ISSN 0973 - 354X

JOURNAL OF HORTICULTURAL SCIENCES

Volume 5 December 2010 Number 2

Society for Promotion of Horticulture
Indian Institute of Horticultural Research, Bangalore 560 089

JOURNAL OF HORTICULTURAL SCIENCES

(Founded in 2005 by the Society for Promotion of Horticulture, Bangalore, India)

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This Journal is abstracted in CABI, Current Contents, AGRIS and Indian Science Abstracts

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JOURNAL OF HORTICULTURAL SCIENCES

Volume 5 **Number 2** **December 2010**

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J. Hortl. Sci.
Vol. 5 (2): 85-93, 2010

FOCUS

Statistical genomics and bioinformatics

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ABSTRACT

Some important and interesting topics in the newly emerging disciplines of *Statistical genomics* and *bioinformatics* have been discussed briefly in relation to plants with possible references to fruit crops. This paper is therefore divided into two parts relating to the two disciplines, respectively. In the first part, mapping of quantitative trait loci (QTL), association mapping, mapping of gene expression transcripts (eQTL), marker-assisted selection, and a systems approach to quantitative genetics have been dealt with. In the second part, generation of databases, annotation, annotated sequence databases, and sequence similarity search have been described.

Key words: Statistical genomics, bioinformatics, fruit crops, eQTL, annotated sequence databases, sequence similarity search

I. STATISTICAL GENOMICS

INTRODUCTION

Most of the traits of economic importance in plants have an underlying genetic basis involving several genes, and, are subject to modification by environmental factors. Statistical considerations have been predominant in dissecting such complex traits into estimable components (Narain, 1990). Heritability of a trait, as a proportion of the phenotypic variation that is attributed to genetic causes, has been a prime indicator helpful in taking decisions for genetic improvement of economic traits. Prediction of response to artificial selection (based on intensity and accuracy of selection) and the existence of genetic variability have been successful across several crop plants. However, relationship between the phenotype and the genotype has been like a black box where inferential approach has been the only way to look into it. This scenario is now changing with advent of the modern technologies of gene sequencing, microarray experiments and the enormous advances made in attempts to understand gene and protein expression within the cell of an organism. In this context, information on molecular markers has been extremely helpful in identifying regions on chromosomes (QTL) that bring about variation in a trait, thereby providing tools that can lead to far more accurate selection procedures for genetic improvement. Saturated genetic maps of markers, giving their order along a chromosome and relative distances between them, have been developed. Gene transcript data from microarray experiments can be integrated with molecular marker information to map expression traits (eQTL) that can possibly lead to causal networks. The network approach connecting data on genes, transcripts, proteins, metabolites, etc. indicates emergence of a systems quantitative genetics (Narain, 2009, 2010).

Mapping of Quantitative Trait Loci (QTL)

Genomic techniques like *restriction fragment length polymorphism* (RFLP), *random amplified polymorphic DNA* (RAPD), *amplified fragment length polymorphism* (AFLP), *variable number of tandem repeats* (VNTR) - that consist of micro satellites (short sequences) termed as *short tandem repeats* (STR) or *simple sequence repeats* (SSR) and mini satellites (long sequences) - and *single nucleotide polymorphisms* (SNP) have been developed that help in identification of QTLs by correlation between a trait and its specific DNA markers (Narain, 2000). The first problem is, therefore, to construct a linkage map that indicates the position and relative genetic distances between markers along the chromosomes. Map distance is based on the total number of cross-overs between the two markers, whereas, physical distance between them is denoted in terms of nucleotide base pairs (bp). A centi-Morgan (cM), corresponding to a cross-over of 1%, may span 10 kbs to 1,000 kbs and can vary across species.

Since marker genotypes can be followed for their inheritance through generations, these can serve as molecular tags for following the QTL, provided they are linked to the QTL. This requires detecting the marker-QTL linkage and, if established, estimating the QTL map position on the chromosome. However, these problems depend on whether we have data on experimental populations obtained from controlled crosses, as in plants, or on natural populations like humans where controlled crosses cannot be made.

The most popular method, given by Lander and Botstein (1989), is that of s*imple interval mapping* (SIM). It involves formation of intervals by pairing of adjacent markers and treating them as a single unit of analysis for detection and estimation purposes. It is based on joint frequencies of a pair of adjacent markers and a putative QTL flanked by the two markers. Suppose markers *A* and *B* are linked with recombination fraction *r* and QTL *Q* is located between them with r_1 recombination from *A* and r_2 from *B*. Then, $r = r_1+r_2-2r_1r_2 \cong r_1+r_2$, on the assumption of no interference and *r* so small that no double cross-overs can be assumed. In the classical back-cross design with three loci each with two alleles, *A-a*, *B-b*, and *Q-q*, the expected frequencies for the eight marker-QTL genotypes can be used to obtain conditional probabilities of the QTL genotypes, given the marker genotypes. By setting up a linear regression model between the trait (Y) and the indicator variable (X) taking the value 1 if the QTL is *QQ* and –1 if it is *Qq*, one can estimate a regression coefficient that defines the allelic substitution effect of this QTL. In such a model, the QTL genotype for a given individual is unknown. X is then a *random* indicator variable with conditional probabilities of obtaining *QQ* or *Qq* at the QTL. This means the observed value is modelled as a mixture-distribution with mixture ratios as the *conditional* probabilities. We have, therefore, a situation often referred to as a linear regression with *missing* data. The problem of estimation then involves the use of EM algorithm. By assuming that the character is normally distributed within each of the eight marker-QTL classes with equal variance σ^2, one can set up a likelihood function in terms of unknown parameters, and develop a log likelihood ratio (Λ) for testing the hypothesis that the QTL is not located in the interval where the log likelihoods are evaluated using the maximum likelihood estimates of the genotypic values for the two QTL genotypes, the variance σ^2 and the recombination fraction r_1 between marker *A* and the putative QTL using iterative procedures based on EM algorithm. This statistic is distributed as χ^2 with 1 d.f. The associated lod score for the interval mapping is then (½) ($\log_{10}$e) Λ. This statistic is evaluated at regularly-spaced points; say 1 or 2 cm distance, covering the interval as a function of the presumed QTL position. Repeating this procedure for each interval along the chromosome and plotting the lod score curve against the interval gives a *QTL likelihood map* that presents evidence for the QTL at any position in the genome. Presence of a putative QTL is assumed if lod score exceeds a certain threshold *T* and the maximum of the lod score function in the map gives an estimate of the QTL position and gene effects. Mapping of QTL by interval method is widely used in practice. Analysis is done through the software MAPMAKER/QTL.

Although SIM is the method for QTL mapping most widely used with advantage in several practical situations, it ignores the fact that most quantitative traits are influenced by numerous QTLs. This is overcome either by adopting a model of *Multiple QTL Mapping* (MQM) or by combining SIM with the method of multiple linear regression, a procedure known as *composite interval mapping* (CIM). In all these methods, one uses the approach of maximum likelihood that produces only point estimates of the parameters such as the number of QTLs, their location, and effects. The corresponding confidence intervals are required to be determined separately by re-sampling methods. Further, the correct number of QTLs is difficult to determine using traditional methods. Their incorrect specification leads to distortion of the estimates of locations and effects of QTLs. To address these problems, a *Bayesian* approach is often adopted wherein the joint posterior distribution of all the unknown parameters given their *prior* distributions and the observed data is computed. For details of these various aspects, one can refer Narain (2003a, 2005).

The first application of interval mapping in plant breeding was to an inter-specific backcross in tomato. The parents for the back-cross were the domestic tomato *Lycopersicon esculentum* (*E*) with fruit mass 65 g and a wild South American green-fruited tomoto *L. chmielewskii* (*CL*) with fruit mass 5 g. A total of 237 back-cross plants were assayed for continuously varying characters like fruit mass, soluble-solids concentration and pH, and, 63 RFLP and 20 isozyme markers spaced at approximately 20 cM intervals were selected for QTL mapping. A threshold *T*=2.4, giving a probability of under 5% that even a single false-positive will occur anywhere in the genome, was used. This corresponds approximately to significance level for any single test as 0.001. The resulting QTL likelihood maps revealed multiple QTLs for each trait (6 for fruit weight, 4

for concentration of soluble solids and 5 for fruit pH) and estimated their location to within 20-30 cm.

Fruit crops

Fruit crops differ from most of the agronomic/forest crops in that they have large plant size, long intergeneration period due to their extended juvenile phase, asexual propagation, high heterozygosity and polyploidy. These practice outcrossing and have a long life. They are mostly woody perennials and their products are usually perishable. The major temperate fruit crops belong to *Rosaceae* family. The most important genera of this family are *Prunus, Malus, Pyrus, Fragaria,* and *Rosa*. Important members of the genus *prunus* are peach, cherry, plum, apricot, almond and of the genus *Malus* is apple. They have been slow to respond to new technologies in breeding, until recently. Characters like yield, blooming, harvesting time and fruit quality have been studied with the help of molecular markers in several fruit crops. Long period from seed to fruiting in such crops is a major problem in breeding studies involving crosses. Vegetative reproduction, on the other hand, allows every population to be immortalized and one can study a given character for as many years and in as many different environments as one wants. Interspecies crosses are possible and most of them have small genomes. For instance peach, the best characterized among *Prunus* species, has a haploid genome size of 164 Mbp only. Most of the *Prunus* species are diploid, with 8 pairs of chromosomes whereas, apple and pear are allotetraploid with 17 pairs of chromosomes.

Saturated linkage maps with transferable markers, RFLPs, and microsatellites have been developed to provide basic tools for studies on QTLs and marker-assisted selection in fruit tree breeding. As a result of a European project, a saturated linkage map of 246 markers (235 RFLPs and 11 isozymes) constructed from an F_2 progeny derived from almond (cv. Texas) x peach (cv. Earlygold) cross – termed TxE map- indicated 8 linkage groups (G1 to G8) with a total distance of 491 cm. This led to a *Prunus* reference map with 652 markers and a further set of 13 maps constructed with a sub-set of these markers has enabled genome comparisons among seven *Prunus* diploid species (almond, peach, apricot, cherry, *Prunus ferganensis, Prunus davidiana,* and *Prunus cerasifera*). These have helped establish the position of 28 major genes affecting various agronomic characters in different species of *Prunus* crops (Dirlewanger *et al*., 2004).

The first linkage map in apples was constructed by a European Consortium based on F_1 progeny derived from the cross cv. Prima x cv. Fiesta (FxF map). There were a total of 290 markers consisting of RFLPs, SSRs, isozymes, RAPD etc., distributed over 17 linkage groups. A more saturated map was constructed with the F_1 progeny derived from the cross cv. Fiesta x cv. Discovery (FxD map) using 840 markers that included 129 SSRs. These maps have been helpful in QTL studies on apple. A comparison between apple and *Prunus* maps suggests a high degree of synteny between these two genera. QTLs for blooming, ripening and fruit quality have been found in peach and apple. Some of these QTLs were found to be located in regions of the genome where major genes were earlier mapped. For instance, in peach a major gene responsible for low fruit acidity was in the same region as QTLs affecting fruit quality, a quantitative trait. In apple too, a major gene coding for malic acid content is located in the same region as QTLs for fruit quality.

Various populations of peach x *Prunus davidiana* crosses with different levels of introgression of the *Prunus davidiana* genome into the cultivated peach viz. F_1, F_2 or BC2 were used to discover the positions of respective QTLs. About 13 QTLs explained up to 65% of the total phenotypic variation for powdery mildew resistance in plants exposed to the disease in different times and environments.

Candidate gene approaches have been adopted for finding associations between genes involved in relevant metabolic pathways and major genes or QTLs in fruit trees. Several resistance gene analogs (RGAs) were mapped in *Prunus* that are at similar genomic positions as genes or QTLs which determine ‘sharka‘ resistance in apricot or root-knot nematode resistance in peach and plum.

Linkage Disequilibrium or Association Mapping

The mapping of QTLs in plants based on data collected from pedigrees of populations formed by crossing inbred lines is on a coarser scale, so that a QTL detected is likely to refer to several genes in a chromosomal region. The approach of population-based association mapping that involves linkage disequilibrium (LD) between markers and the genes underlying complex traits leads, on the other hand, to more accurate mapping of genes. The key idea is that a disease mutation assumed to have arisen once on the ancestral haplotype of a single chromosome in past history of the population of interest is passed on from generation to generation, together with markers at tightly linked loci, resulting in LD. The use of this approach in horticultural crops, though widely prevalent in human genetics, is limited.

Advantages of the two approaches can be combined by detecting QTL initially using linkage mapping with moderate number of markers, followed by a second-stage of high-resolution association mapping in QTL regions that capitalizes on a high-density marker map.

Benefits of linkage and association mapping have recently been combined in a single population of maize by adopting a *nested association mapping* (NAM) approach. The maize NAM population was derived by crossing a common reference sequence strain to 25 different maize lines. Individuals resulting from each of the 25 crosses were self-fertilized for four further generations to produce 5,000 NAM recombinant inbred lines (RILs). This population was first used for initial detection of QTL using the linkage mapping approach. Subsequently, within each diverse strain, high-resolution association mapping was adopted with a high-density marker map. It is significant to note that within each RIL, all individuals are genetically nearly identical. This means we can estimate true breeding value of each line far more accurately by averaging phenotypic measurements of a given trait taken on several individuals with the same genotype.

In a recent experiment, genetic architecture of flowering time in *Zea mays* (maize) was dissected using NAM. About 1 million plants were assayed in eight environments to map the QTLs. About 29 to 56 QTLs were found to affect flowering time. These were small-effect QTLs shared among the diverse families. The analysis showed, surprisingly, absence of any single large-effect QTL. Moreover, no evidence was found of epistasis or environmental interactions. Flowering time controls adaptation of plants to their local environment in an out-crossing species like *Zea mays*. A simple, additive genetic model predicting accurately flowering time in this species is, thus, in sharp contrast to that observed in several plant species which practice self-fertilization (Buckler *et al*., 2009).

Mapping QTLs for Gene Expression profile (eQTL)

The advent of DNA chip technology in the form of cDNA and oligonucleotide microarrays has provided huge and complex data-sets on gene expression profiles of different cell lines from various organisms. Such gene expression profiles have recently been combined with linkage analysis, based on QTL mapping, through molecular markers in what has been termed 'genetical genomics' (Jansen and Nap, 2001). Gene expression, in terms of transcript levels, for each individual of a segregating population are phenotypes that are correlated with markers, genotyped for that individual, to identify QTLs and their location on the genome to which the expression trait is linked. Such expression quantitative trait loci (eQTL) studies are similar to traditional multi-trait QTL studies, but with thousands of phenotypes. It is also important to note that, underlying the gene expression differences, there are two types of regulatory sequence variation. One is *cis*-regulatory that affects its own expression and the other is *trans*-acting or protein coding that affects expression of other genes. The first attempt where transcript abundance was used to study the linkage with QTLs was on budding yeast (Brem *et al*, 2002) based on a cross between a laboratory strain and a wild strain, the parents being haploid derivatives. Heritability estimation was based on haploid segregants and the linkage with a marker was tested by partitioning the segregants into two groups, according to marker genotypes, and comparing expression levels between groups, with Wilcoxon-Mann-Whitney test. They found 8 trans-acting loci, each affecting expression of a group of 7 to 94 genes of related function. Since then, several eQTL studies have been published in species like mice, maize, humans, rats and *Arabidopsis thaliana*.

Apart from study of the eQTL in yeast, Foss *et al*. (2007) investigated protein QTL in the *same* population of the yeast using mass spectrometry. Comparison between genetic regulation of proteins and that of the transcripts revealed that loci that influenced protein abundance differed from those that influenced transcript levels, much against expectations.

Marker–Assisted Selection (MAS)

Molecular markers such as those provided by RFLP have not only made it possible to detect and estimate effects of QTLs, but can also be used as a criterion of indirect selection for genetic improvement of a given quantitative trait – a procedure of selection which has come to be known as *marker-assisted selection* (MAS). The underlying basis of MAS is the correlation between a trait and the marker genotype, which gets generated due to linkage disequilibria between the QTL and marker loci. The fact that such information can be integrated with those of artificial selection on individual and/or collateral basis, to increase the efficiency of selection, was demonstrated by the work of Lande and Thompson (1990). They showed that relative efficiency of the selection index, combining phenotypic and molecular information optimally, is a function of heritability (h^2) of the trait and the proportion (p) of additive genetic variance of

the trait that is associated with marker loci. This efficiency is always one when h^2=1, the phenotype being a perfect indicator of its breeding value. But, for a character with low heritability, the efficiency can be substantially high, provided *p* is high. This means the value of maker information can be very great if a larger proportion of additive genetic variance is associated with the markers. Efficiency is maximum when *p*=1 and is (1/*h*), that becomes infinitely large for extremely small *h*. In that case, all of the weight in selection index is put on molecular information. If we select *only* on the basis of marker information, the efficiency, relative to individual selection with the same intensity, would be. This shows that when $p>h^2$, selection based on marker information *alone* would be more efficient than individual phenotypic selection.

Increased efficiency of MAS, however, is accompanied by increased cost involved in sample collection, DNA extraction and typing of individuals in the sample, compared to that involved in taking simple measurements of the trait. Cost reduction for MAS can be achieved in several ways. Marker technologies such as those based on *polymerase chain reaction* (PCR) may reduce the cost of MAS. *Selective genotyping* of the extreme progeny, as advocated by Lander and Botstein (1989), is another way. Yet another way could be to bring in auxiliary information from other traits that are correlated with the main trait, and are cheaper to measure. This idea has been used in the past by several workers to increase the efficiency of individual and family selection itself, by including in the index one or more auxiliary traits in conjunction with the main trait. As a matter of fact, molecular information in MAS is itself a sort of auxiliary information, but obtained at a higher cost. Narain (2003b), therefore, showed how the efficiency of MAS behaved if information on one or more auxiliary traits with the corresponding molecular scores was combined with that on the main trait, in an optimal manner.

Fruit crops

In fruit crops, molecular markers are used for screening and selecting the best seedlings several years before the characters are evaluated in the field. It saves space and time so important in woody perennials. Marker-assisted selection in such crops is, however, mostly based on major genes, since several characters like disease resistance, flower/fruit/nut quality are found to be controlled by major genes that follow a simple inheritance pattern. Markers tightly linked to such genes are searched for early selection. They are primarily used for characters that cannot be evaluated till the plant has reached the adult stage, such as fruit characters or self-incompatible genotypes. For instance, gametophytic self-incompatibility in almond, apricot and cherry is one such trait that is encoded by a highly polymorphic locus (S/s) located in the distal part of G6 linkage group. With determination of the sequences of the polymorphic S-RNase gene at this locus, a number of species-specific and allele-specific DNA markers were discovered that were used for early and more accurate selection of self-incompatibility or self-compatibility alleles. Markers close to the two genes of resistance to root-knot nematodes are used for selection of resistant *Prunus* rootstocks. The resistance gene *Ma/ma* from Myrobalan plum and located on G7 linkage group, and another one from peach cv. Nemared (*Mi/mi*) located on G2 linkage group, have been screened with markers in a search for rootstocks that pyramid both resistance genes in a three-way progeny obtained from peach, almond and Myrobalan plum.

Marker-assisted selection for disease resistance is quite widespread in apple as a means of early selection, and, to pyramid resistance genes.

Systems approach

As we know, the central dogma of molecular biology stipulates that sequence information flows from DNA to RNA to protein but not in the reverse direction. But, Kimchi-Sarfaty *et al* (2007) reported data that indicate that a protein's three-dimensional structure is *not* necessarily determined by its amino acid sequence that has been specified by the DNA sequence. An mRNA, if subjected to translational braking, can generate a protein with a structure different from that specified by the DNA sequence. This has been termed 'translation-dependent folding' (TDF) hypothesis (Newman and Bhat, 2007). Differential gene expression resulting in transcripts as sub-phenotypes could, then, lead to different proteins and give results similar to those obtained in the yeast experiment, as reported by Foss *et al* (2007). Genes and proteins are, therefore, required to be considered simultaneously to unravel the complex molecular circuitry operating within a cell. One has to have a global perspective of genotype-phenotype relationship, instead of individual components like DNA or protein in a cellular system.

It seems the interplay of genotype-phenotype relationship for quantitative variation is not only complex but also needs a closer look at how we view this relationship – whether purely at the DNA-RNA level (as in the reductionist approach) or at the level of cell as a whole

(where DNA-RNA are just parts of the cellular system with other contextual forces present in the micro-environments of the cell, also playing their own important roles). Such situations have also been noticed in agricultural experimentation where a dialectical approach has been advocated (Narain, 2006, 2008). In the grain production process, it is also important to study how this process affects soil health and the ecosystem surrounding the plant, as is studying the effect of inputs on production. In the dialectical approach, this relationship between the plant and its environment is studied both ways – input to output as well as output to input, a sort of feedback. A similar possibility seems to exist in the genotype and phenotype relationship within a cell. The protein as a phenotype is determined by a DNA sequence as the genotype, but the reverse phenomenon of protein affecting the DNA could also take place at the expense of violating central dogma. In fact, studies are on to explore biochemical signaling pathways that regulate function of living cells through regulatory networks having positive and negative feedback loops, though it is unclear how genetics can be incorporated into it. These feedback loops are basically *cybernetic* concepts that are inherent in the dialectical approach. This approach takes into account dynamics of the system over time as well, in which, development is a consequence of opposing forces. This is based on the concept of *contradiction* inherent in the meaning of *dialectics*. Things change because of the action of opposing forces on them, and things remain what they are because of temporary balance of the opposing forces. Opposing forces are seen as contradictory in the sense that each taken separately would have an opposite effect, but their joint action may be different from result of either acting alone. These forces are, however, part of self-regulation and development of the object is regarded as a network of positive and negative feedback loops, incorporation of which (in the genetic context) would violate the central dogma. Genes, transcripts, proteins, metabolites, physical components, etc., can be regarded as 'parts' of the cellular system and the 'whole' is regarded as a relation of these parts that acquire properties by virtue of being parts of a particular whole. As soon as the parts acquire properties by being together, they impart to the whole new properties that are, in turn, reflected in changes in the parts, and so on. Parts and whole, therefore, evolve as a consequence of their relationship, and the relationship itself evolves. Genes are fixed, but their expression-the transcript-is not. At any given moment of time, genes are expressed as per requirement of the cell and through information contained in its DNA. At this moment of time, the cellular system is said to have a particular *state* of the system. At the next moment of time, the same genes may be expressed, but differently, depending upon the then requirement of the cell and based on the feedback, if any, from the system's state at the previous time point, assuming that the process is Markovian. This gives the next state of the system, which might or might not be different from the previous state. And, the process goes on continually, modifying the relationship between different parts of the system based on interactions and feedbacks. It seems that a dialectical approach could provide the clue for understanding how 'parts' of a system and the 'whole' system behave in the context of genetics.

