorded for culm weight per unit length, grain yield per plant, days to 50 per cent flowering, culm diameter, pulling force and days to maturity indicating least influence of environment for the expression of these traits. High heritability along with high genetic advance was recorded for culm weight per unit length, grain yield per plant, pulling force and culm diameter. This indicated that these traits were governed by additive gene action. Hence selection based on these traits would be effective for improving yield and culm related attributes in kodo millet.

Key words: *Culm strength, yield, variability, heritability, genetic advance.*

Kodo millet (*Paspalum scrobiculatum* L.) is small seeded cereal crop, mainly grown in hilly and semi-arid regions in India and provides food security to farmers in these areas. It is mainly grown for its grain and fodder. In India, the area under millets, especially kodo millet has been declining during post green revolution period because of importance given to other major cereals. But, kodo millet could not be completely replaced by other remunerative crops as it still provides food security in areas where other crops fail to survive (Yadava and Jain, 2006). The seeds have an excellent storage life and can be stored for several years (Hegde and Gowda, 1989). Apart from being a staple food for the poor in the marginal agricultural areas, it is gaining attention as health food as it has more dietary fibre and has anti-oxidative (Chandrasekara and Shahidi, 2010) and anti-diabetic properties (Hegde, *et al.*, 2005). In this present

context, the genetic improvement of yield of kodo millet is imperative. Since the presence of variability is the input for any genetic gain, this study attempts to study the variability present in the germplasm available and to understand the genetics behind the traits for further improvement through selection.

MATERIALS AND METHODS

Experimental site and design

The material for this study included 41 genotypes consisted of germplasm accessions received from National Bureau Plant Genetic Resources, Regional Station, Hyderabad, land races collected from different parts of Tamil Nadu and germplasm accessions available at Millet Breeding Station, Tamil Nadu Agricultural University, Coimbatore and a released variety CO 3. The experiment was carried out at Millet Breeding Station, Centre for Plant Breeding and Genetics (CPBG), Tamil Nadu Agricultural University (TNAU), Coimbatore, during Summer, 2013 (Jan to May). The station is situated at 11°N latitude and 77°E longitude with an elevation of 411.98m above mean sea level, with clay loam soil type and pH around 8.5. The average rain fall at this centre is 730mm per year.

The experiment was laid out in a randomized block design (RBD) with two replications. Each genotype was raised in five rows per replication with a spacing of 22.5 x 10 cm. All the other recommended agronomic practices were followed to ensure a uniform crop stand. Observations on 14 quantitative traits *viz.*, days to 50 per cent flowering, plant height, number of productive tillers, inflorescence length, number of racemes, raceme length, culm diameter (taken at middle of the second inter-node from the base of the plant on the main tiller and measured using Vernier caliper), pulling force (force required to bend the whole plant to ground level, measured using force gauge in Newton), culm weight per unit length (dry weight of the second inter-node from base of the plant on the main tiller divided by its length) recovery angle after bending (the angle made

Table 1. Analysis of variance for 14 quantitative traits in Kodo millet.

Source of variation	Degrees of freedom	Mean sum of squares													
		DF	PH	ProT	InfloL	RN	RL	CD	PF	WL	IntL	RA	DM	GY	TW
Replication	1	10.98	1.48	0.88	0.48	0.20	7.22**	0.05	0.00	7.69**	4.64	34.70	0.05	49.83**	3.11**
Treatment	40	303.76*	158.54*	1.71	6.41**	0.47**	2.40**	1.53**	4.31**	72.85**	5.75*	195.16*	241.76*	69.83**	0.56**
Error	40	34.68	68.47	1.32	2.07	0.21	0.78	0.20	0.58	1.83	3.19	71.82	48.32	5.86	0.23

*significant at 5% level; **significant at 1% level

Abbreviations used: DF-days to 50 per cent flowering; PH- Plant height; ProT- number of productive tillers; InfloL- Inflorescence length; RN- number of racemes; RL- raceme length; CD- Culm diameter; PF- Pulling force; WL- culm weight per unit length; IntL- Inter-node length; RA- Recovery ability after bending; DM- Days to Maturity; GY- grain yield per plant; TW-test Weight.

by the whole plant against the ground after bending the plant to the ground level), inter-node length (the length of the second inter-node from the base of the plant on the main tiller), test weight (weight of random samples of 1000 grains from the total harvest of an accession), grain yield per plant, days to maturity (number of days taken from sowing to physiological maturity of 50 per cent of the main tillers) were recorded in five random plants in each genotype and the mean data was used for statistical analysis.