II. BIOINFORMATICS

INTRODUCTION

Genomic research is creating quantities of data at unprecedented scales by looking at either *all* genes in a genome, or *all* transcripts in a cell, or else *all* metabolic processes in a tissue in several species, in general, and in agriculture in particular. Very soon new genomic technologies will enable individual laboratories to generate terabyte or even petabyte scales of data. To handle these data, to make sense of them and render them accessible to biologists, is the task of a newly emerging field of *bioinformatics* existing at the interface of biological and computational sciences - computer based analysis of large biological data sets. The data sets usually pertain to macromolecular sequences (DNA, RNA and protein sequences), protein structures, gene expression profiles and biochemical pathways. It has three components. Firstly, it involves development of databases to store and search data. Secondly, it deals with statistical tools and algorithms to analyze and determine relationships between data sets. Lastly, it involves application of the tools for analysis and interpretation of various types of genomic data. For a brief discussion on these aspects, reference may be made to Narain (2005). Here, we discuss primarily those aspects that relate to plant genomes.

Generation of Databases

DNA sequences stored in databases are of three types: genomic DNA, cDNA and recombinant DNA. Genomic DNA, taken directly from the genome, contains genes in their natural state which, in eukaryotes, include introns, regulatory elements and a large amount of surrounding inter-genic DNA. cDNA is reverse-transcribed from mRNA and corresponds to only expressed parts of the genome, there being no introns. It gives direct access to genes that represent only a small percentage of the entire sequence. Recombinant DNA comes from the laboratory,

being artificial DNA molecules – sequence of vectors such as plasmids, modified viruses and other genetic elements used in the laboratory.

High quality sequence data is generated by performing multiple reads on both DNA strands. Sequence data of lower quality can, however, be generated by single reads – single pass sequencing on a much larger scale, quickly and cheaply. Expressed sequence tags (ESTs) are generated by single-pass sequencing of random clones from cDNA libraries and are used to identify genes in genomic DNA as well as to prepare large clone sets for DNA microarrays. Most RNA sequences are deduced from the corresponding DNA sequences, or, from a cDNA sequence. The latter is more informative due to it being extensively processed during synthesis. For example, introns are spliced out of a primary transcript to generate mature mRNA.

Plant sequence data are generated through (i) whole genome sequencing, (ii) sample sequencing of bacterial artificial chromosomes (BACs), (iii) genome survey sequencing (GSS), and (iv) sequencing of expressed sequence tags (ESTs). An integrated database and suite of analytical tools to organize and interpret these data, has been developed and is known as PlantGDB (*vide* the website http://www.plantgdb.org/).

Annotation

Annotation means obtaining useful biological information (structure and function of genes and other genetic elements) from raw sequence data. Since prokaryotes and eukaryotes differ in their structure and genome organization, their annotations involve different problems. Prokaryotes have high gene-density with virtually no introns, but in eukaryotes, gene-density is low and the genome has greater complexity.

We have two groups of annotation - structural annotation and functional annotation. In the former, we are concerned with finding genes and other genetic elements in genomic DNA. In the latter, we assign functions to the discovered sequences.

Annotated Sequence Databases

The following three repositories and resources for primary sequence data are available where each entry is extensively annotated. They can be accessed freely over the World Wide Web (www).

(i) Gene Bank of the National Centre for Biotechnology Information (NCBI)

(ii) Nucleotide Sequence Database of European Molecular Biology Laboratory (EMBL)

(iii) DNA Databank of Japan (DDBJ).

New sequences can be deposited in any of the databases, since, these exchange data on a daily basis. The main sequence databases have a number of subsidiaries for storage of particular types of sequence data. For example, dbEST is a division of Gen Bank which is used to store *expressed sequence tags* (ESTs). Other divisions of Gen Bank include dbGSS, dbSTS - used to store *sequence tagged sites* (STSs) - and several others.

These large database providers, however, do not give non-redundant and curated records, so that detailed analysis cannot be performed at the resource site by the user. A data- base like PlantGDB, which downloads raw plant genomic data from Gen Bank, overcomes such difficulties and provides curated records with detailed and updated information. It organizes EST sequences into contigs that represent tentative unique genes. They are duly annotated and linked to their respective genomic DNA. The data-base gives the basis for identifying genes common to particular species by integrating a number of bioinformatics tools that help in gene prediction and cross-species comparison - the goal of comparative genomics.

Besides PlantGDB database, there are species-specific databases like The *Arabidopsis* Information Resource (TAIR), MaizeGDB, Gramene, a tool for grass genomics, and the Stanford Microarray Database. The PlantGDB genome browsing capabilities for *Arabidopsis* are made possible by *A. thaliana* Genome Database (AtGDB; http://www.plantgdb.org/AtGDB/). This database stores EST and cDNA spliced alignments along with current *Arabidopsis* genome annotation.

As we know *Arabidopsis thaliana*, which is a small mustard species – *eukaryotic* and self-pollinating – is already playing an important role as a model organism in development of plant molecular biology, by way of providing increased knowledge and understanding of the plant's functional and developmental processes. It has a rapid life cycle and can be easily grown in laboratory in large numbers. Its entire genome, that is highly compact and consists of about 130 Mb with little interspersed repetitive DNA, has been sequenced. Many thousands of A*rabidopsis* plants can be grown on a bench to search for particular mutants which can then be isolated and genes cloned for use in other crops. It is related to many food plants like rice, wheat, maize, sorghum, millets, etc., and can, therefore, provide a

focus from which genome content of other higher plants can be extrapolated.

Fruit crops

In regard to horticultural crops, an international consortium led by Albert Abbott at Clemson University (Clemson, SC), developed databases on *Prunus* genome. Using RFLPs on the TxE map and a BAC library of peach cv. Nemared, a physical map was assembled. A growing collection of ESTs from peach and almond, based on cDNA libraries, was released to public databases and more than 3,800 peach putative unigenes were detected. About 2,000 of these unigenes were assigned to specific BAC that contain them. Recently, a Rosaceae database (www.genome.clemson.edu/gdr) has been developed that includes apple, peach, cherry, plum, apricot, pear, etc.

Sequence Similarity Searches

Due to molecular evolution, macromolecule sequences share a common ancestor resulting in similarity in their sequences, structure and biological functions. On the other hand, any pair of sequences will share a certain degree of similarity, due to chance alone. For example, DNA sequences are constructed from an alphabet of only four letters, viz., A, T G and C. Any sequence that consists of a mixture of these letters will show some similarity to any other similarly-constructed sequence. We need to distinguish between such a chance similarity and similarity resulting from real evolutionary and/or functional relationship. This requires use of appropriate statistical methods.

Sequences are first aligned in terms of their letters. When identical letters get aligned, we say that these letters were part of the ancestral sequence and have remained unchanged. When non-identical letters get aligned, we say that a mutation has occurred in one of the sequences. It may also happen that some letters in a particular sequence lack an equivalent in the other sequence, resulting in a gap. This could be due to insertion or deletion of letter/s in one of the sequences, with respect to the ancestral sequence. Dynamic programming algorithms – computational methods - can calculate the best alignment of two sequences. The algorithm takes two input sequences and produces the best alignment between them as the output. Well-known algorithms are Smith-Waterman algorithm (local alignment) and Needleman-Wunsch algorithm (global alignment).

To quantify similarity, a simple alignment score measures the number or proportion of identically matching residues. Gap penalties are subtracted from such scores to ensure that alignment algorithms produce biologically sensible alignments, without too many gaps. Gap penalties may be constant, i.e., independent of the length of the gap or be proportional to the length of the gap, or else may be affine, i.e., containing gap-opening and gap-extension contributions.

We have often a query sequence about which we need to predict the structure and/or the function. We perform sequence similarity searches of databases in which the query sequence is aligned (compared) to each database sequence in turn and then rank the database sequences with the highest scoring (most similar) at the top. This can be achieved by the dynamic programming method with Smith-Waterman algorithm but the procedure is very slow, taking hours, for searching large databases. On the other hand, algorithms like BLAST (Best Local Alignment Search Tool) and FASTA provide very fast (about five to fifty times faster) searches of sequence databases. They are however less accurate than the dynamic programming method which provides the best possible alignment to each database sequence. Each of the BLAST and FASTA operates by first locating short stretches of identically or near-identically matching letters (words) –assumed to lead to high scoring alignment - that are eventually extended into longer alignments.

Acknowledgements

This work was supported by the Indian National Science Academy, New Delhi, under their programme “INSA Honorary Scientist”.

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J. Hortl. Sci.
Vol. 5 (2): 94-108, 2010

FOCUS

Guava improvement in India and future needs

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ABSTRACT

Guava (*Psidium guajava* L; Myrtaceae) is an important fruit crop of India. High heterozygosity and frequent cross pollination resulted in the present day variability in seedling populations from which promising genotypes have been selected. As of now, there are about 160 cultivars available in India, among which 'Allahabad Safeda' and 'Sardar' varieties are widely cultivated. Crop improvement work attempted in India resulted in release of several superior selections / hybrids. Also, interspecific hybrids resistant to guava wilt were developed at CISH, Lucknow which are graft compatible with commercial varieties of *P. guajava*. The use of new biotechnological tools like DNA fingerprinting to study the extent of genetic variation among cultivars, rapid multiplication through in vitro shoot-tip culture needs to be employed extensively. Attempts need to be made to spot genetic markers for wilt resistance to improve efficiency in developing wilt resistant clones and rootstocks. Survey to identify superior genotypes with Allahabad Safeda traits and high density planting characters like early bearing, compact plant type, favourable response to pruning, good branch angle to minimize branch breakage even under heavy bearing, and, with a high fruit : shoot ratio need to be paid due attention. Work on aneuploidy breeding, development of autotetraploids and in vitro genetic manipulation of somatic cells needs to be intensified.

Key words: Guava, improvement, varieties/hybrids, *Psidium* sp.

INTRODUCTION

Guava (*Psidium guajava* L.) is an important fruit crop of India. It has gained considerable prominence on account of its high nutritive value, availability at moderate prices, pleasant aroma and good flavour. It is one of the commonest fruits liked by the rich and the poor alike and is popularly known as 'apple of the tropics'. It is one of the hardiest fruit trees, adaptable to a variety of soil and climatic conditions.

It grows well even under neglected conditions and, in fact, is even sometimes considered of a weed in Fiji and Hawaii. It is the fifth most widely grown fruit crop of India. The area under guava is about 0.178 million hectares, producing 1.83 MT of fruit. Popular varieties of guava in India are Allahabad Safeda, Lucknow-49, Nagpur Seedless, Dharwar, etc. Bihar is the leading state in guava production, with 0.26 MT, followed by Maharashtra, Uttar Pradesh, Karnataka, West Bengal, Punjab, Andhra Pradesh, Gujarat, Orissa and Tamil Nadu. At present, it is grown throughout the length and breadth of the country night from sea level to 1300m altitude, and is so acclimatized that it seems like a native of India. Guava is a rich source of vitamin C and pectin. Guava fruit contains 82.5% water, 2.45% acids, 4.45% reducing sugars, 5.23% non-reducing sugars, has 9.73 % TSS, 0.48% ash and 260 mg vitamin C/100g fruit (which differ with cultivar, stage of maturity and season). Guava fruit is relished when mature or ripe, or, when freshly plucked from the tree. It is also used in making many commercial products like jelly, fruit butter, juice, etc.

ORIGIN AND DISTRIBUTION

The guava is said to have originated in tropical America (Hayes, 1953). De Candolle (1904) stated that it originated in Mexico, while Purseglove (1968) opined that it originated in Brazil. It is widely distributed over equatorial regions growing in tropical and sub-tropical climates. It was introduced in to India during the 17th Century. In Spanish, the tree is known as *guayabo* or *guayavo*, the fruit *guayaba* or *guyava*. The French call it *goyave* or *goyavier*; the Dutch, *guyaba*, *goeajaaba*; the Surinamese, *guave* or *goejaba;* and the Portuguese, *goiaba* or *goaibeira.* Hawaiians call it guava or *kuawa*. In Guam, it is *abas*. In Malaya, it is generally known either as guava or

jambu batu, but has also numerous dialectal names as it does in India, tropical Africa and the Philippines where, the name *bayabas*, is often applied. Various tribal names – *pichi, posh, enandi, etc*., are employed among Indians of Mexico and Central & South America.

SPECIES STATUS

The genus *Psidium* belongs to the family Myrtaceae and has a basic chromosome number of x=11. All the cultivars of Indian guava belong to a single species, *Psidium guajava* L. Hayes (1953) reported the genus to contain about 150 species, though only a few have been studied in detail. Bailey (1919) reported that the two species, *pyriferum* and *pomiferum* mentioned by Linnaeus are nothing but trees with pear shaped and round shaped fruits. Subsequently, other species were recognized and documented. The wild species of guava are of considerable importance in breeding programmes.

P. Cattleianum var.cattleianum (Sabine) *syn: P.littorale* (Raddi) *var. longipes (Berg.)*

It is a wild subtropical species closely related to guava. It can adapt to many soil types and is quite cold resistant. It is a small tree or shrub with a smooth bark. Leaves are obovate elliptic and glabrous. Fruits are round, about 2.5 cm in diameter and very fragrant. The skin is thin, pulp is soft with numerous seeds. It has a sweet flavour and good aroma. It is also known as 'Strawberry guava' because of the sweet aroma reminiscent of strawberry. Since this lacks muskiness of the common guava, it is preferred among certain tribals (Normand, 1994).

P. Cattleianum (Sabine) *var. lucidum syn: P. littorale* (Raddi) *var. littorale (Berg).*

It is a relatively hardy subtropical species. The fruits are small, globose, juicy, acidic and sulphur yellow in colour. It is also called 'lemon guava'.

P. guineense (Sw). Syn: *P. molle* (Bertol), *P. araca* (Raddi), *P. Schiedeanum* Berg.

It is also called Brazilian or Castilian guava. It is a slow growing shrub, about 1 to 3m long and withstands short periods of drought. The leaves are oblong, scantily hairy on the upper side but coated beneath with pale or rusty hairs and distinctly dotted with glands. The fruits are round with yellow skin, pale yellow pulp surrounding the white central pulp. It contains numerous hard seeds (Mortan 1987).

P. friedrichsthalianum (Niedenzu)

It is a tall tree about 7-10m high. The branches are slender and smooth. Leaves are oval or oblong/oval, smooth, almost glossy above and pubescent below. Fruits are globose, small and sour. The fruits are good for jelly making because of their high acidity. Reported to be wilt and nematode (*M. incognita*) resistant, it is also called Chinese guava or Costa Rican guava.

P. montanum (Swartz)

It is generally found in the mountains of Jamaica. The branchlets are four angled, leaves oblong to oval, glabrous, fruits are round, pulp white with more number of seeds. It produces fruits of poor quality.

P. araucanum (Soares-Silva and *Proenca)*

A large tree with membranous leaves, brochidodromous venation, long petioles and peduncles, flowers solitary, axillary or ramiflorous or in short racemes, with two pairs of flowers. Fruits globose or pear shaped, thin pericarp, fruits yellowish green when mature, seeds angular or lenticulate.

P. acutangulum DC

The shrub or tree ranges in height from 26 to 40 ft. Its branchlets are quadrangular and winged near the leaf base and the new growth is finely hairy. Leaves are elliptical with very short petioles. Fruits are round to pear shaped, pale yellow to yellowish white acidic pulp but well flavored pulp containing few hard, triangular seeds. The fruits are mixed with honey and eaten or, made into acid drinks or preserves.

CYTOLOGY

In guava, most of the commercial varieties are reported to be diploids, the chromosome number being 2n = 22, except the seedless types which are triploids (Kumar and Ranade, 1952). Cytological studies made on structure and behavior in different varieties of *P. guajava* by several workers indicated that meiosis was normal with formation of 11 bivalents at diakinesis, and normal distribution of chromosomes at later stages (Raman *et al*., 1969). The chromosome number of *P. friedrichsthalianum* Niedenzu was reported to be 2n = 22 (Srivastava, 1977). A natural triploid with somatic chromosome number of 2n = 33 was reported by Kumar and Ranade (1952). The same chromosome number was reported by Raman *et al* (1971) in a seedless variety of *P. guajava* suggesting that triploidy is the cause of seedlessness in guava. Shafaat Mohammed

(1975) studied breeding behaviour of the aneuploids in guava (*Psidium guajava* L.) such as trisomic, tetrasomic and higher aneuploids. He observed that reciprocal crosses between aneuploids and diploids indicated less than 100% crossability. The aneuploids, when used as the male parent, crossed less frequently than as female parents and some aneuploids crossed more readily than others. Differences were observed in fruit size, fruit weight, and seed number in reciprocal crosses. The extra chromosome was found to be transmitted through both the egg cell and the pollen. However, frequency of transmission was greater through the egg cell than the pollen. As high as 26% transmission of extra chromosomes were obtained through the egg cell. There was no clear cut difference between trisomics and higher aneuploids with regard to frequency of transmission of the extra chromosomes. In guava, where large number of seeds is a disadvantage, aneuploidy breeding appears to be beneficial.

FLORAL BIOLOGY

The knowledge of flower bud development, time of anther dehiscence and anthesis, extent of fruit set and degree of cross pollination are a pre-requisite for planned hybridization for crop improvement.

In guava, flower buds are borne in leaf axil on current season's growth, either singly or in cymose of two or three (Braganza, 1990). Guava is reported to require about 30 days in Northern India from flower bud differentiation to complete development upto the calyx cracking stage (Singh and Sehgal, 1968). However, under Bangalore conditions, Braganza (1990) reported that the period varied from 45 to 51 days. The flowers consist of a superior calyx with five lobes and the corolla consists of 6 to 10 petals arranged in one or two whorls. The androecium consists of 160 to 400 thin filaments carrying bilobed anthers, closely packed together. The gynoecium consists of an inferior ovary, syncarpous, with axillary placentation and subulate style. The style is smooth and bearded at the summit. Three flowering seasons, *viz.*, 'Ambe bahar', 'Mrig bahar' and 'Hatti or Hasta bahar' have been reported in the peninsular regions of India (Cheema *et al* 1954). However, some workers reported only two flowering seasons (Sehgal and Singh, 1967; Sachan *et al*, 1969; Srivastava, 1974; Syamal *et al*, 1980; Ojha *et al*, 1986). In guava, it has been observed that the flowering season does vary between regions. Generally, three flowering seasons are recognized in the tropical South India and only two seasons in the subtropical North India.

In guava, peak anthesis was found to be between 6 and 7.30 A.M. under North Indian conditions (Singh and Sehgal, 1968). Dehiscence of anthers was observed to take place 15 to 30 minutes after anthesis and continued upto 2hrs (Balasubramanyam, 1959). Pollen fertility has been generally found to be high in guava (78 to 91%) in diploid varieties. Balasubramanyam (1959) found 4% sucrose solution to be the best medium for artificial germination of pollen. The pollen is reported as round with large grains (Srivastava, 1974). Stigmatic receptivity, as studied by fruit set following controlled pollination, was observed to be maximum on the same day as anthesis. Stigma was found to be receptive two days before dehiscence, extending upto 4 days (Singh and Sehgal, 1968).

VARIETIES

Varietal description and nomenclature of different guava varieties grown in India are greatly confusing. Some varieties were named according to shape of the fruit, skin colour and pulp colour, while, several other varieties were named after in the place of origin. Pandey (1968) made detailed studies in different cultivars of guava and classified them into the white pulp group and the red pulp group.

Guava is largely a self-pollinated crop, but cross-pollination also does occur. This results in a large variability in the seedling population from which promising genotypes have been selected in different agro-climatic regions of the country. Promising cultivars of different Indian states are given below:

State	Cultivars
Andhra Pradesh	Allahabad Safeda, Anakapalli, Banarasi, Chittidar, Hafsi, Sardar, Smooth Green and Smooth White
Assam	Amsophri, Madhuriam, Safrior Payele
Bihar	Allahabad Safeda, Chittidar, Hafsi (Red Fleshed), Harijha, Seedless
Gujarat	Nasik, Seedless, Sindh
Karnataka	Allahabad Safeda, Arka Mridula, Sardar, Navalur
Maharashtra	Dharward, Dholka, Kothrud, Lucknow-24, Sardar
Tamil Nadu	Anakapalli, Banarasi, Bangalore, Chittidar, Hafsi, Nagpur Seedless and Allahabad Safeda
Uttar Pradesh	Allahabad Safeda, Apple Colour, Chittidar, Red Fleshed, Banarasi Surkha, Sardar, Mirzapur Seedless
West Bengal	Bariampur and cvs. of Uttar Pradesh

About 160 genotypes, including some *Psidum* spp., are available in Indian collections and are maintained at several centres within the country in field gene banks. Nomenclature of the cultivars of guava grown in India is not yet well established. Some of the varieties have been

named according to shape, colour, and smoothness of skin or by the place of their origin. Characters like plant growth, yield and physico-chemical composition of different guava varieties were reported by several workers. Varietal evaluation was carried out by many workers who reported performance of these varieties under different agro-climatic conditions (Golberg and Levy, 1941; Teaotia *et al*, 1962; Srivastava and Srivastava, 1965; Singh *et al*, 1979; Chadha *et al*, 1981; Dinesh and Reddy, 2001). Characteristic features of some of the important guava cultivars grown in India are given below:

Allahabad Safeda : It is the most popular variety in India and has acquired large variations due to seed propagation. This is the progenitor of many Indian varieties and occupies the largest area under cultivation. Fruits are round, large in size, skin with smooth, light yellow on ripening, pulp white, firm, excellent in quality with high TSS and Vitamin C, pleasant flavour and a few, soft seeds.

Anakapalli: Fruits are medium sized with red pulp. Seeds are soft and plenty. Fruits are slightly oval.

Apple Colour: The trees are medium in vigour and are moderate yieldes. Fruits are medium sized, with apple coloured skin; cool temperature is required for good colour development. The pulp is white and firm, sweet to taste.

Bangalore: Fruits are medium to large in size, pulp is white with good taste and flavour.

Chittidar: This variety is very popular in Western Uttar Pradesh. The fruits are characterized by numerous, red dots on skin. Fruits are sub globose, with white pulp, high TSS and Vitamin C content of 240 mg /100g pulp.

Hafsi: Fruits are spherical in shape with thin skin and medium size. The pulp is red with good taste and flavour. Seeds are comparatively less in number, but hard.

Red Fleshed: Fruits are medium sized with red pulp, round, smooth skinned, seeds are plenty and medium soft. Fruits possess sweet flavour, are rich in Vitamin C (386 mg /100g pulp).

Sardar (Lucknow 49): It is a selection from Allahabad Safeda made at Poona during 1927. The plant has a spreading nature. The tree is dwarf with open, rounded crown. The fruits are medium to large and contain a crisp, soft and creamy pulp; it is a heavy bearing variety. The fruit has a slightly acidic flavour, attractive aroma, with many seeds, and good keeping quality.

Smooth Green: Fruits are round and medium sized. Skin is glossy and greenish yellow when mature. Pulp is white, good in taste and flavour.

Nasik: Fruits are medium sized, round, with white pulp, sweet with good flavour.

Banarsi Surkha : Trees are medium sized (5.1m) with a broad crown, fruit shape is round and surface smooth, skin colour golden yellow, pulp colour pink, seed number very high, seed texture very hard.

Seedless: The plants are very vigorous. There are different varieties, like, Saharanpur Seedless, Nagpur Seedless, Sringeri Seedless, etc., which are nearly identical. Two types of fruits, *viz.*, long, big sized with warty surface, and yellow, thin skin with swollen calyx end and round; small, with very few seeds. The pulp is white, good to taste and has aroma, contains moderate to high levels of Vitamin C (240 mg / 100g pulp).

Navalur: It is a variety grown in Dharwad district of Karnataka. It is hardy in nature, drought tolerant and resistant to canker. The important cultivars are: CIW-2 (Channappa Itigatti White), CIW-3, CIW-4, CIW-5, GR 1 (Ghatage's Red number one), GR 3, GW-1 (Ghatage's white number), GW-4, SR-1 (Shivammanavar red number one), SR-2, SW-2 (Shivammanavar white), SWY-1 (Shivammanavar white yalakki).

Apart from these prominent commercial cultivars, other cultivars grown in localized areas are: Pear Shaped, Apple Colour, Banarasi Surkha, Sangam, Seedless, Dholka, Sindh, Karela, Mirzapuri Seedling, Superior, Pourtgal, Spear Acid, Superior Sour Licidium, White Fleshed, Behat Coconut, Smooth white, Amsophri, Madhuriam, Bariampur, Harijha,

Dharwar, Safri or Payera, Soh-Pryiam, Am-Sophri, Kaffree, Supreme, White Supreme, Bangalore, Bhavnagar, Gwalior-27, Kafri (Pear shape), Kafri (Round shape), Rewa-72, Hazi sahib, Kohir, etc.

CROP IMPROVEMENT

In India, during the early days, guava plants were generally propagated by seeds from limited varieties available with nurserymen and pomologists. The seedling population obtained by open pollination gave rise to considerable variation in the form and size of fruit, the nature and flavour of pulp, seediness and other morphological characters such as spreading or erect growth habit of trees (Naik, 1949). Cheema *et al*., (1954) observed all the cultivars of guava to be highly heterozygous. Commercial producers utilized the variation thus obtained for selection of desirable genotypes and propagated them vegetatively. Assessment of genetic diversity and relationship among *Psidium* spp was carried out by Sharma *et al* (2007). They observed a high genetic similarity between Chinese guavas grouped with *Psidium guajava* cultivars.

It has been observed that only a few named varieties are under cultivation. Most of these varieties suffer from one defect or the other. Hence, guava improvement by breeding was started mainly with the following objectives for developing new cultivars:

i) dwarf plant habit suitable for high density planting

ii) fruits with uniform shape, size, good colour, firm and thick pulp, good aroma, few and soft seeds, high TSS and high pectin

iii) long shelf life

iv) resistance to *Fusarium* wilt

Plant introduction

Most of the guava varieties have evolved as selections from seedling variants. Variability has come about because of open pollination from highly heterozygous parents. Several introductions of promising genotypes have been made in guava growing countries. In India, many introductions made from Hawaii, Brazil, Thailand, etc. are being cultivated and used in breeding programmes. Similarly, introductions of Indian cultivars like Allahabad Safeda and Sardar have given excellent results in other parts of the world (Gonzaga *et al*, 1999).

Although introduction is a potential tool in any crop improvement programme as it considerably saves time, there is also the danger of new diseases getting introduced. It has been our observation that some of the introduced exotic acidic types like Beaumont were prone to 'Stylar End Rot' caused by *Phomopsis psidii*. This character was inherited even by hybrids. Hence, utmost care needs to be taken while introducing varieties. Unless these are observed under plant quarantine, further multiplication or usage on field scale should not be made.

Selection

At Ganeshkhind Fruit Experimental Station, Pune, India, guava improvement work was initiated in1907, primarily with collection of seeds of varieties grown in different places, to isolate superior strains. One strain from open pollinated seedlings of 'Allahabad Safeda' collected from Lucknow was selected and released as 'Lucknow-49' (Cheema *et al*, 1954) which became very popular and has now been renamed as 'Sardar' (Phadnis, 1970) after trials at Saharanpur (Singh,1953) and Kodur (Rangacharlu, 1954). The plant has a spreading nature. The tree is vigorous, dwarf with open rounded crown. Fruits are medium to large and contain a crisp, soft and creamy pulp. It is a heavy bearing variety. Fruits have a slightly acidic flavour, attractive aroma, with many seeds and good keeping quality.

At the Fruit Research Station, Saharanpur, one superior selection, *viz.*, S-1, with good fruit shape and quality, few seeds, sweet taste and high yield was isolated (Singh, 1959).

At Faizabad, seedling selections were made from 'Allahabad Safeda' and many selections were made under 'Faizabad Selection' (Pathak and Dwivedi, 1988).

At CISH, Lucknow, four seedling selections of guava, namely, CISH-G-1, CISH-G-2, CISH-G-3 (Lalit), CISH-G-4 (Swetha) have been released and their performance was studied by Marak and Mukunda (2007).

CISH-G-1: It is a selection from local red fleshed type, with attractive fruits having deep red skin, firm pulp with high TSS, soft seeds.

CISH-G-2: Selection from local red fleshed type, crimson colored attractive fruit, stripes in groove, seeds soft.

CISH-G-3 (Lalit): It is a selection from a high yielding variety, responsive to primary and high density planting. Fruits are round, weighing 150g, pink pulp suitable for both table and processing purposes.

CISH-G-4 (Swetha): Plants are semi vigorous, medium in height and are prolific bearers. Fruits are round, weighing 225g, with white pulp with good keeping quality.

At the Indian Institute of Horticultural Research, Bangalore, out of 200 open pollinated seedlings of the variety 'Allahabad Safeda' (collected from Uttar Pradesh), one seedling selection, 'Selection-8', was found to be promising and was released as 'Arka Mridula'. This variety has been reported to have also performed well with respect to yield and quality under rainfed conditions of Bihar (Ramkumar, 1998). The plants are semi-vigorous and spreading in nature. Fruits are round in shape and weigh about 180g. Fruit are yellow in colour with smooth skin.The pulp is white, firm, sweet with few soft seeds. The TSS is 12.0°B, 100 seed weight is 1.6g. Keeping quality is good. Pectin content is 1.04 %. The variety is good for jelly making.

At Allahabad Agricultural Institute, Allahabad, a selection from the local red pulp type has been released as 'Allahabad Surkha'. The plans are vigorous, dome shaped and compact. Trees are high yielding, producing upto 120kg per plant. The fruits are round with uniform, pink skin and deep pink pulp, sweet, strongly flavoured and with few seeds.

At Bulakihar (Malihabad), Lucknow, a selection has been made as **'**G. Vilas Pasand'. The trees are vigorous, wide spreading with bushy, low growing habit. Fruits are round to ovoid, skin texture is course to smooth, fruit skin pale yellow to golden, colour of flesh is creamy white texture creamy soft, very large (400g to 800g) fruit, less seeds, very productive throughout the year. High content of Vitamin C makes it stand out among guava varieties.

At Aurangabad and Bihir districts of Marathwada, three promising selections, *viz*., ABD3, BHR3 and BHR5 were made out of the 12 strains collected (Thonte and Chakrawar,1981).

At Narendra Dev University of Agriculture and Technology, Faizabad (UP), of the 23 strains collected from a survey of guava growing regions, 3 seedlings of Allahabad Safeda (AS1, AS2 and AS3) and 2 of Faizabad selection (FS 1 and FS 2) were found to be promising with respect of fruit quality and yield (Pathak and Dwivedi, 1988).

At GBPUA&T, Pantnagar (Uttaranchal), A selection was made as 'Pant Prabhat' for commercial cultivation. Plant growth in this line is upright; with broad leaves, the tree is highly productive (100 -125kg). Fruit skin is smooth and light yellow in colour, fruits medium sized with average fruit weight of 150-172g , pulp is white, seeds are small and medium soft, the fruit has a sweet taste with pleasant flavour, ascorbic acid content varies from 125mg (rainy season) to 300mg/ 100g fruit weight (winter season). TSS varies from 10.5 to 13.5^{0}B.

At the Fruit Research Station, Kuthulia, Rewa, a selection from an old, seedling orchard was made as 'Dhareedar'. The trees are vigorous, medium tall with erect and upright branching and a flat crown. Fruits are medium to large sized, roundish ovate in shape with 5-7 raised lines on the surface of mature fruits, the peel in greenish yellow, the pulp soft and sweet (TSS 11.7^0 Brix).

Hybridization technique

In guava, flowers that are chosen for crossing are emasculated when at the 'calyx break stage', a day before opening. Pollen from the pollen parent is brought from an unopened flower, at preferrably at calyx break stage. The stigmatic surface is gently smeared with the pollen and flowers are bagged. Under Bangalore condition pollination carried out during the morning hours between 10 AM to 12 noon has given better results.

Intervarietal Hybridization

In general, intervarietal crosses in guava are successful, having no crossability barriers. However, varietal cross incompatibility in guava is reported in crosses made between 'Behat Coconut' and 'Sardar', 'Behat Coconut' and 'Apple Colour'. In India, breeding work for guava improvement has been going on at several research institutions. At HETC, Basti, a number of cross combinations of 'Seedless' x 'Allahabad Safeda', 'Seedless' x 'L-49', 'Allahabad Safeda' x 'Patillo', 'Apple Colour' x 'Red Fleshed' and 'Apple colour' x 'Kothrud' were made. None of the 55 hybrids obtained from these crosses were found to be promising (Chadha, 1998).

At the Fruit Research Station, Sangareddy, A.P., two hybrids, 'Safed Jam' and 'Kohir Safeda' were selected out of crosses of 'Allahabad Safeda' x 'Kohir' and 'Kohir' x 'Allahabad Safeda', respectively, and released. These hybrids are particularly recommended for semi arid tropical areas. These have also been found to be suitable for the preparation guava juice (Mitra and Bose, 1985; Shanmugavelu *et al*, 1987).

At the Indian Institute of Horticultural Research, Bangalore, 'Arka Amulya' a hybrid was released through intervarietal hybridization involving 'Allahabad Safeda' and 'Triploid' (Subramanyam and Iyer, 1998; Anon., 1996).

At the Fruit Research Station, Anantharajupet, Andhra Pradesh, out of 6 hybirds, two (H1 and H6) were

found promising with regard to fruit quality and precocious bearing (Rama Rao and Dayanand, 1977).

At Rajendra Agricultural University, Sabour, Bihar, 210 F_1 hybrid seedlings were raised from various intervarietal crosses. A hybrid from the cross 'Apple Colour' x 'Sardar' had maximum fruit weight, Vitamin C and pectin content (Singh and Hoda, 1994).

Chaudhary Charan Singh Haryana Agricultural University, Hisar, released two guava hybrids, namely, Hisar Safeda (Allahabad Safeda x Seedless) and Hisar Surkha (Apple Colour x Banarasi Surkha).

Traits of some of the guava hybrids released recently are given below:

Safed Jam: This is a hybrid between the cross 'Allahabad Safeda' X 'Kohir' developed at FRS, Sangareddy. The tree is medium in height and a heavy yielder. Fruits are round in shape, large in size with a thin peel, good taste and few, soft seeds.

Kohir Safeda: It is a cross between a selected, heavy yielding line of Kohir X Allahabad Safeda. The tree is vigorous, fairly large in size and dome shaped. Fruits are large, with few, soft seeds and white pulp.

Arka Amulya: As stated above, this is from the cross 'Allahabad Safeda' X 'Triploid' developed at the Indian Institute of Horticultural Research, Bangalore. Plants are semi-vigorous and spreading type. Fruits are medium sized (180-200g), weight of 100 seed is 1.8g, pulp is white, with high TSS (12.5°B), fruits have good keeping quality.

Arka Kiran: It is from the cross 'Kamsari' X 'Purple Local'. Plants are semi vigorous, amenable to high density planting. The fruits are sub globose, weighing about 200-230g .The pulp is deep pink, thick and has good flavour. The seeds are medium soft (9.0 kg cm^{-2}), with high lycopene content (7.45 mg /100g) and TSS of 12.0 to 12.5°B.

Hisar Safeda: It is from the cross 'Allahabad Safeda' X 'Seedless', developed at CCSHAU, Hisar. Plants are upright, trees have a compact crown. Fruits are round, with a smooth surface, creamy yellow skin; average fruit weight is 92g, creamy white pulp, few soft seeds high TSS (13.4%).

Hisar Surkha: It is from the cross 'Apple Color' X 'Banarasi Surkha'. The tree crown is broad to compact. Fruits are round, skin yellow with red dots, average fruit weight 86g, pulp pink, seed count medium, TSS high (13.6%).

Autopolypoloidy: From several parts of our country, seedless varieties have been reported. At Poona, Kumar and Ranade (1952) reported a triploid guava variety with 33 chromosomes, and suggested it to be autotriploid. The chromosome status of seedless varieties available at IARI and Saharanpur was studied by Majumder and Singh (1964) and were found to be autotriploids. Iyer and Subramanyam (1971) were of the opinion the production of any more triploids was futile since fruit shape in triploids is highly irregular with mis-shapen fruits because of differential seed size. A natural autotetraploid in *P. guajava* was reported by Naithani and Srivastava (1966). Tetraploidy in guava has been induced too with colchicine treatment (Janaki Ammal, 1951; Ram Kumar, 1975).

Aneuploidy: At the Indian Agricultural Research Institute, New Delhi, with a view to evole a variety with fewer seeds and high yield, crosses were made between seedless (triploid) and seeded (diploid) 'Allahabad Safeda'. Of the 73 F_1 hybrid seedlings raised, 26 were diploids, 9 trisomics (2n+1), 5 double trisomics (2n+1+1) and 14 tetrasomics (2n+2). They showed distinct variation in tree growth habit and, leaf and fruit characters. Three tetrasomic plants had dwarf habit and, normal shape and size of fruits, with less number of seeds (Majumder and Mukherjee, 1972a,b; Mukherjee, 1977). In the progeny of open pollinated triploid, and triploid with diploid (Mohammad, 1974), 30 trisomics, 2 double trisomics, 1 tetrasomic and higher aneuploids were obtained. Reduction in growth and size of leaf distinguished aneuploids from diploids. Aneuploids, particularly trisomics, had promising qualities and may prove useful in developing plants with reduced seediness and, possibly, in providing dwarfing rootstocks.

Sharma (1982) identified a promising tetrasomic dwarfing rootstock (Aneuploid No. 82), through selection, out of 48 aneuploid seedlings at IARI, New Delhi. Studies conducted on the effect of aneuploid No. 82 rootstock on growth and yield of 'Allahabad Safeda' showed that the aneuploid induced substantial dwarfing in Allahabad Safeda in terms of plant height, plant spread and tree volume. Overall yield/unit volume of the plant was highest in aneuploid

No. 82, which indicated its strong potential for use as dwarfing rootstock on a commercial scale, for increasing production and profitability of guava orchards (Sharma *et al*, 1992).

Mutation: According to Cheema and Deshmukh (1927), naturally occurring mutations are not rare in guava. Brar and Bal (2003) investigated the effect of gamma rays (1,2,3,4 and 5 kR) on buds of guava cv. Sardar. After the treatment, these were budded onto Lucknow-49 rootstock. Variability for plant height, internodal length and stem diameter was maximum in 2 kR treatment; while, for number of branches, number of leaves and breadth of leaves, maximum variability was noted in 4, 1 and 3 kR treatments, respectively. Mutagenic treatments had no significant effect on stomatal size.

In vitro mutagenesis, followed by micropropagation *via* axillary bud proliferation of shoot tips in guava, was carried out by Zamir *et al* (2003). Shoot tips irradiated with gamma rays at 15-90 Gy and cultured in Murashige and Skoor's (MS) medium containing 3% sucrose, 6-benzylaminopurine (benzyladenine) (BAP) and L-glutamine. Optimum shoot proliferation was recorded in MS medium supplemented with 1.0mg BAP and 250mg L-glutamine/litre. Rooting of cultured shoots was observed in half-strength MS medium supplemented with IAA and IBA. LD_{50} was observed at 45 Gy. Rates of more than 75 Gy were lethal to explants.

Biotechnological techniques

Biotechnological techniques can be useful chiefly in breeding for disease resistance and in germplasm storage using tissue culture techniques. Use of tissue culture and micropropagation of superior guava cultivars has been discussed by several researchers (Amin and Jaiswal, 1988; Jaiswal and Amin, 1987; Loh and Rao, 1989; Papadatou *et al*, 1990). Jaiswal and Amin (1992) felt that somatic cell genetics could be useful in guava breeding for specific objectives. A technique for successful *in vitro* propagation of guava germplasm using shoot-tip explants from mature trees was reported by Jaiswal and Amin (1987). These workers also demonstrated that adding activated charcoal to the medium enhanced rooting of explants and vegetative growth of established plantlets. Risterucci *et al*, (2005) constructed a library of microsatellite-enriched (GA)n and (GT)n and, 23 nuclear simple sequence repeat (SSR) loci were characterized in the guava species *Psidium guajava* L.). All the SSR loci were found to be polymorphic after screening for diversity in different cultivars, and across-taxa amplification tests showed potential transferability of most SSR markers in three other *Psidium* species. First to be published for *P. guajava*, this new SSR resource will be a powerful tool for genetic studies in guava, including cultivar identification and linkage mapping, as well as potentially, for, interspecific genetic studies within the genus *Psidium*. Rapid multiplication of seedling plants of guava through *in vitro* shoot-tip culture and subsequent plant establishment also has been successful (Papadatou *et al*, 1990).

Somaclonal variation, which normally occurs in several tissue cultures (Larkin *et al*, 1985; Evans and Sharp, 1988; Lee and Philips, 1988) could be useful for selecting guava plants resistant to the wilt disease. An efficient protocol for plant regeneration from callus culture would also be helpful in selecting plants resistant to disease or environmental stresses. Recovery of plants of haploid origin from anther/pollen culture of guava could offer advantages in breeding (Jaiswal and Amin, 1992).

Chandra *et al* (2004) attempted embryogenesis and plant regeneration from mesocarp of *Psidium guajava* L. (guava) and developed a protocol for induction, maturation and germination of somatic embryos from this tissue. Explants were cultured on modified MS medium fortified with 2,4-D (2.0 mg/l), ascorbic acid (100 mg/l), L-glutamine (400 mg/l) and sucrose (6%). Embryogenic proliferating tissue was induced, which was found to be translucent, mucilaginous and it differentiated into many, small somatic embryos. The somatic embryos were retained in the same medium, where simultaneously differentiation of new somatic embryos and their conversion into plantlets was observed. Thus, embryogenesis in guava can be perpetuated and could be used in the future for carrying out cellular selection against wilt causing organism.