Statistical analysis

The data collected were subjected to analysis of variance (ANOVA) as suggested by Panse and Sukhatme (1967). The phenotypic and genotypic variances were calculated by utilizing the respective mean sum of square from variance table (Lush, 1940). Phenotypic and Genotypic coefficient of variation were estimated using the formula suggested by Burton (1952) and categorized into three range of variation such as low (<15%), medium (15-30%) and high (>30%) as proposed by Sivasubramanian and Madhava Menon (1973). Heritability in broad sense (h^2) was calculated according to Lush (1940) and expressed in percentage. The range of heritability was categorized as low (0-30), medium (31-60) and high (> 60) as suggested by Johnson *et al.* (1955). Genetic advance was worked out based on the formula suggested by Johnson *et al.*, 1955) and traits were classified as having high (>20%), moderate (10- 20%) or low (<10%) genetic advance as per the method suggested by Johnson, *et al.*, 1955.

RESULTS AND DISCUSSION

Analysis of variance

The result of ANOVA for the 14 quantitative characters studied is presented in Table 1. Mean square

of all the characters showed significant difference ($P < 0.05$) except for number of productive tillers per plant indicating the presence of variability for all the other traits which can be exploited through selection.

Variance components

The extent of variability in respect of range, phenotypic and genotypic coefficients of variation, is furnished in Table 2. In the present study, the traits, days to 50 per cent flowering, culm diameter, culm weight per unit length and days to maturity showed narrow differences between phenotypic and genotypic coefficients of variation indicating low effect of environment and greater role of genetic factors on the expression of these traits. High Genotypic Co-efficient of Variation (GCV) and Phenotypic Co-efficient of Variation (PCV) estimates were recorded for pulling force (38.18% and 43.74% respectively), culm weight per unit length (52.46% and 53.79% respectively) and grain yield per plant (39.29% and 42.74% respectively). Hence, presence of wide variability ensures more scope for improvement in these traits. Amawate and Dabral, 1970, Choudhary and Singh, 1970 and Rao, 1991 also observed wide range of variability for morphological characters and grain yield in land races of Madhya Pradesh. Low GCV and PCV estimates were recorded for inflorescence length (10.11% and 14.12% respectively), days to maturity (7.93% and 9.71% respectively) and test weight (8.99% and 13.80% respectively) suggesting low amount of variation for these traits. Similarly, Verma and Satpute (1969) and Ahluwalia *et al.* (1970) also reported high magnitude of PCV for grain yield per plant and low PCV for developmental traits like days to flowering and days to maturity in kodo millet. Subramanian *et al.*, 2010 also reported high GCV and PCV for seed yield per plant and low GCV and PCV for test weight in kodo millet.