Phylogenetic affinity, usefulness of wild species and interspecific hybridization

Utilization of wild species for crop improvement has been one of the ways to introduce certain gene(s) for specific purposes like hardiness, disease and pest resistance, etc. However, in perennial crops, because of the long time gap, efforts have not been made to their full potential. Exploitation of wild species requires extensive knowledge of taxonomy, reproductive biology & cytogenetics, genetics, crossability barriers and fertility of the hybrids. Although success obtained in fruit crops is low, many desirable traits with great potential for crop improvement are found in the

wild species. Interspecific crosses in many fruit crops, between cultivars and species, have resulted in hybrids that are partially sterile due to ploidy level differences genomic incompatibilities and cytoplasmic imbalances. In order to develop a rootstock tolerant/resistant to guava wilt and to test the possible role of different species, studies have been initiated on interspecific hybridization, in the genus *Psidium*. Phylogenetic studies carried out in *Psidium* species utilizing differences in flavonoid patterns showed a close affinity between *P. guajava* and *P. molle*. Two species, *P. molle* and P. *guineense* were found to be morphologically similar with minute differences in their chromatographic pattern (Das and Prakash, 1981). These workers also found a close affinity between *P. guineense*, *P. pumilum* and *P. chinensis*. It was observed that *P. guajava* and *P. chinensis* were crossable. However, *P. guajava* and *P. molle* are cross incompatible when *P. guajava* is used as the female parent (Subramanyam and Iyer, 1982). Leslie *et al* (1995) and Edward & Shankar (1964) reported that *Psidium friedrichsthalianum* Niedenzu to be resistant to guava wilt. The other species reported to be resistant to the wilt are *P. cumuni*, *P. cattleianum* var. *lucidum*, *P. molle*, and *P. guineense*. However, Singh et al (1977) reported that *P. cattleianum* var *lucidum, P. corecium, P. cajuvalis, P. guineense* and *P. friedrichsthalianum* developed wilt infection. Hence, intensive work is needed to develop useful interspecific hybrids resistant to wilt using the resistant species in breeding programmes.

The contribution of wild species to crop improvement and management programmes mainly involves their utilization as rootstocks for regulation of vigour, yield, fruit quality and, disease and pest resistance. Pathak and Ojha (1993) enumerated their potential uses: *P. cujavalis, P. molle, P. cattleianum* and *P. guineense* can be used as rootstock. Chinese guava (*P. friedrichsthalianum)* and Philippine guava are compatible rootstocks and have been reported to be resistant to the wilt disease. 'Allahabad Safeda' trees grafted on to *P. pumilum* had a dwarfing influence. *P. cujavallis* produced the largest trees but with non-uniform and rough skinned fruits. Singh *et al* (1976) observed that fruits of 'Allahabad Safeda' contained higher sugar content on to *P. pumilum* while, higher ascorbic acid content was recorded in these grafted on *P. cujavallis*. High yields were obtained using *P. cattleianum* rootstock. Other species reported to be resistant to the wilt are: *P. cumunii, P. cattleianum var. lucidum, P. molle* and *P. guineense*. However, Singh *et al* (1977) reported that *P. araca, P. cattleianum var lucidum, P. corecium, P. cujavalis, P. guineense* and *P. friedrichsthalianum* developed infection. At the Central Institute for Subtropical Horticulture, Lucknow, Uttar Pradesh, interspecific hybridization was carried out between *P. molle* and *P. guajava*. The interspecific hybrids have been found resistant to guava wilt and are graft compatible with commercial varieties of *P. guajava* (Anon., 2003-04).

Inheritance studies

Genetic studies conducted in guava at IIHR, Bangalore, indicated that red pulp colour was dominant over white and that this character was governed monogenically. Many cultivated red fleshed varieties were found to be heterozygous for this character. Bold seeds were found to be dominant over soft seeds and this was also found to be determined monogenically. Linkage was also found between flesh colour and seed size, i.e., red flesh with bold seeds (Subramanyam and Iyer, 1982). Mitra and Bose (1985) have reported heterosis in guava. Studies conducted at Coimbatore by Raman *et al*, (1969 and 1971), have shown that triploidy and genetic factor(s) are responsible for female sterility and, that; variation among triploids is due to their independent origin from a distinct diploid variety. Iyer and Subramanyam (1971) opined that seedless varieties of guava were triploids, grew vigorously and bore fruits that were irregular in shape with ridges, because of irregular distribution of seeds of various sizes. Dinesh and Yadav (1998) carried out half-sib analysis in progenies of the variety 'Apple Colour'. They observed that genotypic variance was lower than the phenotypic variance, and heritability was moderately high for all the characters implying, that, selection may be practiced for improvement of fruit characters. Hence, hybridization among less seeded diploids can be adopted in an improvement programme. It is our observation on inheritance pattern using 'Purple guava' and 'Arka Mridula' as parents that hybrids segregate in a ratio of 3:1 for green leaf types and purple leaf types.

Characterization and evaluation

Cluster analysis was carried out using fifteen morphological characters in 29 varieties and 5 species. The cluster diagram showed four main clusters (Fig. 1). In the first cluster, the species *P. cattleianum, P. friedrichsthalianum* and *P.molle* are placed. The second cluster consists of 8 varieties and one species, viz., Bangalore Local, Benaras, Dharwad, Karela, Kamsari, Spear acid, Surka Chitti, Surka Chitti Neputani and *P. quadrangularis*.

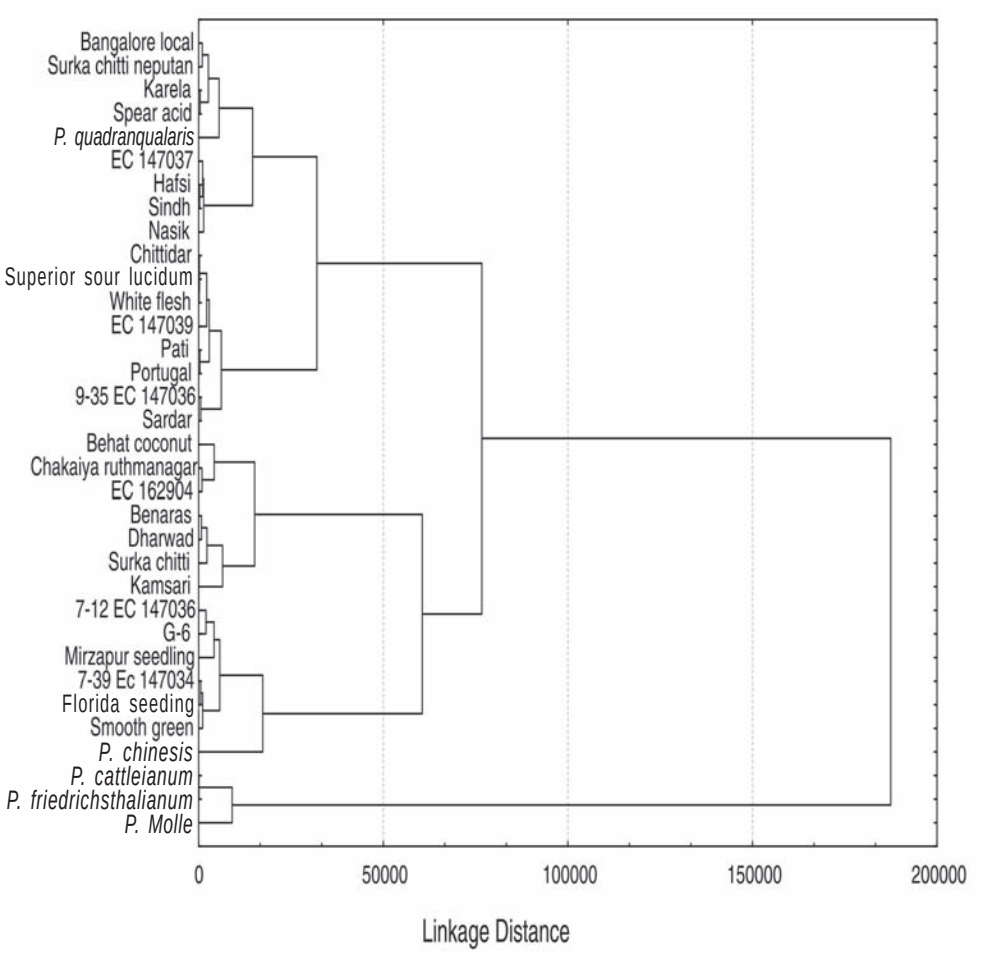

Fig 1. Tree diagram for 29 varieties and 5 species

In the third cluster, 10 varieties are grouped, viz., Chittidar, EC 147039, 147037, Hafsi, Nasik, Pati, Portugal, Sindh, Superior sour lucidum and White flesh. In the fourth cluster, Behat Coconut, Chakaiya Ruthmanagar, EC 147306, EC 147036, EC 147034, EC 162904, Florida seedling, G-6, L-49, Mirzapur seedling, *P.chinensis* and Smooth Green are grouped together.

The cluster means indicate that fruit weight, fruit volume, fruit length and width are greater more in Cluster II and low in Cluster I. Members of cluster I are mainly species that usually bear small sized fruits, except *P. quadrangularis*. The mean Vitamin 'C' content and fruit / seed ratio was maximum in cluster IV, which indicates that hybridization involving these accessions would be expected to result in maximum hybrid vigour. Mean acidity ranged over 1.00 to 1.65 % and total sugar was about 7.23 to 7.86g among different clusters. Crosses between Cluster I and Cluster II crosses may result in desirable combinations leading to development of varieties with good processing traits.

Principal component (Fig. 2) analysis shows that the species *P.molle, P. friedrichsthalianum* and *P. cattleianum* var. *lucidum* are closely related and are away from varieties of *P. guajava* and the species, *P. chinensis*. Due to the edible nature of *P. chinensis*, it is closely related to *P. guajava. Although P. quadrangularis* is not placed with the species group,it is different from *P. guajava* varieties as well. Fruits of *P. quadrangularis* are not edible, but because of fruit size, it is placed with the *P. guajava* varieties. Seeds of *P. quadrangularis* are unusually large compared to *Psidium guajava* varieties or any other species. The cluster analysis clearly shows that the species are different from cultivated *Psidium guajava* varieties and considerable diversity is present for various characters within the species of *P. guajava* for breeding varieties with good fruit size, fewer number of seeds or for dwarfness.

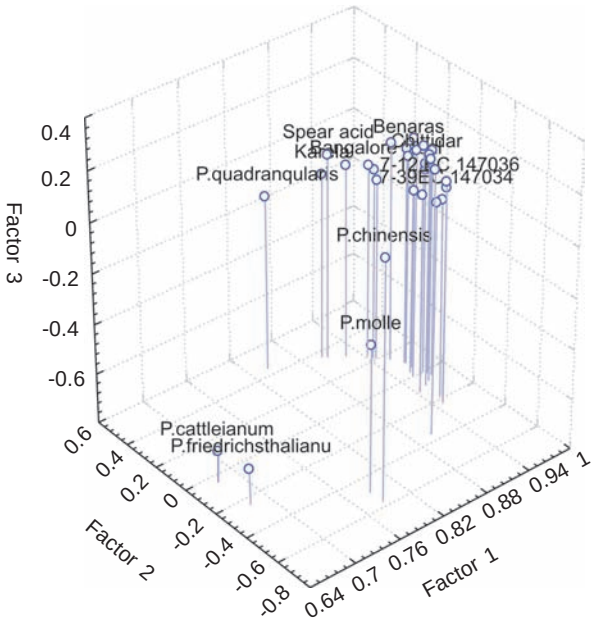

Fig 2. Principal component analysis

Guava varieties based on shape, colour and weight

Classification of guava varieties

Based on fruit shape

Variety : Lalit

Globose: Allahabad Safeda, Apple Colour, Arka Amulya, Arka Mridula, Benaras, Behat Coconut, Chittidar, Dharwad, EC 147037, Hafsi, Local 2, Mirzapur seedling, Nasik, *P. cattleianum var. lucidum, P. chinensis, P. friedrichsthallianum, P. molle, P. quadrangularis,* Phili (pink), Philippine guava, Red flesh, Sindh, Smooth green, Surka Chitti, Superior Sour Lucidum, Dhareedar, Aneuploid 2, Lalit

Subglobose: Chakaiya Ruthmanagar, 7-12 EC 147036, 9-35 EC 147036, EC 14089, EC 162904, kamsari, Karela, Local 1, Lucknow 42, Pati, Portugal, Sardar, GR-1, Spear acid, Abu Ishakwala

Pyriform: Bangalore Local, G-6, White flesh

Ovate: Florida seedling, Surka Chitti Neputani

Oblong: Oblong, Aneuploid-1, 7-39 EC 147034, Nagpur Seedless, Seedless triploid, Thailand 2, Lucknow 42

Based on fruit weight

Small (16-100g)

Aneuploid 1, Apple colour, EC 14039, EC 147037, G-6, Hafsi, Local 1, Local 2, Nagpur seedless, *Psidium cattleianum var. Lucidum, P. chinensis, P. friedrichsthalianum, P. molle*, Pati, Philippine guava, Portugal, Seedless (triploid), Sindh, GR1

Medium (100-140g)

Allahabad safeda, Arka Amulya, Arka Mridula, Bangalore Local, Chittidar, 7-39, EC 147034, 7-12 IC 147036, 9-35 EC 147036, EC 162904, Florida seedling, Lucknow 42, Mirzapur seedling, Nasik, Phili (pink), Red Flesh, Sardar, Smooth Green, Spear acid, Surka chitti, Surka Chitti Neputani, White Flesh

Large (>140g)

Abu Ishakwala, Benaras, Behat Coconut, Chakaiya Ruthmanagar, Dharwad, Kamsari, Karela, *P. quadrangularis*, Superior Sour lucidum

Based on skin colour

White: Abu Ishakwala, Allahabad safeda, Aneupoloid 2, Apple colour, Arka amulya, Arka Mrudiula, Behat coconut, Benaras, Chakaiya Ruthmanagar, Chittidar, Dharwad, Florida seedling, Hafsi, Karela, Local 1, Llocal 2, Lucknow 42, Mirzapur seedling, Nagpur Seedless, Nasik, *P. chinensis*, Sardar, Seedless (triploid), Singh, Smooth Green, Superior Sour lucidum, Surka Chitti, Surka Chitti Neputani, White Flesh

Shades of red: Aneuploid 1, 7-39, EC 147034, 7-12 EC 147036, 9-35 EC 147036, EC 147039, EC 163904, EC 147037, GR-6, Kamsari, *P. chinensis*, Pati, Phili (pink), Portugal, Red Flesh, GR1

Variety: Kamsari

Shades of yellow: Spear acid, Bangalore Local, *P. cattleianum var. lucidum*, *P. quadrangularis*

Purple: Purple Local

Variety: Purple Local

Probable Gene donors

Probable donor parents were identified for various horticultural traits as follows:

Character	Accession Name
Dwarfness	Apple Colour, Aneuploid, *Psidium molle, P. chinensis, P. friedrichsthalianum*
Seedless	Seedless
Good yielder	Benaras, 7-39 EC147034, EC 162904, Behat Coconut,
Globose fruit shape	Smooth Green, Allahabad Safeda, Apple Colour, Arka Amulya, Arka Mridula, Benaras, Behat Coconut, Hafsi, Sindh, Mirzapur seedling, Dharwad
Purple pericap	Phillippine guava
Processing	7-12, EC 147036, 7-39 EC 147034
Big sized fruit	One kg guava, Behat Coconut, Benaras, Kamsari, Dharwad, Chaikaiya Ruthmanagar
High TSS	Dhareedar, Allahabad Safeda, Arka Mridula, Seedless, Sindh, Hafsi, Bangalore Local, Surka Chitti, Behat Coconut
High Vitamin C	Mirzapur seedling, *P. chinensis*, EC 162904, G-6, Chakaiya Ruthmanagar, Dhareedar
Suckering habit	*P. chinensis*

The accessions were screened for their variable reactions to insect pests and the least susceptible sources were identified:

Pest	Least susceptible varieties
Fruitfly	EC 147037, EC147039, Kamsari, Red Flesh, Superior Sour lucidum
Tea mosquito bug	EC 147036, EC 147039, Hafsi, Superior Sour lucidum
Spiralling whitefly	Arka Amulya, Benaras, Spear acid, *Psidium chinensis*, *P . friedrichsthalianum* ,EC 147039

Utilization of germplasm

The accessions Allahabad Safeda and Seedless were used in our breeding programme for developing varieties like Arka Amulya (Allahabad Safeda x Seedless) and Arka Mridula (selection from Allahabad Safeda). Interspecific hybridization was carried out using the wild species *Psidium chinensis* with *P. guajava* cv. Beaumont to produce rootstocks resistant to wilt disease. 'Apple Colour' and 'Sardar' were also used in various combinations. 'Red Flesh' and 'Philippine guava' are under use in the breeding programme for imparting red/purple colour to the progenies.

FUTURE NEEDS

Priority needs to be given to developing good fruit quality, since, there is little merit in improving yield and disease resistance if not accompanied by high quality. High quality should include high TSS, good sugar-acid blend, good aroma, attractive skin pulp colour and soft seeds; processing quality, which includes juice colour, high Vitamin C content, higher lycopene content, good pectin content and good flavour. While selecting new varieties, keeping quality may be accorded adequate importance. In this connection, flavour and firmness of the pulp, (that could contribute towards better keeping quality) of the 'Apple Colour' guava as the gene donor could be attempted to improve other commercial cultivars. Hence, attempts to hybridize these genotypes with 'Allahabad Safeda' and other commercial cultivars should be intensified and selections may be made of progenies without apple colour, provided they have all the other desirable characteristics.

In the recent past, efforts have been intensified to develop apple coloured cultivars to make them attractive for local as well as export markets. However, deep red colour has been found to be a very unstable character, the skin colour changing from deep red to yellowish white from season to season, as well as within the same tree. Hence, 'stable' types need to be identified and great care should be taken while selecting hybrids from a large population.

The cultivar 'Allahabad Safeda' possibly represents a population of guava trees grown extensively in Uttar Pradesh (India) rather than being descendent from a single clone. Hence, enormous variation has been observed in this so-called cultivar. It is for horticulturists to make rigorous screening of the population to identify superior genotypes, keeping specific objectives in mind. A survey to locate such genotypes is certainly required, especially in Uttar Pradesh.

While breeding or selecting superior types suitable for high density planting characters like early bearing, compact plant type, favourable response to pruning, good branch angle to minimize branch breakage even under heavy bearing, and with a high fruit-shoot ratio need to be given

due attention. Aneuploidy breeding should be intensified to develop high yielding, high quality varieties with fewer and soft seeds, and to develop dwarfing rootstocks. Autotetraploids of less-seeded, superior diploid varieties may be developed. Induction of mutation by physical and chemical mutagens may be attempted where improvement in a specific character is required in an otherwise acceptable variety.

Since guava cultivation in many locations is threatened by wilt (*Fusarium* spp), work on interspecific hybridization to develop wilt resistant rootstocks should be intensified. Efforts should also be made to develop wilt resistant scion varieties, of which, self-rooted plants could be used for commercial cultivation. The Philippine Guava (Purple type) has shown some promise as a wilt-resistant rootstock but needs extensive experimentation. As this species segregates into the purple and white types on crossing with *P. guajava* cultivars, there is a need to look for possible linkages between purple leaf types and resistance to wilt. Extensive screening of other related *Psidium* species needs to be made for assessing their resistance to wilt. In this connection, reliable pathological screening techniques need to be standardized to hasten the process of disease resistance breeding in guava.

Varietal introduction, though, becomes an essential part of any crop improvement programme and needs to be made with great caution and with strict, customary plant quarantine measures. To quote some examples of caution, the 'Beaumont' variety of guava, when introduced in to India, was found to be severely infected with 'Stylar End Rot' (*Phomopsis psidii)* although it is not a severe problem in its original habitat. Similarly, the 'Giant Thailand Guava' when introduced in to Bangladesh, was found to be highly susceptible to several insect pests. Such examples are many and should be borne in mind.

Molecular characterization of germplasm needs to be accelerated with a view to work out genetical distance so that good recombinants can be arrived at by crossing suitable parents. Biotechnological tools need to be employed extensively. DNA fingerprinting and similar tools may be used to study extent of variation, even with in 'Allahabad Safeda' cultivar. Attempts to spot genetic markers for wilt resistance may be made to improve efficiency for developing wilt resistant clones and rootstocks.

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J. Hortl. Sci.
Vol. 5 (2): 109-113, 2010

Variability studies in Palayankodan ecotypes (AAB genomic group) of banana (*Musa* spp.)

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ABSTRACT

Six Palayankodan ecotypes of banana belonging to AAB genomic group were evaluated for genetic variability among quantitative traits. Genetic and phenotypic coefficient of variation, heritability and genetic advance were estimated for eighteen traits that included plant height, pseudostem girth, number of leaves per plant, leaf width, number of suckers per plant, days taken from planting to shooting, total crop duration; length, girth, weight and volume of finger; hand weight, bunch weight, number of fingers per bunch, number of fingers per hand, ripe-fruit weight, sugar/acid ratio and pulp weight. Remarkable variability was observed among the collections for these characters. Bunch weight, number of fingers per bunch and number of suckers per plant with very high value of PCV, GCV, heritability and genetic advance makes it prime traits for direct selection. Plant height, pseudostem girth, total crop duration, sugar:acid ratio, finger length and days taken from planting to shooting with high value of heritability and moderate value of genetic advance. PCV are other important traits which need to be considered for selection. The volume of finger with low values for GCV, PCV, heritability and genetic advance as per cent of mean implies that it is highly influenced by environment and should not be taken as a criterion for selection. Plant height, total crop duration, sugar:acid ratio, finger length, pseudostem girth, number of fingers per bunch and days taken from planting to shooting showed high genetic advance and heritability and important characters to be considered for selection of ecotypes.

Key words: Banana, Palayankodan, ecotypes, heritability, genetic advance, PCV, GCV

INTRODUCTION

The primary objective of a crop improvement programme is to assess genetic variability existing in that particular crop the extent to which the character to be improved is heritable. Critical estimation of variability existing in the base population is a prerequisite for successful crop improvement through various plant breeding methods. Burton (1952) pointed out that calculating Genetic Coefficient of Variation (GCV) along with heritability could assess the best picture of amount of advancement to be expected by selection. Ramanujan and Thirumalachar (1967) suggested that heritability estimate in the broad sense is reliable if accompanied by high genetic advance. Johnson *et al* (1955) and Swarup and Chaugle (1967) also considered that heritability estimates along with genetic gain were useful and more reliable than heritability estimates alone in predicting selection response. Effectiveness of selection based on phenotypic performance can be more useful and reliable only when selection is based on heritability estimates combined with genetic gain. Above all these, knowledge of the extent of variability in germplasm is an essential prerequisite in any breeding programme. Banana (*Musa* spp.) is the most important fruit crop grown in the tropical and subtropical regions of India. Clones of 'AAB' genomic group occupy major banana growing area in India. This group comprises several popular desert types, of which, Palayankodan (syn. Mysore Poovan) is the most widely cultivated single clone because of its drought tolerance and suitability for ratooning (Rajeevan and Geetha, 1982). The vast difference in agroclimatic conditions under which the clone is grown in India, is likely to generate numerous mutants of the clone. However, only one mutant, namely 'Mottapoovan', has been reported so far. Progress of a breeding programme depends upon the extent to which desirable traits are heritable. High heritability estimate is

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used to predict the usefulness of traits in a selection programme. Hence, the present investigation was undertaken to study genetic variability, heritability and genetic advance in eighteen morphological traits of six Palayankodan banana ecotypes.

MATERIAL AND METHODS

The present experiment was carried out in the Department of Pomology and Floriculture, College of Agriculture, Vellayani, Thiruvananthapuram, Kerala. A total of six Palayankodan ecotypes procured from Banana Research Station, Kannara, Thrissur and College of Agriculture, Vellayani, Thiruvananthapuram, were planted and maintained at the College Orchard, College of Agriculture, Vellayani. Six ecotypes of Palayankodan banana were raised in Completely Randomized Block design (CRD) with five replications as per Panse and Sukhatme (1985). Cultural practices as per the Package of Practice Recommendations were followed (KAU, 1996). The six ecotypes of Palayankodan banana (all dessert type, having 3x ploidy and AAB genomic composition) are as follows: Palode Palayankodan, PKNNR, Chandra Bale, Pisang Ceylon, Mottapoovan Vellapalayankodan.

The genetic and phenotypic coefficient of variation, heritability and genetic advance were estimated for eighteen characters which included plant height, pseudostem girth, number of leaves per plant, leaf width, number of suckers per plant, days taken from planting to shooting, total crop duration, bunch weight, number of fingers per hand, number of fingers per bunch, hand weight; length, girth, weight and volume of finger, ripe fruit weight, pulp weight and sugar:acid ratio. Biometric data were collected and statistically analyzed following Fischer (1960). From the analysis of variance, genetic parameters like phenotypic and genotypic coefficient of variation (PCV and GCV) (Burton, 1952), habitability estimates (Burton and de Vane, 1953) and genetic advance (Allard, 1960) were calculated.

RESULTS AND DISCUSSION

Phenotypic and genotypic coefficients of variation for eighteen morphological characters of six Palayankodan ecotypes were studied. PCV were higher than their respective GCV for all the characters, which reflects influence of environment on phenotypic expression of these characters. Significant difference was recorded among ecotypes of banana for various plant parameters studied. Results presented in Table 1 indicate the range and general mean for each character studied. The highest range of variation was recorded for plant height, total crop duration, days taken from planting to shooting and number of fingers per bunch. The lowest range of variation was recorded for the number of suckers per plant, number of leaves per plant, number of fingers per hand, hand weight, bunch weight, finger length and girth of finger.

Table 1. Mean, range and coefficient of variation (CV) for eighteen traits in Palayankodan ecotypes of banana

Trait	Mean ± S.E.	Range	CV (%)
Plant height (cm)	311.03 ± 23.3	417.2 – 264.20	17.6
Pseudostem girth (cm)	65.80 ± 6.3	96.06 – 56.46	23.5
Number of leaves per plant	8.53 ± 0.7	11.20 – 6.80	20.3
Leaf width (cm)	78.98 ± 2.1	87.64 – 72.92	6.6
Number of suckers per plant	9.4 ± 1.5	15.80 – 6.20	39.0
Days taken from planting to shooting	227.87 ± 15.6	300.0 – 188.80	16.8
Total crop duration (days)	323.87 ± 17.3	407.0 – 286.80	13.1
Bunch weight (kg)	16.19 ± 1.9	23.04 – 10.60	28.8
Number of fingers per hand	16.90 ± 0.9	18.93 – 14.30	12.2
Number of fingers per bunch	189.73 ± 15.2	254.20 – 72.20	19.6
Hand weight (kg)	2.03 ± 0.2	2.80 – 1.50	22.0
Length of finger (cm)	11.09 ± 0.8	13.60 – 8.36	17.6
Girth of finger (cm)	9.76 ± 0.5	10.52 – 8.14	12.3
Finger weight (g)	99.38 ± 3.0	109.22 – 90.08	7.4
Volume of finger (cc)	92.49 ± 2.8	101.74 – 82.54	7.3
Ripe fruit weight (g)	82.80 ± 2.5	88.78 – 72.74	7.4
Pulp weight (g)	63.66 ± 2.6	68.30 – 52.32	9.9
Sugar:acid ratio	48.01 ± 4.3	68.53 – 37.53	22.2

Phenotypic and genotypic coefficients of variation, heritability and genetic advance are presented in Table 2. Highest PCV was observed for bunch weight (42.07%), followed by the number of suckers per plant (31.99%), hand weight (24.84%) and pseudostem girth (23.62%). Lowest PCV value was seen for leaf width (7.18%), followed by ripe fruit weight (7.78%). GCV ranged from 38.14 per cent for bunch weight to 6.27 per cent for volume of finger. Highest GCV was recorded for bunch weight (38.13%), followed by number of suckers per plant (27.94%), hand weight (21.18%) and sugar:acid ratio (22.03%). Work of Rajeevan and Geetha (1982) and Valsalakumari and Nair (1986) also supported this, with high estimates for GCV.

Table 2. Phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), environment coefficient of variation (ECV), heritability (broad sense) and genetic advance (GA) as percentage of mean for eighteen traits of Palayankodan ecotypes of banana

Trait	GCV (%)	PCV (%)	ECV (%)	Heritability (Broad sense) %	GA as % of mean
Plant height (cm)	17.58	17.68	1.86	98.90	36.02
Pseudostem girth (cm)	23.42	23.62	3.09	98.29	47.83
Number of leaves per plant	19.69	23.36	10.59	77.56	35.72
Leaf width (cm)	6.47	7.18	3.11	81.26	12.02
Number of suckers per plant	27.94	32.00	15.59	82.16	38.68
Days taken from planting to shooting	16.73	16.97	2.78	76.25	34.01
Total crop duration (days)	13.09	13.19	1.58	98.57	26.77
Bunch weight (kg)	38.13	42.07	17.77	97.31	71.21
Number of fingers per hand	9.77	10.58	4.01	85.38	18.60
Number of fingers per bunch	19.48	20.23	5.42	92.81	50.26
Hand weight (kg)	21.18	24.84	12.97	72.73	37.21
Length of finger (cm)	17.41	18.46	6.12	89.02	33.84
Girth of finger (cm)	11.84	14.01	7.47	71.53	20.63
Finger weight (g)	7.24	8.16	3.75	78.88	13.26
Volume of finger (cc)	6.27	10.42	8.32	36.20	7.77
Ripe fruit weight (g)	7.30	7.78	2.69	88.07	14.11
Pulp weight (g)	9.73	1.34	3.48	88.63	18.87
Sugar:acid ratio	22.03	22.73	5.74	93.63	43.91

Lowest GCV value was observed for volume of finger (6.27%), followed by leaf width (6.47%). Rajeevan and Geetha (1982) observed high PCV and GCV values for bunch weight, number of fingers per bunch and weight of finger for 40 banana cultivars. Valsalakumari and Nair (1986) reported highest PCV and GCV for bunch weight, hand weight, number of fingers per bunch, pseudostem girth and finger weight. The vast difference in PCV and GCV and very low estimates for GCV indicate an immense influence of environment on manifestation of this character. Similar findings were also made by Sreerangaswamy *et al* (1980) in banana. Significant difference between phenotypic and genotypic coefficients of variation for number of leaves per plant, number of suckers per plant, number of fingers per bunch, bunch weight, finger weight and volume of finger suggests that these characters were not influenced by environment. The estimates of heritability separate genetic variability from phenotypic variability and indicate possibility and the extent to which improvement can be brought about through proper selection. Moderate phenotypic and genotypic coefficients of variation were registered for plant height (17.68% and 17.58%), total crop duration (13.19% and 13.10%), number of fingers per bunch (20.22% and 19.49%), finger length (18.46% and 17.41%), finger girth (14% and 11.84%) and sugar:acid ratio (22.76% and 22.02%), respectively. These characters offer much scope for improvement by selection and hybridization. Heritability, in a broad sense, gives the amount of heritable portion of a character. Environmental coefficient of variation was high in bunch weight (17.78%) and the lowest in total crop duration (1.58%).

Characters possessing high heritability can be improved directly through selection, as, these are relatively less affected by environment. The magnitude of heritability indicates effectiveness of selection based on phenotypic performance (Johnson *et al*, 1955). In the present study, all traits exhibited high heritability. This ranged from 72.53% for leaves per plant, to 98.90% for plant height. Characters like plant height (98.90%), total crop- duration (98.57%), pseudostem girth (98.29%), bunch weight (97.31 %), number of fingers per bunch (92.81%), sugar:acid ratio (93.63%) and length of finger (89.02%) show high heritability. Relatively higher values of heritability for these characters imply that a large proportion of phenotypic coefficient of variance (PCV) was attributable to the genotypic coefficient variance (GCV). High heritability values for number of fingers per bunch, plant height, days taken from planting to shooting and number of fingers per bunch obtained in the present study are in agreement with findings of Sreerangaswamy *et al* (1980) in banana. High heritability has also been reported for pulp weight and length of finger (Singh and Sharma, 1997), pseudostem girth, bunch weight, finger length and number of fingers per bunch (Rajeevan and Geetha, 1982), plant height, days taken from planting to flowering, finger length, sugar:acid ratio, bunch weight and pesudostem girth (Valsalakumari and Nair, 1986), crop duration (Rosamma and Namboodiri, 1990) and bunch weight, plant height and crop duration (Uma *et al*, 2000). Katiyar *et al* (1974) demonstrated that heritability values alone are inadequate to cannot be taken as a tool to calculate the amount of genetic progress achieved by selecting the best individual. Ramanujam and Thirumalachar (1967) opined that heritability estimates could be reliable if accompanied by a high genetic advance.

In the present investigation, there was wide variation among characters for genetic advance. Genetic advance as per cent of mean, varied from 7.77% for volume of finger to 71.21% for bunch weight. Characters like bunch weight (71.21%), plant height (36.02%), pseudostem girth (47.83%), days taken from planting to shooting (34.01%), number of fingers per bunch (50.26%), sugar:acid ratio (43.91%), hand weight (37.21%), number of suckers per plant (38.68%) and finger length (33.84%) showed higher genetic advance, along with high heritability. This clearly suggests that these characters are mainly of the additive type as reported by Johnson *et al* (1955). Lowest genetic advance was obtained for volume of finger (7.77%), leaf width (12.02%), ripe fruit weight (14.11%), number of fingers per hand (18.61%) and pulp weight (18.87%). Number of suckers per plant and bunch weight with high value for PCV, GCV and heritability, coupled with genetic advance, indicate that the character is predominantly controlled by additive gene action. This is supported by the hypothesis proposed by Panse (1957) suggesting that characters exhibiting high heritability and GA were governed by additive gene effects. Similar results were reported by Rosamma (1982) and Uma *et al* (2000). This implies that selection of bunch weight, number of fingers per bunch and number of suckers per plant can bring about effective improvement, and may be exploited in breeding programmes. High heritability does not necessarily mean a high genetic advance for a particular character (Allard, 1960). Heritability, along with genetic advance, is more useful than heritability alone in predicting the result and effect of selecting the best individuals (Johnson *et al*, 1955). Uma *et al* (2000) reported plant height, with very high value of heritability and moderate value of genetic advance, revealed relatively low influence of environment on the trait for silk ecotypes of banana. In a study with 48 banana varieties, falling under different genomic group traits such as weight of finger, number of fingers per bunch and bunch weight, recorded a high estimate of heritability along with high genotypic gain in the crop (Rosamma, 1982). In the present study, plant height, with high value of heritability (98.92%) and moderate value of genetic advance (36.02%) revealed relatively low influence of environment on this trait. A study by Uma *et al* (2000) also revealed that plant height had high value of heritability (91.11%) and moderate genetic advance (27.54%). Sugar:acid ratio, pseudostem girth, days taken from planting to shooting, number of fruits per bunch and finger length showed high values of heritability, coupled with moderately high genetic advance, indicative the influence of environment on expression of these characters to some extent and, that, rigid selection might bring about improvement in these traits. Though ripe fruit weight, number of fingers per hand, leaf width, finger width and finger weight showed moderate estimate of heritability, lower value of genetic advance reflected favourable influence of environment rather than that of the genotype, and simple selection may not be rewarding. Thus it may be suggested that improvement is likely to be very effective for these characters in banana.

ACKNOWLEDGEMENT

The authors gratefully acknowledge Kerala Agricultural University, Thrissur, Kerala, India, for providing funds and facilities to carry out the research work.

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(MS Received 4 May 2009, Revised 21 June 2010)

J. Hortl. Sci.
Vol. 5 (2): 114-116, 2010

Comparative performance of mango varieties grafted on Vellaikolamban and mixed rootstock

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ABSTRACT

Research on rootstock in mango is very limited in our country. Kalapady was reported to be a dwarfing rootstock. Recent trend among mango growers is to high density orcharding with dwarfening nature of the varietie. Efforts were made at Agriculture Research Station, Mulde, to study comparative performance of Ratna, Alphonso and Kesar mango on Vellaikolamban and mixed rootstock i.e., heterozygous seedling stock and the effect of rootstock on a scion under high density of 5m x 5m spacing. Results indicated that use of Vellaikolamban rootstock reduced plant volume in scion cv. Alphonso by 39.1%, followed by 24.9% in Ratna and 26.5% in cv. Kesar. As volume of the canopy was reduced, it directly influenced fruit yield cvs. Alphonso and Ratna. However, reduction in canopy volume had a positive influence on yield in cv. Kesar. Net returns of Rs.38,629/- per ha were maximum for Kesar with the rootstock Vellaikolamban.

Key words: Mango, Ratna, Alphosno, Kesar, Vellaikolamban, Mixed rootstock, polyembryonic, dwarfing, volume, yield

INTRODUCTION

Konkan region of Maharashtra is a traditional belt of mango cultivation particularly, cv. Alphonso which occupies an area of 1,64,000 ha. Though Alphonso is the chief commercial variety of the region, cultivars Kesar and Ratna are gaining popularity with the concept of high density orcharding. However, the weather of the region favours crop viguor. Wide variation in performance of the same variety within an orchard using grafts prepared out of heterozygous seedling stocks restricts establishment of high density orchards.

Rootstock research work in mango is still in its infantly. Good work was done on selection criteria for dwarfness, way back in 1985 at IARI (Bose, 1985). Studies conducted at various places indicated that the varieties Kalapady (Sen, 1939), Olur, Ambalavi (Jauhari *et al*, 1972), Vellaikolamban (Singh and Singh, 1976) and Belkhas and Parikhas (Mukherjee and Das, 1976) have the potential for imparting dwarfness. Avilan *et al* (1996) reported influence of rootstock on fruit size and shape of the grafted cultivars, showing strong scion/rootstock relationship. Singh and Singh (1976) recorded maximum reduction in height of Dashehari scion when grafted to Vellaikolamban rootstock. With this in view, the present study was carried out to study comparative performance of Ratna, Alphonso and Kesar mango varieties on Vellaikolamban and mixed rootstock, i.e., heterozygous seedling stock, and, to study the effect of rootstock within a scion variety under a high density orchard with a spacing of 5m X 5m.

MATERIAL AND METHODS

The present study on comparative performance of different, leading varieties namely Alphonso, Ratna and Kesar grafted onto Vellaikolamban and mixed rootstock under high density viz., 5m x 5m spacing was carried out at Agriculture Research Station, Mulde, from September, 1992 to May, 2007. The experimental station is located at 16°2' latitude, 73°42' longitude and at 17m above msl in Konkan region of Maharashtra, which is a coastal region with annual rainfall of 3000mm. Soils are well drained sandy loam with pH 6.01. Treatment combinations are detailed below:

T_1 Ratna on Vellaikolamban
T_2 Ratna on Mixed rootstock (heterozygous seedling stock)
T_3 Alphonso on Vellaikolamban
T_4 Alphonso on Mixed rootstock (heterozygous seedling stock)
T_5 Kesar on Vellaikolamban
T_6 Kesar on Mixed rootstock (heterozygous seedling stock)

T_4 trial was laid out in Randomized Block Design with five replications and two plants per replication as a unit. The experimental material was prepared by stone grafting and one year old grafts were planted at 5m x 5m

spacing during September, 1992. Annual growth (height and spread) was recorded in the month of May every year since 1994. However, data on growth and yield from year 2003 to 2006 only have been used here. Low spreading branches upto 60cm height above ground and overcrowded branches in the canopy of the tree were chopped by way of light pruning in the year 2004. Plant volume was calculated using the following formula:

Plant Volume (m^3) = Πr^2 x h

Where h = plant height and r = $\frac{E - W + N - S \text{ spread}}{4}$

Data on growth and yield attributes were subjected to statistical analysis (Panse and Sukhatme, 1989).

Cost of cultivation was calculated using standard cost concepts applied by Gorivale *et al* (1997*)* and Nikam *et al* (2004).

RESULTS AND DISCUSSION

Growth and yield observations for the years 2003 to 2006 are presented in Table 1.

It is evident from the data (Table 1) that significant difference between treatments was observed for pooled data on plant height, plant volume and yield. Grafts of Ratna variety on Vellaikolamban showed significantly lower plant height (3.6 m) compared to Kesar on Vellaikolamban and mixed rootstock (4.6 m), Alphonso on mixed rootstock (4.7 m) and was on par with Alphonso on Vellaikolamban, and Ratna, on mixed rootstock. Effect of rootstock on plant height within a scion variety was significant only in Alphonso variety. Alphonso grafts on Vellaikolamban showed marked reduction in plant height (3.8 m) over the mixed rootstock (4.7 m). Singh and Singh (1976) recorded maximum reduction in plant height in Dashehari grafted on Vellaikolamban rootstock. However, rootstock did not show any effect on plant height in Kesar variety.

Data on average plant volume from 2003 to 2006 and pooled data over the years revealed that Ratna grafts on Vellaikolamban had reduced plant volume compared to that in other treatments. Similarly, grafts of all varieties on Vellaikolamban rootstock showed reduction in plant volume over the mixed rootstock.

Data on average plant volume pooled over the years revealed that Ratna variety on Vellaikolamban had recorded the lowest plant volume (285.9 m^3) compared to the grafts on mixed rootstock and other scion varieties, irrespective of the rootstock. Maximum plant volume (651.5 m^3) was recorded in 'Kesar' on mixed rootstock.

In the present study, reduction in plant volume was observed in cv. Alphonso (39.1%), followed by 'Kesar' (26.5%) and 'Ratna' (24.9%) when grafted onto Vellaikolamban rootstock. These results are in line with earlier results reported by Avilan *et al* (1996), Singh and Singh (1976) and Reddy *et al* (2003). Vellaikolamban rootstock not only important reduced plant volume to the scion variety, but four year pooled yield data revealed that it also reduced the yield by 24.4% in 'Ratna' and 21.6% in 'Alphonso' scions. Similar effect of Vellaikolamban rootstock on yield of Dashehari plants was reported by Singh and Singh (1976), whereas, Reddy *et al* (2003) reported higher yields with the dwarfing Vellaikolamban rootstock. However,

Table 1. Growth and yield of 'Alphonso', 'Kesar' and 'Ratna' mango varieties grafted onto Vellaikolamban and mixed rootstock during 2003 - 2006 and data pooled over the years

Treatments	Pooled mean height (m)	Pooled over the years (m^3)				Pooled over the year (m^3)	% Reduction in volume	Yield (t/ha)				Pooled over the	% Decrease/ increased in yield over mixed rootstock
		2003	2004	2005	2006			2003	2004	2 005	2006		
Ratna / Vellaikolamban	3.6	289.0	263.3	270.6	320.7	285.9	24.9	13.4	4.2	2.8	3.1	5.9	(-) 24.4
Ratna / Mixed rootstock	3.7	360.8	334.6	329.5	498.0	380.8		13.8	5.8	4.4	6.9	7.8	
Alphonso / Vellaikolamban	3.8	306.1	303.1	270.7	414.2	323.5	39.1	5.1	1.7	3.9	1.0	2.9	(-) 21.6
Alphonso / Mixed Rootstock	4.7	582.6	465.4	483.3	593.9	531.3		7.4	2.6	3.8	1.1	3.7	
Kesar / Vellaikolamban	4.6	512.4	397.2	406.1	599.5	478.8	26.5	17.8	8.0	4.5	3.9	8.6	(+) 10.3
Kesar/ Mixed rootstock	4.6	679.5	569.0	576.0	781.4	651.5		15.7	8.5	3.6	3.2	7.8	
C.V.	8.2	28.1	27.7	28.7	32.0			44.0	35.3	75.2	50.6		
SE $\pm$	0.17	57.3	48.2	50.0	76.5	27.9		2.4	0.8	1.3	0.7	0.8	
CD *(P=0.05)*	0.5	169.0	142.2	147.4	225.7	78.4		7.1	2.4	N. S.	2.1	2.3	

Table 2. Cost of cultivation (Rs./ha) of mango varieties on rootstocks

Particulars	R x V	R x M	A x V	A x M	K x V	K x M
Hired labour	21,775	21,775	21,775	21,775	21,775	21,775
Manures and fertilizers	18,640	18,640	18,640	18,640	18,640	18,640
Plant protection	7,000	7,000	7,000	7,000	7,000	7,000
Input cost (Rs.)	47,415	47,415	47,415	47,415	47,415	47,415
Depreciation on implements and machinery	500	500	500	500	500	500
Land revenue & other cesses	50	50	50	50	50	50
Interest on working capital for 12 months @ 13%	7,682	7,682	7,682	7,682	7,682	7,682
Interest on fixed capital @ 10%	500	500	500	500	500	500
Rental value of land ($1/6^{th}$) of the gross value. land revenue	14,700	19,400	9,733	12,400	21,400	19,400
Amortization value	7,482	7,332	7,482	7,332	7,482	7,332
Supervision charges @ 10% of input cost	4,742	4,742	4,742	4,742	4,742	4,742
Total cost (Cost C)	83,071	87,621	78,104	80,621	89,771	87,621
Yield (t/ ha) and Gross Returns	5.88	7.76	2.92	3.72	8.56	7.76
Sale price of 'Alphonso @ Rs.25/kg Ratha and kesar @ Rs.15/kg	88,200	1,16,400	73,000	93,000	1,28,400	1,16,400
Net returns at Total Cost	5,129	28,779	(-) 5,104	12,379	38,629	28,779
Benefit:Cost ratio	1.06	1.32	0.93	1.15	1.43	1.33

R= 'Ratra', V= 'Vellaikolumban', A= 'Alphonso', M= Mixed rootstock', K= Kesar

Vellaikolamban rootstock showed beneficial effect on 'Kesar' variety, where the yield increased by 10.3% over mixed rootstock.