Table 2. Genetic parameters for 13 quantitative traits in kodo millet genotypes

Sl. No	Traits	Range	Mean±SE	GCV %	PCV %	ECV %	Heritability %	GA	GA(% of mean)
1	Days to flowering (days)	62.00 -101.50	83.15 ±1.92	13.95	15.65	7.08	79.51	21.31	25.62
2	Plant height (cm)	37.00-74.00	52.25 ±1.39	12.84	20.39	15.84	39.68	8.71	16.66
3	Inflorescence length (cm)	11.50-19.17	14.58 ±0.28	10.11	14.12	9.87	51.21	2.17	14.90
4	Raceme length (cm)	6.25-11.17	7.80 ±0.17	11.54	16.15	11.30	51.04	1.32	16.98
5	Number of racemes (count)	2.00-4.33	3.05 ±0.08	11.94	19.17	15.00	38.80	0.47	15.32
6	Culm diameter (mm)	2.04-5.58	3.66 ±0.14	22.30	25.40	12.16	77.08	1.48	40.33
7	Pulling force (Newton)	1.24-6.41	3.58 ±0.23	38.18	43.74	21.33	76.21	2.46	68.67
8	Culm weight per unit length (mg per cm of dry culm)	5.02-32.70	11.36 ±0.94	52.46	53.79	11.90	95.10	11.97	105.38
9	Internode length (cm)	8.75-16.17	11.13 ±0.26	10.16	19.00	16.05	28.58	1.24	11.18
10	Recovery angle after bending (degrees)	23.33-68.34	45.43 ±1.54	17.29	25.43	18.66	46.20	11.00	24.20
11	Days to maturity (days)	109.00-145.00	124.07 ±1.72	7.93	9.71	5.60	66.68	16.54	13.33
12	Grain yield per plant (g)	4.49-26.56	14.39 ±0.92	39.29	42.74	16.82	84.51	10.71	74.40
13	Test weight (g)	3.63-5.53	4.54 ±0.08	8.99	13.80	10.46	42.49	0.55	12.08

Heritability in broad sense and genetic advance

In plant breeding, the magnitude of heritability of the trait of interest determines the success of the breeding program as it has a close bearing on the response to selection. The genotypes under study showed low level of heritability for the trait, inter-node length (28.58%) suggesting higher influence of environment for the expression of this trait. The highest heritability was recorded for culm weight per unit length (95.10%) followed by grain yield per plant (84.51%), days to 50 per cent flowering (79.51%), culm diameter (77.08%), pulling force (76.21%) and days to maturity (66.68%) indicating high influence of genetic components for these traits. The remaining traits, plant height, inflorescence length, raceme length, raceme number, inter-node length, recovery ability after bending and test weight, exhibited moderate level of heritability (Table 2). Similar reports of high level heritable variation are available for days to flowering (Verma and Satpute, 1969 and Ahluwalia, *et al.*, 1970) and low level of heritability for panicle length (Dhagat, *et al.*, 1971) in kodo millet. Dhagat, 1978, Rao and Tiwari, 1988 and Kandaswamy, *et al.*, 1990 reported high heritability for grain yield and low heritability for plant height and days to flowering.

Heritability estimates in conjunction with the predicted genetic advance will be more reliable (Johnson, *et al.*, 1955) in formulating suitable selection procedures. In the present study, the maximum genetic

advance was recorded for culm weight per unit length (105.38%), followed by grain yield per plant (74.40%), pulling force (68.67%) and culm diameter (40.33%) (Table 2). These four traits also registered high heritability, which indicated that these traits were governed predominantly by additive genes and selection could be effective for improving them. Hence there is more scope for improvement in yield and culm attributes from the genotypes studied. Subramanian *et al.*, 2010 also observed high heritability coupled with high genetic advance for the trait, seed yield per plant in kodo millet genotypes studied by them.

High level of genotypic and phenotypic variation for traits such as pulling force, culm weight per unit length and grain yield per plant suggests that there is more scope for improving the culm strength as well as yield. The trait internode length cannot be relied upon as it is highly influenced by environment as evidenced by a low value for heritability. High heritability for traits such as, days to 50 per cent flowering, culm diameter, culm weight per unit length, grain yield per plant, pulling force and days to maturity shows that these traits are least affected by environment and hence more reliable for selection and further genetic improvement of the respective traits. Out of these six traits, four traits *viz.*, culm weight per unit length, grain yield per plant, pulling force and culm diameter exhibited high heritability along with high genetic advance as well, which indicated predominance of additive gene action for the expression of these four traits and selection would be effective in improving them.

ACKNOWLEDGEMENT

Funding by University Grants Commission, New Delhi, to undertake the above research is gratefully acknowledged.