Per hectare cost of cultivation of different mango varieties grown on Vellaikolamban and mixed rootstock is presented in Table 2. Data reveale that 'Ratna' variety on mixed rootstock exhibited 1.32 benefit to cost ratio as against 1.06 in 'Ratna' on Vellaikolamban rootstock. Cultivation of 'Alphonso' on Vellaikolamban suffered a loss by recording only 0.93 benefit to cost ratio compared to 'Alphonso' on mixed rootstock (1.15). Maximum net returns (Rs. 38,629/ -) per hectare were recorded in 'Kesar' on Vellaikolamban, exhibiting B:C ratio of 1.43 as against that on mixed rootstock with, Rs. 28,779 and 1.33 B:C ratio.

The present study revealed that plant height, plant volume and yield decreased by use of Vellaikolamban rootstock in 'Alphonso' and 'Ratna' whereas, Vellaikolamban rootstock reduced the plant volume of 'Kesar' variety with increased per hectare yield under high density planting of 5m x 5m.

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(MS Received 9 March 2010, Revised 26 October, 2010)

J. Hortl. Sci.
Vol. 5 (2): 117-119, 2010

High density planting in mango cv. Alphonso

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ABSTRACT

A trial was conducted to optimize spacing for high density planting in mango cv. Alphonso to obtain higher yield/ unit area at the Agriculture Research Station, Mulde, during 2006-07 to 2008-09 with four close spacings and one normal spacing as control. Highest yield (6.4 MT/ha) was recorded with a spacing of 5 m x 5 m without reduction in fruit size in 10 year old plants compared to the mean yield of 1.12 MT/ha in 10m x 10m normal spacing. High density plantation helped to get significantly higher yield per unit area compared to the normal spacing, without affecting size and quality of mango fruits. The highest cost:benefit ratio (2.33) was recorded in high density plantation of 5m x 5m, with maximum net returns of Rs.1,12,000/- per hectare. The present findings show promise for more yield and returns per unit area during the initial years of mango plantation by adopting 5m x 5m high density planting.

Key words : Mango, Alphonso, spacing, high density planting

INTRODUCTION

Alphonso mango (*Mangifera indica* L.) is a leading cultivar grown commercially in the Konkan region of Maharashtra and occupies an area of 1,64,000 ha. The variety is highly preferred for export. But, alternate bearing and low productivity (3.0 t/ha) realized with normal spacing gives low net returns to the farmer. To overcome this constraint, a trial was conducted at the Agriculture Research Station, Mulde with five different spacings during the period 1997 to 2009. Efforts were made to accommodate higher number of plants per unit area so as to get higher yield from the mango plantation during the initial period of orchard development. It takes at least 15 to 20 years to cover all the area with canopy in a mango orchard. This leads to low net returns to the grower. As a result, there is a feeling among mango growers that mango cultivation is not economical. To increase net returns per unit area of mango cultivation, this trial was undertaken. The major objective of the study was to optimize spacing for high density planting to obtain higher yields per unit area during the early period of the plantation.

MATERIAL AND METHODS

The study was conducted at Agriculture Research Station, Mulde, Sindhudurg District, Maharashtra State. The trial was laid out in Randomized Block Design, with five replications. The soil was red laterite, with pH range 5.5 to 6.5, and was rich in iron content. Soil nutrient status of this experimental plot was as follows : N (2.24%), P (0.10%), K (0.72%) and minor nutrients Zn (63.7 ppm), Cu (12.1 ppm), Fe (72.70 ppm) and Mn (68.8 ppm).

Average rainfall in this region is 3000–4000 mm, with relative humidity of 85–90%. Maximum average temperature is 35^{O}C and minimum average temperature 16^{O}C, with average sunshine hours of 9.00 h. Weather conditions are ideally suited to mango cultivation.

Five spacings used for the planting were: 1) 2.5m x 10m, 2) 5m x 5m, 3) 5m x 7.5m, 4) 5m x 10m and 5) 10m x 10m. Unit area /treatment /replication and details of number of plants /treatment are given below:

Treatment	Spacing	Number of plants/ plot/ treatment	Number of replications	Total number of plants/ treatment
T_1	2.5m x 10m	20	5	100
T_2	5m x 5m	20	5	100
T_3	5m x 7.5m	13	5	65
T_4	5m x 10m	10	5	50
T_5	10m x 10m	5	5	25

The trial was conducted under rainfed conditions. Planting was done during 1997. Plants were given recommended fertilizer doses and prophylactic measures (with standard dose of paclobutrazol) were practised during July – August every year. Age of the trees was ten years. Regular pruning of overcrowded branches was done in the trial. Three vegetative flushes occurred in June, October and March every year which led to luxuriant growth. Observations were recorded at fortnightly intervals. To get better yield during the initial years, pruning of dead, diseased, weak and intermingling branches of mango plants was done at the age of eight years. Observations on vegetative growth, flowering and number of fruits/plant and average yield kg/plant were recorded and yield expressed as metric tons/ha. Data reported here is the average of three years (2006-07 to 2008-09) and was statistically analyzed as per Panse and Sukhatme (1985) for Randomized Block Design. Mean values are reported for the physico chemical properties like fruit length, breadth, size, weight, TSS, acidity, stone, peel, pulp ratio and shelf life.

RESULTS AND DISCUSSION

Vegetative parameters

Vegetative parameters of plants under different treatments are presented in Table 1. Plant height was found to be significantly higher (9.10m) with 2.5m x 10m treatment, whereas normal spacing (6.99m) was at par with the spacing 5m x 7.5m (9.05m). No significant differences were observed with respect to plant girth and spread among the treatments. In high density planting natural tendency of the plant is to put forth vertical growth rather than horizontal, due to mutual shading of plants. These findings are in line with earlier reports of Ram *et al* (1996) and Gunjate *et al* (2003).

Table 1. Vegetative parameters in high density orchard of 'Alphonso' mango

Treatment	Spacing	No of trees/ha	Tree height (m)	Tree girth (cm)	Tree spread (cm)	
					E –W	N – S
T_1	2.5 m x 10 m	400	9.10	59	4.85	4.60
T_2	5 m x 5 m	400	7.12	57	5.12	5.30
T_3	5 m x 7.5 m	267	9 .05	51	4.75	4.63
T_4	5 m x 10 m	200	7.80	53	5.10	5.05
T_5	10 m x 10 m	100	6.99	61	6.70	5.78
	SEm±		0.31	0.81	0.20	0.24
	CD *(P=0.05)*		0.93	NS	NS	NS

* The figures are average/mean values of three years' data (2006-07 to 2008-09)

Flowering and fruit yield parameters

High density planting with 2.5m x 10m (T_1) spacing recorded a mean of 42 fruits/tree during both years and average fruit yield of 16.9 kg/tree, also in the same treatment. Maximum fruit yield (6.4 t/ha) was recorded in 5m x 5m spacing, whereas, normal spacing recorded the lowest fruit yield (1.12 t/ha). All the high density treatments recorded higher fruit yield compared to normal spacing. Maximum fruit yield (6.4 t/ha) in 5m x 5m spacing was due to higher number of plants and maximum number of fruiting branches. It was seen that under the Konkan agroclimatic zone, the hot and humid climate favours luxuriant growth of cv. Alphonso. During the initial years, high density orcharding with 2.5m x 10m, 5m x 5m and 5m x 7.5m spacings appears promising. These results are in line with those reported by Ram *et al* (1996) in 'Dashehari' mango. More number of plants /unit area resulted in more number of fruits/plant, higher yield/ha, and thereby, more tonnage from the same unit area. These results are similar to those reported earlier by Gunjate *et al* (2003) and Nath *et al* (2007).

Fruit quality

Data on physico chemical properties are presented in Table 3. The study on fruit quality attributes of 'Alphosno' mango showed that maximum fruit weight (248g) was recorded in the spacing 2.5m x 10m, which was significantly

Table 2. Flowering and yield parameters in 'Alphonso' mango as influenced by high density planting

Treatment	Spacing	No. of trees/ha	Flowering (%)		No. of fruits/tree		Average. fruit yield (kg/tree)		Average. yield (t/ha)
			2008	2009	2008	2009	2008	2009	
T_1	2.5 m x 10 m	400	31.80	30.83	32	52	8.3	13.1	4.280
T_2	5 m x 5 m	400	27.52	29.83	45	89	11.2	22.5	6.400
T_3	5 m x 7.5 m	266	16.50	15.83	39	74	9.7	18.4	3.737
T_4	5 m x 10 m	200	07.83	08.33	43	71	10.2	18.0	2.820
T_5	10 m x 10 m	100	15.80	15.00	31	48	8.0	12.3	1.12
	SEm±		2.21	2.57	1.1	1.4	0.42	0.79	0.19
	CD *(P=0.05)*		6.72	7.60	3.4	4.1	1.3	2.5	0.67

Table 3. Fruit quality attributes of 'Alphonso' mango under high density planting during Year 2009

Treatments	Spacing	Fruit length (cm)	Fruit breadth (cm)	Pulp weight (g)	Stone weight (g)	Peel weight (g)	Fruit weight (g)	Pulp: stone ratio	TSS(^{0}B)	Acidity(%)	Shelf life at room temperature (days)
T_1	2.5 m x 10 m	7.0	6.8	122.0	29.0	40.0	248.0	3.2	18.0	0.30	13
T_2	5 m x 5 m	7.0	7.0	120.0	35.0	39.0	194.0	3.2	18.0	0.29	14
T_3	5 m x 7.5 m	6.5	6.5	119.6	38.0	39.4	197.0	3.1	16.75	0.35	17
T_4	5 m x 10 m	8.0	7.0	122.2	38.0	39.8	200.0	3.2	17.00	0.29	18
T_5	10 m x 10 m	6.5	6.2	123.0	34.0	40.0	243.0	3.2	19.50	0.28	15
	SEm±	0.4	0.8	3.2	0.6	0.8	4.3	0.2	0.7	0.6	0.1
	CD *(P=0.05)*	N.S.	N.S.	N.S.	N.S.	N.S.	12.9	N.S.	N.S.	N.S.	0.3

N.S.=Non-Significant

Table 4. Cost: benefit ratio under high density planting in 10-year old Alphosno mango trees

Treatment	Expenditure/ ha (Rs.)	Receipts realized (Rs.)*	Net profit/ha	C:B ratio
T_1	48,000	1,0,7000	59,000	1.23
T_2	48,000	1,60,000	1,12,000	2.33
T_3	42,640	93,425	50,785	1.19
T_4	40,000	70,500	30,500	0.76
T_5	36,000	28,000	-8,000	-0.22

*Fruits were sold @ Rs. 25/kg

superior over the spacing 5m x 10m, and was at par with normal spacing (10m x 10m). The rest of quality parameters like TSS, acidity, pulp to stone ratio, etc., did not show significant differences between treatments. These results show that a closer spacing and high density mango plantation does not influence or hamper the quality of fruit. Regular training and pruning helps generate good aeration, thus ensuring better quality. The present findings are in line with earlier reports of Krishna *et al* (2003) and Gunjate *et al* (2003).

Cost : Benefit ratio

Data in Table 4 show that maximum net returns (Rs.1,12,000/-) and cost benefit ratio (2.33) was recorded in the spacing 5m x 5m, whereas, normal spacing of 10m x 10m fetched lower net returns (Rs.8,000/-) and cost: benefit ratio (0.22). Normal spacing 10m x 10m may have yielded lower net returns as the trees were 10 years old and 'Alphonso' orchards become profitable only after 15 years. These findings indicate that high density planting of 'Alphonso' mango not only gives higher yield/unit area during the initial years, but also promises higher net returns subsequently.

Though the present findings are based on three years yield data these sufficiently indicate that high density plantation in 'Alphonso' mango with 5m x 5m spacing is helpful for getting higher yield and more net returns/unit area.

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(MS Received 14 March 2010, Revised 7 October, 2010)

J. Hortl. Sci.
Vol. 5 (2): 120-123, 2010

Effect of pruning intensity on leaf tissue micronutrient status in three mango (*Mangifera indica* L.) cultivars under high density planting

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ABSTRACT

An experiment was conducted to study the effect of pruning on leaf micro nutrient (Cu, Zn, Fe and Mn) status in non-floral and floral shoots of three mango cultivars ('Amrapali', 'Mallika' and 'Dashehari') under high density planting during 2005-2007. All the three cultivars differed significantly in Cu, Zn, Fe and Mn content in leaves of non- floral as well as floral shoots. Pruning showed marked influence only on Cu and Zn content in the leaves of non- floral and floral shoots. Leaf nutrient status in terms of Fe and Mn also varied in cultivars irrespective of pruning intensity, and pruning did not have significant impact on Fe and Mn status in leaf tissue. Non-floral shoots had greater concentration of Cu and Zn than floral shoots in both the years of experiment. Highest Cu, Fe and Mn content was recorded in 'Mallika' mango, while, Zn content was highest in 'Dashehari' mango. Severe pruning (90 cm from apex) improved Cu and Zn content in leaves of non-floral shoots as well as floral shoots. The lowest amount of Cu and Mn was noted in 'Dashehari' leaves, while, 'Amrapali' had the lowest Zn and Fe content in both non-floral and floral shoots. Severely pruned 'Mallika' trees registered the highest amount of Cu, while lightly pruned 'Dashehari' trees had highest Zn content in their floral and non-floral leaves. Moderate pruning in' Mallika' enhanced Mn content in leave of non-floral and floral shoots. No-pruning in 'Dashehari' trees led to lower Cu content but Zn content was the least in lightly pruned 'Amrapali' trees. Severe pruning in 'Dashehari' trees drastically reduced Mn content. Thus, severe pruning in old mango trees may be advisable to improve micronutrient status in floral and non floral shoots.

Key words: Mango, *Mangifera indica*, pruning, micronutrients, Cu, Zn, Fe, Mn

INTRODUCTION

Mango (*Mangifera indica* L.), member of the family Anacardiaceae, is the most important fruit crop of both subtropical and tropical regions of the world. There is ample scope for enhancing production and productivity of mango through pruning under high density planting (HDP). Pruning is also practised to avoid overlapping/ intermingling of branches, improve light interception, increase photosynthetic rate, reduce relative humidity within the plant canopy, etc. (Lal *et al*, 2000). Not much work has been reported on determining optimum pruning intensity in close spaced orchards compared to wider spaced (traditional) ones. The practice of mango pruning is followed immediately after harvest (heading back branches) which encourages shoot growth just beneath the site of the first bud break (Sauco, 1996). These shoots [newly emerged] have different physiological responses post-pruning, i.e., changes in biochemical, physiological and nutritional status, which subsequently affect overall performance of the trees in the long run. Pruning decreases yield in the initial years due to simulative growth of shoots, while minerals absorbed by roots are readily available to a few fruits only (Mika, 1986). Root shortening coupled with stem pinching, followed by spray of PBZ or TIBA on shoots is the most effective treatment enhancing root and shoot branching and also for increasing leaf content of N, Ca, Mg, Fe and Zn (Helail and Eissa, 1997). Hence, the present work was carried out to study effect of pruning intensity on micronutrient status in leaf tissues of mango obtained from non-floral (vegetative) and floral (reproductive) shoots, which may reflect futurie performance of trees, especially under high density planting.

MATERIAL AND METHODS

The field experiment was conducted at the Main Orchard of the Division of Fruits and Horticultural Technology, IARI, New Delhi, during 2005-2007. Three mango cultivars, *viz*., 'Amrapali' (23-year-old), 'Mallika' (24-year-old) and 'Dashehari' (26-year-old) were selected

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for the present study. These cultivars were planted under high density with spacings of 2.5m x 2.5m, 3.0m x 4.0m and 3.0m x 2.0m for cvs. Amrapali (V_1), Mallika (V_2) and Dashehari (V_3), respectively. Trees were provided with uniform agronomic and cultural practices during the course of investigation. Pruning was done in mid August, 2005 and pruning intensities were**:** I0 (Control): un-pruned, I1 (Light): 30 cm from the apex, I2 (Moderate): 60 cm from the apex, and I3 (Severe): 90 cm from the apex. Each cultivar had three replications with four levels of pruning intensities. Thus, the total number of treatment combinations was 12, with one tree per replication. Balanced pruning was performed in all directions by removing the inner and a few peripheral branches of the canopy that were dense and overcrowded. The control trees were maintained without pruning. As a result of pruning, trees did show some flowering and fruiting during 2006, i.e. the first year (presumed to be an 'off' year) and the second year (2007, the 'on' year). The leaves (7-8 month old) from non-flowered (vegetative), flowered (reproductive) shoots were collected from all directions and immediately shifted to the laboratory where these were washed quickly and rinsed with distilled water. The samples were air dried, cut into small pieces and oven dried at 70^0C for 48h in paper bags until gaining constant weight and milled to a powder in a stainless steel grinder. The powder was stored in paper bags at room temperature. The powdered plant material (500 mg) was digested in 20 ml di-acid mixture [nitric acid (HNO_3):perchloric acid ($HClO_4$) 3:1] and the volume was made up to 100 ml with distilled water. Micro-nutrient concentration was determined on an atomic absorption spectrophotometer directly from the di-acid digest, using an air-acetylene flame. Content of Cu, Fe, Mn and Zn was measured at 386 nm (Lamp current 7 mA), 22.6 nm (Lamp current 3 mA), 403.1 (Lamp current 5 mA) and 213.9 nm (Lamp current 5 mA) wavelength, respectively. The sensitivity was 0.05, 0.008, 0.02, and 0.025 µg/ ml for Fe, Zn, Mn and Cu, respectively. Final concentration (in ppm) was calculated by multiplying the concentration with a suitable dilution factor. Experimental data were subjected to statistical analysis in factorial Randomized Block Design and two years data from non-floral and floral shoots were analyzed as per methods suggested by Gomez and Gomez, 1984. Interpretation of results was based on 'F' test and critical difference (CD) at P=0.05 was worked out for comparing means.

RESULTS AND DISCUSSION

Role of micronutrients in plant nutrition is vital because several deficiency symptoms occur in plants due to which performance of the entire tree declines markedly. Although micronutrient deficiencies produce characteristic symptoms, the symptoms are very confusing under field conditions, especially, when more than one nutrient is deficient. Mango cultivars, irrespective of pruning intensity, had significantly different concentrations of Cu, Zn, Fe and Mn in the leaves in the 'off' as well as the 'on' year of our experiment. Highest concentration of Cu and Mn was observed in 'Mallika', and lowest in 'Dashehari' (Table 1) which may be due to the biennial nature of 'Dashehari' mango (Thakur *et al*, 1981). It was also noted that in the 'on' year, Cu and Mn content leaves was lower than in the 'off' year (Thakur *et al*, 1973) because fruiting terminals numbered more in the 'on' year than in the 'off' year which acted as a sink for mineral nutrients (Thakur *et al*, 1981). Similarly, pruning intensity showed marked influence on Cu and Zn content in mango leaves. Severely pruned trees (I_3) had the highest Cu content, followed by moderately pruned (I_3) trees and the least Cu content was observed in un-pruned trees (I_0), as, pruning destabilizes the root:shoot ratio. In addition, defoliation along with root pruning and stem pinching invariably increases Cu content in shoots as noted by Helail and Eissa, 1997. In contrast, Mn content in leaves did not differ significantly (Table 1). Content of Zn in mango leaves improved after severe pruning (I_3), followed by light pruning while, moderate pruning reduced Zn level in mango leaves of both non-floral and floral shoots.

The interaction effect of cultivar and pruning intensity also affected Cu (except flowered shoots in the 'off' year), Zn and Mn content in leaves of non-floral and floral shoots. Cu and Mn content were highest in severely (V_2I_3) and moderately pruned 'Mallika' (V_2I_2) trees, respectively, while the lowest Cu concentration was estimated in un-pruned 'Dashehari' mango. In contrast, severely pruned 'Dashehari' had the lowest Mn in leaves (Table 2). Cultivar 'Mallika' encouraged greater vegetative growth (and produced substantial number of non-fruiting terminals in the beginning) /and non-fruiting terminals had higher Cu and Mn content (Thakur *et al*, 1979). Un-pruned trees had slow growth (less number of new shoots), thus resulting in deficiency of Cu.

Among the three cultivars, 'Dashehari' leaves had highest Zn content (Kumar *et al*, 1985), while 'Amrapali' had the lowest concentration of both Zn and Fe due to continuous production of fruiting terminals in both the years of experiment (Thakur *et al*, (1981). On the other hand, severely pruned tree had the lowest Zn content (26.66, 23.25; 25.64, 22.35 ppm) probably due to higher number of new

Table 1. Effect of cultivar and pruning intensity on micronutrient content in leaves of three mango cultivars under high density planting

Treatments†	Cu (ppm)				Zn (ppm)				Fe (ppm)				Mn (ppm)			
	2005-06*		2006-07**		2005-06*		2006-07**		2005-06*		2006-07**		2005-06*		2006-07**	
	NFS	FS	NFS	FS	NFS	FS	NFS	FS	NFS	FS	NFS	FS	NFS	FS	NFS	FS
Amrapali (V_1)	18.78	15.58	16.00	13.07	19.44	16.40	18.59	15.59	201.11	126.50	162.78	146.86	100.50	95.84	88.62	85.10
Mallika (V_2)	22.80	18.92	20.02	16.80	22.12	19.39	21.39	18.62	238.62	218.92	215.05	196.02	113.45	110.27	102.33	101.57
Dashehari (V_3)	14.30	10.72	12.27	8.83	30.90	26.91	29.60	26.01	202.85	181.36	174.75	156.44	76.42	73.75	67.18	63.62
SEm ±	0.41	0.29	0.32	0.97	0.80	0.74	0.82	0.79	11.85	12.02	13.31	13.64	5.09	5.00	5.45	5.12
CD *(P=0.05)*	1.18	0.86	0.93	0.80	2.31	2.14	2.37	2.20	34.05	34.33	38.22	45.24	14.64	14.60	15.60	14.71
Un-pruned (I_0)	17.17	13.56	14.27	11.23	23.67	20.34	22.42	19.71	208.46	183.96	174.96	156.84	92.14	88.83	80.91	81.24
30 cm (I_1)	18.13	14.40	15.20	12.10	25.33	21.97	24.08	20.83	196.25	175.94	172.21	134.20	102.07	99.26	92.14	88.83
60 cm (I_2)	18.52	15.02	16.21	13.00	20.95	18.03	20.62	17.41	237.23	216.13	197.53	182.88	96.07	91.85	83.05	79.20
90 cm (I_3)	20.72	17.32	18.72	15.21	26.66	23.25	25.64	22.35	213.54	192.04	192.00	171.79	96.88	93.21	88.07	84.20
SEm ±	0.47	0.34	0.37	0.32	0.93	0.86	0.95	0.92	13.69	13.88	15.36	15.75	5.88	5.87	6.30	5.91
CD *(P=0.05)*	1.36	0.99	1.07	0.92	2.67	2.47	2.74	2.64	NS	NS	NS	NS	NS	NS	NS	NS

* 'off' year; ** 'on' year
NFS = Non-floral shoots; FS = Floral shoots
†For details of the treatments, please see text

Table 2. Interaction effect of cultivar and pruning intensity on micronutrient content of leaves in three mango cultivars under high density planting

Treatments†	Cu (ppm)				Zn (ppm)				Fe (ppm)				Mn (ppm)			
	2005-06*		2006-07**		2005-06*		2006-07**		2005-06*		2006-07**		2005-06*		2006-07**	
	NFS	FS	NFS	FS	NFS	FS	NFS	FS	NFS	FS	NFS	FS	NFS	FS	NFS	FS
V_1I_0	18.56	15.16	13.66	12.83	23.23	19.53	21.16	18.86	202.90	176.13	159.13	144.60	88.30	84.66	76.76	72.80
V_1I_1	16.80	13.26	13.83	10.56	17.08	14.20	17.06	13.0	200.03	180.66	167.00	149.68	108.90	105.43	98.66	95.33
V_1I_2	19.03	15.90	16.36	13.40	17.56	15.43	17.46	14.76	226.20	202.23	173.90	161.96	86.10	73.66	67.60	64.23
V_1I_3	20.73	18.00	18.16	15.26	19.93	16.43	18.66	15.73	175.33	147.00	151.13	131.26	118.73	114.60	112.06	108.06
V_2I_0	21.60	17.90	18.10	15.33	22.63	20.23	22.56	19.60	218.10	193.60	185.63	167.16	97.00	92.60	82.13	90.20
V_2I_1	23.40	19.46	20.20	17.43	21.43	18.40	19.90	17.33	206.16	182.86	190.33	168.93	117.73	115.56	109.30	106.20
V_2I_2	21.06	16.96	18.40	15.10	21.33	18.56	20.73	17.90	267.60	251.13	244.96	228.13	134.46	181.80	122.66	110.56
V_2I_3	25.16	21.36	23.40	19.36	23.10	20.36	22.36	19.66	262.63	246.16	239.30	219.86	104.63	101.13	95.22	91.33
V_3I_0	11.36	7.63	9.06	5.53	25.16	21.26	23.33	20.66	205.90	183.16	180.16	158.76	91.13	89.23	84.43	80.73
V_3I_1	14.20	10.46	11.56	8.30	37.33	33.33	35.60	32.16	192.56	164.30	159.30	144.16	79.60	76.80	68.46	64.96
V_3I_2	15.46	12.20	13.86	10.50	23.96	20.10	23.66	18.56	217.90	195.03	173.73	158.56	67.66	65.10	58.90	54.80
V_3I_3	16.26	12.60	14.56	11.0	36.96	32.96	35.30	31.66	202.66	182.96	185.83	164.26	67.30	63.90	56.93	53.20
SEm ±	0.82	0.60	0.64	0.55	1.61	1.49	1.65	1.59	23.71	24.04	26.61	27.29	10.19	10.17	10.91	10.24
CD*(P=0.05)*	2.36	NS	1.85	1.59	4.63	4.29	4.74	4.57	NS	NS	NS	NS	29.20	29.21	31.36	29.43

* 'off' year; ** 'on' year
NFS = Non-floral shoots; FS = Floral shoots
† For details of treatments, please see text

leaves and perhaps due to Zn regulating enzymes synthesized after pruning and then rapid translocation due to high activity of cytokinins in leaves. The lowest level of Zn in moderately pruned trees (I2) may be due to existence of old leaves and fruiting leading to exhaustion of nutrients (Table 1). The rest of the treatments were at par. Lightly pruned 'Dashehari' recorded maximum Zn content (37.33, 33.33, 35.60 and 32.16 ppm) due to varietal characters, while lowest was seen in lightly pruned 'Amrapali' (17.08, 14.20, 17.06 and 13.0 ppm) may be due to a higher number of fruiting terminals (Table 2) (Thakur *et al*, 1981). During the 'on' year, Zn and Fe content decreased in mango leaves compared to the'off' year (Mishra and Dhillon ,1978; Thakur *et al*, 1979). Highest Fe content was noted in leaves of 'Mallika', while the minimum in 'Amrapali'. Effect of pruning intensity and its interaction with cultivar on Fe content was non-significant, which could be due to the several factors regulating nutrient composition in plant tissues. It is also clear from the data (Tables 1 and 2) that 'on' year had low levels of all the micronutrients studied in leaves than in the 'off' year. Similarly micronutrient content declined during the reproductive stage compared to the vegetative stage. The result of this study indicates that severe pruning in old mango trees may be preferred to improving micronutrient status, especially Cu and Zn, in flowering and non-flowering shoots.

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(MS Received 23 March, 2010, Revised 17, November 2010)

J. Hortl. Sci.
Vol. 5 (2): 124-127, 2010

Effect of organic nutrition practices on papaya (cv. Surya) fruit yield, quality and soil health

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ABSTRACT

A field experiment was conducted during 2005-07 at Indian Institute of Horticultural Research, Bangalore, on papaya cv. Surya with six organic treatments along with recommended dose of fertilizers and no manure/fertilizer application. Results indicated that crop growth and fruit yield were higher in inorganic fertilizer treatment (55 t ha^{-1}) compared to organic treatments (26.9 to 38 t ha^{-1}). There was no significant variation in average fruit weight and TSS, but shelf life of the fruit was significantly higher in organic treatments (6.2 to 7.9 days) as compared to inorganic fertilizer treatment (5.1days). Among the treatments, application of 7 kg urban compost plant^{-1} or 10 kg FYM plant^{-1} was found to be ideal for improving soil health in terms of microbial population, and biochemical reaction compared to other treatments.

Key words: Papaya, organic practices, fruit yield, quality, shelf life

INTRODUCTION

Organic farming is becoming increasingly popular, with a rapidly growing global demand for organic products. It offers considerable benefits over conventional farming systems particularly with respect to sustainable yield, better quality and health hazard free produce. Fruits, often eaten raw, are more vulnerable to contamination with chemicals due to the latter's residual toxicity as compared to cereals and pulses. Thus, organic production of fruits is gaining popularity over that of other crop groups.

Papaya is grown in an area of 98,000 ha with production of 36.29 lakh tons in India (National Horticultural Board, 2009). Since papaya bears fruits and flowers round the year, it is likely to respond well to organic production systems compared to other perennial fruit crops. In almost all the states, area under papaya is increasing, and limited information is available on organic production system in this crop. Hence, the present investigation is very important in crops like papaya.

MATERIAL AND METHODS

A field trial was conducted during 2005-2007 at the experimental farm of Indian Institute of Horticultural Research, Bangalore. The soil in the experimental plot was red loam with pH 6.12, 0.73% organic carbon, 158 kg available nitrogen/ha, 13 kg phosphorus/ha and 196 kg potash/ha. There were 8 treatments details of which are as follows;

T_1**:** Recommended dose of NPK fertilizers (250g N + 250 g P_2O_5 + 500 g K_2O plant^{-1}year^{-1}),

T_2**:** 10 kg FYM plant^{-1} year^{-1}

T_3**:** 7 kg urban compost plant^{-1} year^{-1}

T_4**:** 20 kg sun hemp + 150 g rock phosphate plant^{-1} year^{-1}

T_5**:** 2 kg neem cake + 0.5 kg wood ash plant^{-1} year^{-1}

T_6**:** 18 kg rural compost plant^{-1} year^{-1}

T_7**:** 2.5 kg vermi compost + 12.5 kg sun hemp plant^{-1} year^{-1}

T_8**:** No manure or fertilizer

Nutrient content of organic manures used in the experiment is as follows:

Organic manure used	Percentage		
	N	P	K
FYM	0.91	0.166	0.88
Rural compost	1.22	0.304	0.98
Urban compost	0.86	0.284	0.80
Vermicompost	1.41	0.299	0.55

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Standard procedures

Soil samples were collected at 2 years from experimentation for nutrient and microbial analysis. Microbial properties and soil enzymes were estimated as per standard procedures. Vegetative parameters such as plant height, plant girth and number of leaves were recorded at 6 month intervals. Fruit yield was recorded periodically. Fruit quality attributes such as TSS and keeping quality of fruits were also recorded as per standard procedures. Statistical analysis of data was done based on methods given by Panse and Sukhatme (1985). Plant spacing was 1.8 m×1.8 m in the trial.

RESULTS AND DISCUSSION

Vegetative characters : Vegetative parameters such as plant height, plant girth and number of leaves at 24 months from planting were affected by various nutrient treatments (Table 1). Maximum plant height, girth and number of leaves were recorded in the recommended dose of fertilizer treatment, whereas, no manure or fertilizer treatment recorded the least. Similar results were reported by Singh and Sharma (1996) Reddy *et al* (1986), Purohit (1977), Awada and Long (1978), Jauhari and Singh (1971), and Kumar *et al* (2006). Increased growth in recommended fertilizer dose treatment was mainly attributed to sufficient availability of all the nutrients during different growth stages of the plant, compared to other treatments

Fruit yield: Fruit yield in terms of number of fruits and their weight were found to be significantly different among various treatments (Table 2). Maximum fruit yield was recorded under recommended dose of fertilizer treatment, and the least with control treatment. Similar results were reported by Kumar *et al* (2006), Reddy *et al* (1986) and Singh & Sharma (1996). The increased fruit yield was attributed to better plant growth compared to that in other

Table 1. Vegetative characters, fruit yield and quality of 'Surya' papaya as influenced by various treatments (24 months after planting)

Treatment	Plant height (m)	Plant girth (cm)	No. of leaves plant^{-1}
T_1: Recommended dose of NPK fertilizers (250 g N + 250 g P_2O_5 + 500g K_2O plant^{-1}year^{-1})	2.49	52.0	25.8
T_2 : 10 kg FYM plant^{-1} year^{-1}	2.27	40.6	19.9
T_3 : 7 kg urban compost plant^{-1} year^{-1}	2.32	46.5	23.9
T_4 : 20 kg sun hemp + 150 g rock phosphate plant^{-1} year^{-1}	2.27	47.9	20.5
T_5 : 2 kg neem cake + 0.5 kg wood ash plant^{-1} year^{-1}	2.10	36.0	20.3
T_6 : 18 kg rural compost plant^{-1} year^{-1}	2.24	44.6	21.5
T_7 : 2.5 kg vermin compost + 12.5 kg sun hemp plant^{-1} year^{-1}	2.06	42.4	19.7
T_8 : No manure or fertilizer	1.68	33.3	16.1
SEm ±	0.01	0.30	0.38
CD *(P=0.05)*	0.04	0.90	1.12

Table 2. Fruit yield and quality of 'Surya' papaya as affected by various treatments

Treatment	Fruit yield				Fruit quality		
	No. of fruits plant^{-1}	Fruit yield (kg plant^{-1})	No. of fruits ha^{-1}(000)	Fruit yield (t ha^{-1})	Average fruit weight (g)	TSS (^{o}Brix)	Shelf life (days)
T_1: Recommended dose of NPK fertilizers (250 g N + 250 g P_2O_5 + 500 g K_2O plant^{-1}year^{-1})	37.9	17.8	116.8	55.0	472.6	11.1	5.1
T_2 : 10 kg FYM plant^{-1} year^{-1}	19.9	9.7	61.3	30.0	498.2	11.4	7.6
T_3 : 7 kg urban compost plant^{-1} year^{-1}	25.2	11.8	77.7	36.5	476.5	11.3	6.6
T_4 : 20 kg sun hemp + 150g rock phosphate plant^{-1} year^{-1}	30.0	12.6	92.5	38.7	427.7	12.2	7.1
T_5 : 2 kg neem cake + 0.5 kg wood ash plant^{-1} year^{-1}	20.0	9.9	61.6	30.6	495.3	11.3	6.2
T_6 : 18 kg rural compost plant^{-1} year^{-1}	27.0	11.5	83.2	35.5	430.0	11.6	6.6
T_7 : 2.5 kg vermi compost + 12.5 kg sun hemp plant^{-1} year^{-1}	18.7	8.7	57.7	26.9	473.8	11.4	7.0
T_8 : No manure or fertilizer	12.0	5.7	39.4	17.5	441.3	12.2	7.9
SEm ±	4.6	1.9	14.4	5.9	28.5	0.28	0.66
CD *(P=0.05)*	13.7	5.6	42.4	17.3	NS	NS	0.91

Table 3. Microbial population, soil respiration and mineralizable nitrogen in organic papaya (C.V.Surya field

Treatment	Bacteria (10^8cfug^{-1})	Fungi (10^4 cfug^1)	Actionomycetes (10^5 cfug^{-1})	Total Diazotrophs (10^4 cfu g^{-1})	Soilrespiration (mg C kg^{-1}soilhr^{-1})	Soil mineralizable nitrogen (mg N kg^{-1} of soil)
T_1: Recommended dose of NPK fertilizers (250 g N + 250 g P_2O5 + 500 g K_2O plant^{-1} year^{-1})	98.4	6.0	8.3	6.3	7.19	10.5
T_2 : 10 kg FYM plant^{-1} year^{-1}	141.8	18.3	16.0	21.0	8.57	46.25
T_3 : 7 kg urban compost plant^{-1} year^{-1}	139.6	16.4	17.8	19.4	7.26	47.25
T_4 : 20 kg sun hemp + 150 g rock phosphate plant^{-1} year^{-1}	116.4	8.4	14.0	16.5	10.10	42.00
T_5 : 2 kg neem cake + 0.5 kg wood ash plant^{-1} year^{-1}	119.6	11.0	14.8	15.0	9.70	45.50
T_6 : 18 kg rural compost plant^{-1} year^{-1}	136.4	18.0	16.4	23.2	8.70	56.00
T_7 : 2.5 kg vermi compost + 12.5 kg sun hemp plant^{-1} year^{-1}	127.3	11.6	13.6	19.1	9.85	43.75
T_8 : No manure or fertilizer	80.2	5.4	9.2	7.9	5.60	14.00
SEm ±	5.70	0.61	0.66	0.79	0.40	3.55
CD *(P=0.05)*	11.67	1.25	1.34	1.63	0.81	7.28

organic treatments or control. Although yield was higher in inorganic treatment (recommended dose of fertilizer) soil quality improvement was not noticed in terms of soil microflora and soil enzymes. Fruit yield reduction was 30-51% in organic treatments as compared to inorganic treatment at two years from experimentation. This may be due mainly to higher and quick availability of nutrients for growth and development under inorganic fertilizer treatment. In addition, pest and disease problem too may have resulted in reduced fruit yield in organic treatments (although progressive nutrient built up was seen in the soil due to addition of organic manures).

Fruit quality attributes: Fruit quality attributes like average fruit weight and TSS were found to be non significant but shelf life was found to be significantly different among various treatments (Table 2). Maximum shelf life was (7.9 days) seen in control, whereas, minimum shelf life (5.1 days) was noticed in recommended dose of fertilizer treatment. The finding is quite interesting but needs to be confirmed at different locations.

Soil health: Results on soil microbial population indicated that in general bacteria, fungi, actinomycetes and total diazotrophos were significantly higher in all the organic treatments compared to no manure and recommended dose of fertilizers (Table 3). The organic treatments recorded significantly higher soil respiration and mineralizable nitrogen content compared to recommended dose of fertilizer and control treatment. The finding clearly indicated an increase in microbial population in organic treatments, which may have improved soil respiration and mineralizable nitrogen content. Reduction in soil microorganisms in inorganic fertilizer treatment could be due to toxicity from metal contaminants found in inorganic fertilizers (Marschner *et al*, 2004), In the present study, treatments that resulted in higher organic carbon content in soil had higher microbial population. Similar results were reported by Chang *et al* (2007).

The results on soil enzyme activity (Table 4) indicated that among various treatments, dehydrogenase and glusosidase activity was significantly higher in 7 kg urban compost plant^{-1} treatment, whereas acid phosphatase and urease were significantly higher in 20 kg sunhemp plus 150 g rock phosphate plant^{-1} treatment compared to control and inorganic fertilizer applied treatments. These results reveal that treatments that received FYM or compost had greater microbial population, which may have increased soil enzyme activity compared to inorganic fertilizers alone or control. Higher levels of enzyme activity have been reported by many researchers in soils treated with vermicompost and organic manure compared to inorganic fertilizers (Krishna Kumar *et al*, 2005; Chang *et al*, 2007).

Results clearly revealed that organic nutrition practices in papaya production significantly improve soil health in terms of soil microbial and biochemical properties

Table 4. Soil enzyme activity in organic papaya field (24 months after planting)

Treatment	Soil enzyme activity			Urease[4]
	Dehydro-genase[1]	βGluco-sidase[2]	Acid.phos-phatase[3]	
T_1: Recommended dose of NPK fertilizers (250g N + 250 g P_2O_5 + 500 g K_2O $plant^{-1}year^{-1}$)	27.4	69.2	86.1	29.4
T_2 : 10 kg FYM $plant^{-1}$ year	83.5	169.3	106.8	66.5
T_3 : 7 kg urban compost $plant^{-1}$ $year^{-1}$	102.4	226.2	109.2	60.2
T_4 : 20 kg sun hemp + 150 g rock phosphate $plant^{-1}$ $year^{-1}$	78.9	147.7	121.0	79.8
T_5 : 2 kg neem cake + 0.5 kg wood ash $plant^{-1}$ $year^{-1}$	69.6	141.1	112.1	63.0
T_6 : 18 kg rural compost $plant^{-1}$ $year^{-1}$	83.7	225.4	107.5	46.9
T_7 : 2.5 kg vermicompost + 12.5 kg sun hemp $plant^{-1}$ $year^{-1}$	74.7	177.5	112.3	39.9
T_8 : No manure or fertilizer	39.8	68.7	94.4	28.6
SEm	3.39	7.32	4.96	2.50
CD *(P=0.05)*	6.95	14.98	10.15	5.12

[1]. µg TPF released g^{-1} of soil h^{-1}, [2]. µg g^{-1} soil h^{-1}, [3]. µg *p*-nitrophenol released g^{-1} soil h^{-1} [4]. µg NH_4 formed g^{-1} soil h^{-1}

compared to application of inorganic fertilizers alone. Among the treatments, application of 7 kg urban compost $plant^{-1}$ or 10 kg FYM $plant^{-1}$ was found to be ideal for improving soil qualities, but fruit yield was significantly higher under recommended dose of fertilizers compared to that under organic treatments.

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(MS Received 15 February 2010, Revised 13 August 2010)

J. Hortl. Sci.
Vol. 5 (2): 128-133, 2010

Role of Paclobutrazol and Ethephon in reproductive growth of 'Allahabad Safeda' guava (*Psidium guajava* L.) plants at different spacing

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ABSTRACT

A study on 4-year 'Allahabad Safeda' guava plants was made to assess the influence of Paclobutrazol (PP 333), [(2RS, 3RS)-1-(4-chlorophenyl)-4,4-dimethyl-2-(1,2,4 triazol-1-yl) pentan-3-ol], a gibberellin-inhibitor, and Ethephon [(2-chloroethyl) phosphonic acid], a vegetative growth inhibitor and a ripening promoter, on reproductive growth of plants. Treatments in the form of foliar application at 500 and 1000 ppm were applied consecutively during March 2007 and 2008 on plants at 6m x 2m, 6m x 3m, 6m x 4m and 6m x 5m spacing. Maximum flowering and fruit set was recorded in paclobutrazol treated plants in both rainy and winter crops. Ethephon reduced flower bud density (FBD) and fruit set during both the cropping seasons. However, Ethephon treated plants exhibited slightly higher fruit retention. Ethephon advances fruit maturity by upto a week during rainy season and two weeks during winter season. Paclobutrazol treated plants exhibited significantly higher fruit number, fruit yield, yield efficiency, fruiting density compared to Ethephon treated and control plants. Reproductive growth of plants at wider spacing of 6m x 5m and 6m x 4m significantly improved compared to closer spacings of 6m x 2m and 6m x 3m during both cropping seasons. Plants at wider spacing responded better to Paclobutrazol applications with respect to flowering and fruiting.