LITERATURE CITED

- Ahluwalia, M., Singh, Diljit, Solaman, C. and Naqvi, S. 1970. Studies on variability in kodo millet (*Paspalum scrobiculatum* L.). *The Andhra Agricultural Journal*. **17**(2):58-60.
- Amawate, J.S. and Dabral, K. C. 1970. Variability in kodo millet (*Paspalum scrobiculatum* L.). *Indian Journal of Agricultural Sciences*. **40**(7): 616-9.
- Burton, G.W. 1952. Quantitative inheritance in grasses. *Proc. 6th International Grassland Congress*, **1**: 277-283.
- Chandrasekara, A. and Shahidi, F. 2010. Content of insoluble bound phenolics in millets and their contribution to antioxidant capacity. *Journal Agricultural and Food Chemistry*. **58**:6706–14.
- Choudhary, L.B. and Singh, Rameshwar. 1970. Variety x environment interaction in kodo millet under two levels of fertility. *Indian Journal Agricultural Sciences*. **40**(11):977-83.
- Dhagat NK , Joshi RC, and Sharma D. 1971. Correlation and heritability in kodo millet (*Paspalum scrobiculatum* L.). *Indian Journal Agricultural Sciences*. **41**(8):682-4.
- Dhagat, N.K. 1978. Variability in inter-relationship and genetic divergence in *Paspalum scrobiculatum* (L.). Ph.D. thesis, Jawaharlal Nehru Krishi Vishwa Vidyalaya (JNKVV), Jabalpur, Madhya Pradesh, India.
- Hegde, P.S., Rajasekaran, N.S. and Chandra, T.S. 2005. Effects of the antioxidant properties of millet species on oxidative stress and glycemic status in alloxan-induced rats. *Nutrition Research*. **25**: 1109–1120.
- Hegde, B.R. and Linge Gowda, B.K. 1989. Cropping systems and production technology for small millets in India. In: *Small Millets in Global Agriculture* ((eds. Seetharam, A., Riley, K.W. and Harinarayana, G.) India: IDRC, Oxford & IBH Co.
- Johnson, H.W., Robinson, H.F. and Comstock, R.E. 1955. Estimate of genetic and environmental variability in soybeans. *Agronomy Journal*. **47**:314-318.
- Kandaswamy, G, Ramamoorthy N and Manoharan V 1990. Genetic variability in kodo millet (*Paspalum scrobiculatum* L.). *Madras Agricultural Journal*. **77**:9-12.
- Lush, J.L. 1940. Inter size correlation and regression of offspring on dams as a method of estimating heritability of characters. In : *Proceedings of American Society for Animal Production*., **33**: 293-301.
- Panse, V.G. and Sukhatme, P.V. 1967. *Statistical Methods for Agricultural Workers*. (2nd Edn.), ICAR Publication, New Delhi. pp. 152-161.
- Rao, S. S. 1991. Genetic variability in minor millets. *Indian Journal Agricultural Sciences*. **61**(5):322-23.
- Rao S.S and Tiwari R.D 1988. Variability and correlation studies in kodo millet . *Millets Newsletter*. **7**:53-4.
- Sivasubramanian, S. and P. Madhavamenon. 1973. Genetic analysis of quantitative characters in rice through diallel crosses. *Madras Agricultural Journal*. **60**: 1097-1102.
- Subramanian, A., Nirmalakumari, A. and Veerabhadhiran, P. 2010. Trait based selection of superior Kodo millet (*Paspalum scrobiculatum* L.) genotypes. *Electronic Journal of Plant Breeding*, **1**(4): 852-855.
- Verma, S.N.P and Salpute R.G. 1969. Heritability studies in kodo millet (*Paspalum scrobiculatum* L.). *JNKVV Research Journal*. **3**(2):77-9
- Yadava, H.S., Jain, A.K., 2006. *Advances in kodo millet research*. New Delhi, Directorate of Information and Publications of Agriculture, Indian Council of Agricultural Research.

Received on 11-07-2014

Accepted on 16-07-2014