Key words: Guava, Paclobutrazol, Ethephon, flowering and fruiting

INTRODUCTION

Guava (*Psidium guajava* L.) is an important fruit crop grown throughout the tropics and sub-tropics of the world. It is a very hardy plant, a prolific bearer and highly remunerative fruit crop. It can also be grown satisfactorily under adverse soil and climatic conditions. Productivity of guava can be further enhanced by increasing planting density and by better canopy management. Therefore, high density plantation with a managed canopy, that has balanced vegetative and reproductive growth, is the need of the hour to achieve high productivity per unit area.

In the absence of dwarfing rootstocks in guava, techniques that restrict vegetative growth and promote reproductive growth are important in orchard management. Therefore, apart from pruning and training of roots and shoots, certain growth retardants (alone or in combination) may be exploited. Although a flowering and fruit set under normal planting system is not a problem, there is a wide scope for enhancing the productivity potential of guava plants, particularly under reduced plant spacing. As guava tree responds well to canopy modification with respect to vegetative and reproductive growth (Singh and Chanana, 2005), modification of canopy through pruning and use of certain growth regulators in high density orchards may be required to enhance production efficiency. Some growth regulators like Paclobutrazol and Ethephon may also be very useful in high density planting, as Paclobutrazol helps make the plants dwarf by a retarding effect on vegetative growth of the tree while increasing the number of flower buds. Ethephon acts as a ripening hormone and enhances the ripening process besides having a growth retarding effect. Paclobutrazol @ 500 ppm improved fruit set in winter season crop of guava (Singh and Bal, 2006). Similarly, Jain and Dashora (2007) also recorded highest yield under 500 ppm PBZ treatment.

MATERIAL AND METHODS

The present investigation, relating to reproductive growth of 'Allahabad Safeda' guava (*Psidium guajava* L.) plants at different spacings with Paclobutrazol and Ethephon treatments, was carried out in the New Orchard, Department of Horticulture, Punjab Agricultural University, Ludhiana, during the years 2007-2009. Plants of guava cv.

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'Allahabad Safeda' were planted in March, 2003 at different spacings viz., 6m x 2m, 6m x 3m, 6m x 4m and 6m x 5m, with three replications. Experimental plants were sprayed with Paclobutrazol and Ethephon in both the years @ 500 and 1000ppm, while control plants were sprayed with water, during the month of March. Observations were recorded on fruiting characters, i.e., flower bud density, fruit set, fruit retention, fruit maturity, yield per tree and yield efficiency for both the rainy and winter season crops in May-July and September-December, during both years of study.

For calculation of flower bud density, three tertiary shoots (one meter) of medium vigour each in the upper, middle and lower parts of the tree canopy of every plant were randomly selected and tagged. Number of flowers on each shoot was counted and the average worked out. Fruit set was recorded by counting the number of fruits that had set on tagged shoots after the petal-fall stage and per cent fruit set was calculated. Similarly, fruit retention was calculated by counting the number of fruits left on each tagged shoot 8-10 days before harvest, which was expressed as per cent fruit retention. Fruit maturity was recorded by noting the number of days taken from fruit set to maturity, by counting mature fruits in all the parts of the tree canopy during both the cropping seasons. Maturity was judged on the basis of parameters like colour break, total soluble solids, acidity, firmness and TSS/acid ratio. Fruit yield per tree under each treatment was recorded at harvest and yield efficiency was determined by dividing average fruit load on a tree by canopy volume and was expressed in percentage. Data were analyzed in this Randomized Block Design with split plot.

RESULTS AND DISCUSSION

Flower Bud Density (FBD) : The highest flower bud density for rainy season crop was noted in plants sprayed with PBZ 1000 ppm (35.87 flowers/shoot), followed by 30.74 in plants treated with PBZ 500 ppm. Ethephon 1000 ppm treated plants exhibited the least FBD of 26.9 flowers/meter shoot (Table 1). However, in the winter season guava, FBD was found to be highest (8.53) in Ethephon 500 ppm treated plants and the least (6.32) in untreated plants. Reduction in FBD under higher doses of Ethephon may be due to Ethephon induced leaf shed, causing reduced transfer of the stimulus necessary for induction of flower buds. However, higher FBD in Paclobutrazol treated plants may be due to enhancement of reproductive growth at expense of vegetative growth, reduced by the gibberellin inhibiting effect of Paclobutrazol. Manivannan and Bharthikannan (2005) also found positive a response with respect to number of flowers produced, days to first flower bud appearance, size and yield of fruits, at 1500 ppm Paclobutrazol. In earlier investigations on mango (Winston, 1992, Tongumpai *et al,* 1997, Singh, 2000) and lychee (Manzel and Simpson, 1990), improvement was reported in flowering, with PBZ application. Similarly, Mohammed *et al* (1984) and Castelen and Becerril (1994) reported that Ethephon was appreciably effective in increasing flower bud production in guava.

The mean maximum number of flower buds/1m shoot during rainy season was observed to be 35.93 at 6m x 5m, and 32.31 flowers per meter shoot at 6m x 4m spacing. Plants at closer spacings of 6m x 2m and 6m x 3m produced minimum average number of flower buds/shoot, i.e., 22.44 and 29.4, respectively. During winter, the highest FBD of 14.12 was noted in plants at 6m x 5m spacing, and the least (4.38) in 6m x 3m spaced plants. FBD in the winter season was quite low due to overbearing during the rainy season, i.e., metabolic reserves required for induction of flowering may have got reduced due to heavy flowering in April-May, thereby affecting FBD in August- September in the winter season crop. Reduction in FBD with decrease in plant spacing may be attributed to reduced radiant energy received

Table 1. Effect of Paclobutrazol and Ethephon on flower bud density of 'Allahabad Safeda' guava planted at different spacings

Treatment (ppm)	Spacing (m)									
	Rainy season					Winter season				
	6x2	6x3	6x4	6x5	Mean	6x2	6x3	6x4	6x5	Mean
Paclobutrazol 500	24.18	32.45	38.28	28.06	30.74	4.10	4.75	6.44	17.48	8.19
Paclobutrazol 1000	24.20	34.71	39.92	44.66	35.87	4.58	4.70	8.15	15.39	8.21
Ethephon 500	22.70	27.82	23.87	38.00	28.10	4.88	4.63	8.92	15.68	8.53
Ethephon 1000	21.15	28.85	29.02	28.59	26.90	5.00	3.83	6.69	11.81	6.83
Control	19.96	23.19	30.47	40.36	28.50	5.37	3.97	5.69	10.26	6.32
Mean	22.44	29.40	32.31	35.93	30.02	4.79	4.38	7.18	14.12	7.62
CD *(P=0.05)*	Treatment (A): 6.16 Spacing (B): 5.51 Interaction: A x B: 9.64					Treatment (A): 1.20 Spacing (B) : 1.07 Interaction: A x B: 1.5				

NS = Non-Significant

Table 2. Effect of Paclobutrazol and Ethephon on fruit set (%) of 'Allahabad Safeda' guava planted at different spacings

Treatment (ppm)	Spacing (m)									
	Rainy season					Winter season				
	6x2	6x3	6x4	6x5	Mean	6x2	6x3	6x4	6x5	Mean
Paclobutrazol 500	45.00	53.48	59.39	48.94	51.70	64.86	66.51	75.19	80.08	71.66
Paclobutrazol 1000	45.00	55.75	61.03	65.78	56.89	65.49	68.26	71.38	78.21	70.84
Ethephon 500	43.78	48.70	44.68	59.11	49.07	64.06	67.00	71.69	76.92	69.92
Ethephon 1000	41.93	49.79	49.96	49.51	47.80	61.20	65.71	69.57	74.41	67.72
Control	40.70	43.99	51.44	61.48	49.40	62.00	68.49	73.07	74.69	69.56
Mean	43.28	50.34	53.3	56.96	50.97	63.52	67.19	72.18	76.86	69.94
CD *(P=0.05)*	Treatment (A): 2.68 Spacing (B): 3.00 Interaction: A x B: NS					Treatment (A): 1.85 Spacing (B) : 2.07 Interaction: A x B: NS				

NS = Non-Significant

and, also, reduced substrate level with a reduction in shoot growth. The present results are in conformation with those of Lal *et al* (1996), who also found guava trees at closer spacing (2m x2m) producing lower number of flower buds compared to higher spacing (8m x 8m). Similarly, Singh (2003) also recorded an increase in flower bud density with increased plant spacing in guava.

Fruit set: Fruit set in both the seasons improved significantly with PBZ application (Table 2). Average fruit set during the rainy season was found to be maximum when plants were sprayed with PBZ 1000ppm (56.89%) followed by 51.07% in PBZ 500ppm treated plants. The least fruit set (47.8%) was recorded in Ethephon 1000ppm treated plants. In winter season too, PBZ enhanced fruit set. Maximum mean fruit set was found with PBZ 500ppm (71.66%), followed by 70.84% in PBZ 1000ppm treated plants. Minimum average fruit set (67.72%) was noted in Ethephon 1000ppm sprayed plants, followed by 69.56% in untreated plants. Lim and Nualsri (1992) also observed improvement in fruit set of Neck orange with PBZ treatment. Similarly, Singh (2000) reported increase in fruit set in mango with PBZ treatment. Fruit set in plants sprayed with Ethephon, particularly at higher dose, was recorded to be low; this may be due to Ethephon induced leaf shed thus causing an uncongenial microclimate in the tree canopy, which may be responsible for reduced fruit set. On the other hand, PBZ was found to increase fruit set, by channelizing energy available for the vegetative growth to reproductive growth.

During rainy season, the maximum mean fruit set of 56.96% and 53.3% was recorded in the wider spacings of 6m x 5m and 6m x 4m, respectively. The minimum average per cent fruit set of 43.28% was recorded in the close spacing of 6m x 2m. However, in the winter season crop, fruit set was found to be higher than in the rainy season crop due the low number of flowers in winter season. Maximum fruit set (76.86%) was observed in plants at 6m x 5m spacing and the least average fruit set was observed in close spacing of 6m x 2m (63.52%). Low fruit set at close spacing may be due to less spread of trees, and to lower light and air penetration into the canopy. Data in winter season crop are in line with observation of Lal *et al* (1996) who reported lower fruit set in close spacing (2m x 2m) compared to a wider spacing (8m x 8m). However, results of this study are in contradiction to those of Singh (2006), who recorded maximum fruit set at close spacing.

Fruit retention: Growth regulators had significant effect on fruit retention in both rainy and winter season crops of guava. In rainy season guava fruit retention recorded in plants treated with Ethephon 500 and 1000 ppm was 47.66 and 44.36%, respectively and lowest fruit retention (39.62%) was observed in PBZ 500 ppm treated plants (Table 3). In the winter season guava, similar trend of fruit retention was observed. Maximum mean fruit retention was recorded in plants treated with Ethephon 500 ppm (62.56%) and least retention was noted in PBZ 500 ppm (53.73%) and untreated (53.74%) plants. Higher retention in Ethephon treated plants may be due to low FBD and fruit set, resulting in low fruit load on the trees, thereby, enabling plants to hold higher number of fruits owing to higher availability of translocates.

Average per cent fruit retention was not significantly affected by various different spacings during both seasons. However, mean fruit retention was maximum (43.5%) at 6m x 4m, and 59.11% at 6m x 5m spacing. However, the least average per cent fruit retention (41.64 and 55.17%) was observed in 6m x 2m and 6m x 3m spacings during rainy and winter crop seasons, respectively. Retention of winter season fruits in both cultivars was higher than in the rainy season fruits. This may be due to very low fruit count during winter season, there by resulting in higher allocation of nutrients and translocates to fruits on the plants.

Fruit maturity : The treatments had significant effect on

fruit maturity in both rainy and winter season crops. Rainy season fruits took relatively less number of days to maturity (Table 4) when sprayed with Ethephon 500 ppm (65.9 days), followed by 66.5 days in Ethephon 1000 ppm treatment. Similarly, in the winter season, fruit maturity was attained earlier, i.e., 105.9 and 106.3 days, with Ethephon 1000 and 500 ppm sprays, respectively. Therefore, Ethephon treatment induced advance maturity by approximately 5-6 days (i.e., earlier) in the rainy season and 10-12 days in the winter season compared to Paclobutrazol treated and control plants. This might be due to Ethephon inducing early ripening, as it is the key plant hormone responsible for fruit ripening. Further, partial leaf shedding due to Ethephon, particularly at the higher dose, may be another factor which enabled light- penetration and a rise in temperature, resulting in early fruit ripening.

Maturity of rainy and winter season crops was not significantly affected by different spacings. However, fruit maturity in widely spaced plants was slightly earlier than in the closer spacing, probably due to higher solar radiation penetration and canopy temperature.

Fruit Yield : Per plant fruit yield was maximum (34.79 kg/plant) in plants sprayed with PBZ 1000 ppm, followed by 31.55 kg/plant in PBZ 500 ppm treated plants during the rainy season. PBZ 1000 ppm sprayed plants also gave the highest yield of 18.71 kg/plant during the winter season. Lowest per plant yield during rainy season was 23.54 kg and 13.42 kg/plant in winter season (Table 5) in untreated plants. Benjawan *et al* (2006) also reported increase in 'Kaew' mango fruit yield with PBZ treatment. Overall yield, primary crop yield and number of primary clusters were seen to be significantly reduced in 'Chenin Blanc' grapevine treated with Ethephon upto two weeks after bloom (Szyjewicz and Kliewer, 1983). Yadav *et al* (2001) also recorded minimum yield in 'Sardar' guava plants treated with Ethrel compared to control plants. However, Suleman *et al* (2006) found that Ethephon application during May reduced the yield of rainy season guava crop significantly over control, and subsequently increased the yield of winter season crop.

Highest yield was obtained from plants at the widest (6m x 5m) spacing during rainy and winter crop seasons, i.e., 35.3 and 28.65 kg/plant, and, lowest yield of 18.56 and

Table 3. Effect of Paclobutrazol and Ethephon on fruit retention (%) of 'Allahabad Safeda' guava planted at different spacings

Treatment (ppm)	Spacing (m)									
	Rainy season					Winter season				
	6x2	6x3	6x4	6x5	Mean	6x2	6x3	6x4	6x5	Mean
Paclobutrazol 500	39.12	38.63	40.37	40.37	39.62	54.23	51.96	52.92	55.80	53.73
Paclobutrazol 1000	38.11	39.52	41.40	41.00	40.01	51.96	51.80	54.70	57.62	54.02
Ethephon 500	46.38	47.40	49.23	47.63	47.66	59.86	62.05	63.01	65.30	62.56
Ethephon 1000	43.21	43.59	45.97	44.65	44.36	58.00	59.25	58.34	63.13	59.68
Control	41.37	41.25	40.54	41.73	41.22	51.81	54.11	55.33	53.71	53.74
Mean	41.64	42.08	43.50	43.08	42.57	55.17	55.83	56.86	59.11	56.74
CD *(P=0.05)*	Treatment (A): 3.10 Spacing (B): NS Interaction: A x B: NS					Treatment (A): 1.34 Spacing (B) : 1.50 Interaction: A x B: 1.90				

NS = Non-Significant

Table 4. Effect of Paclobutrazol and Ethephon on fruit maturity (days) of 'Allahabad Safeda' guava planted at different spacings

Treatment (ppm)	Spacing (m)									
	Rainy season					Winter season				
	6x2	6x3	6x4	6x5	Mean	6x2	6x3	6x4	6x5	Mean
Paclobutrazol 500	76.5	69.5	72.0	67.8	71.5	115.5	116.8	111.2	119.5	115.8
Paclobutrazol 1000	72.6	72.3	70.5	68.5	71.0	114.8	114.2	110.5	113.2	113.2
Ethephon 500	69.2	65.5	66.5	62.5	65.9	108.6	106.5	105.0	105.2	106.3
Ethephon 1000	68.5	66.8	67.5	63.2	66.5	107.0	106.7	106.5	103.2	105.9
Control	74.2	70.2	71.3	71.2	71.7	114.0	113.8	109.8	110.7	112.1
Mean	72.2	68.9	69.6	66.6	69.3	112.0	111.6	108.6	110.4	110.6
CD *(P=0.05)*	Treatment (A): 0.94 Spacing (B): NS Interaction: A x B: NS					Treatment (A): 0.96 Spacing (B) : NS Interaction: A x B: NS				

NS = Non-Significant

Table 5. Effect of Paclobutrazol and Ethephon on fruit yield (kg/tree) of 'Allahabad Safeda' guava planted at different spacings

Treatment (ppm)	Spacing (m)									
	Rainy season					Winter season				
	6x2	6x3	6x4	6x5	Mean	6x2	6x3	6x4	6x5	Mean
Paclobutrazol 500	20.47	32.06	42.88	30.77	31.55	6.83	8.95	11.93	38.00	16.43
Paclobutrazol 1000	19.30	29.96	48.06	41.85	34.79	8.96	11.71	19.50	34.66	18.71
Ethephon 500	19.73	23.15	23.78	40.67	26.83	12.93	10.40	16.00	33.35	18.17
Ethephon 1000	18.21	24.46	27.80	28.06	24.63	11.68	7.53	16.90	21.00	14.28
Control	15.07	18.05	25.87	35.15	23.54	6.83	13.36	17.25	16.22	13.42
Mean	18.56	25.54	33.68	35.30	28.27	9.45	10.39	16.32	28.65	16.20
CD *(P=0.05)*	Treatment (A): 4.50 Spacing (B): 4.64 Interaction: A x B: NS					Treatment (A): 2.51 Spacing (B) : 2.24 Interaction: A x B: 3.17				

NS = Non-Significant

Table 6. Effect of Paclobutrazol and Ethephon on yield efficiency (%) of 'Allahabad Safeda' guava planted at different spacings

Treatment (ppm)	Spacing (m)									
	Rainy season					Winter season				
	6x2	6x3	6x4	6x5	Mean	6x2	6x3	6x4	6x5	Mean
Paclobutrazol 500	55.7	84.1	113.6	69.8	80.5	18.6	23.5	31.6	86.2	41.9
Paclobutrazol 1000	50.4	71.6	95.8	94.2	79.6	23.4	28.0	38.9	78.0	42.8
Ethephon 500	47.2	51.1	64.9	100.5	65.4	30.9	23.0	43.6	82.4	44.3
Ethephon 1000	44.6	60.0	72.7	57.5	58.4	28.6	18.5	44.2	43.0	33.9
Control	32.2	32.9	46.8	60.2	43.7	14.6	24.3	31.2	27.8	24.9
Mean	45.4	57.8	77.2	74.8	64.3	23.1	23.5	37.4	60.7	36.8
CD *(P=0.05)*	Treatment (A): 26.52 Spacing (B): 23.88 Interaction: A x B: 23.20					Treatment (A): 7.10 Spacing (B) : 13.92 Interaction: A x B: 5.85				

NS = Non-Significant

9.45 kg/plant was recorded in plants at the closest spacing of 6m x 2m, respectively. Similar results were reported by Chundawat *et al* (1992), Kalra *et al* (1994), Lal *et al* (2000) and Bal and Dhaliwal (2003) in guava. However, Singh *et al* (2007) also recorded highest yield in guava at 3m x 1.5m spacing from rainy and winter season crops, respectively, and the minimum at 6m x 6m spacing.

Yield efficiency : Yield efficiency of plants under different treatments (Table 6) was influenced significantly during both seasons. During the rainy season, maximum average yield efficiency (80.5%) was noted in PBZ 500 ppm treated plants, followed by 79.6% in PBZ 1000 ppm treatment. Treatments with Ethephon significantly reduced yield efficiency during the rainy season. Lowest efficiency of yield (43.7%) was observed in untreated plants. In the winter season, Ethephon 500 and PBZ 1000 ppm sprays gave the highest yield efficiency of 43.3 and 42.8%, respectively, while, untreated plants exhibited the least yield efficiency of 24.9%, followed by 33.9% in Ethephon 1000 ppm treated plants.

Yield efficiency significantly increased with increase in plant spacing. Maximum average efficiency of yield (77.2%) was recorded in 6m x 4m spacing, followed by 74.8% in 6m x 5m spacing. Least yield efficiency (45.4%) was obtained in plants at 6m x 2m spacing. Similarly, in winter season, maximum mean yield efficiency (60.7%) was noted in 6m x 5m spacing, and the lowest average yield efficiency of 23.1% was recorded in the close spacing of 6m x 2m.

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(MS Received 4 February 2010, Revised 28 June 2010)

J. Hortl. Sci.
Vol. 5 (2): 134-137, 2010

Effect of spacing and crop duration on growth, flowering and bulb production in tuberose (*Polianthes tuberosa* L.) cv. Double

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ABSTRACT

Field experiments were conducted at Junagadh during 2002-05 to study the response of spacing (45 x 45, 45 x 30, 45 x 15, 30 x 30 and 30 x 15 cm) and crop duration (first year crop, first ratoon and second ratoon) on growth, flowering, cut flower yield and bulb production in tuberose cv. Double. The widest spacing (45 cm x 45 cm) registered the highest values for plant height (46.18 cm), number of leaves per clump (67.25), spike length (89.64 cm), spike diameter (0.95 cm), diameter of open flower (4.6 cm), rachis length (34.8 cm), number of spikes per clump (4.1), number of florets per spike (48.2), number of bulbs per clump (18.40) and number of bulblets per clump (31.60). It also induced early spike emergence and flowering. A planting distance of 30 x 30 cm realized the highest cut flower yield (2.72 lakh ha^{-1}) and that of 30 cm x 15 cm recorded the highest bulb production (22 lakh ha^{-1}). Ratoon crops showed higher plant height, number of leaves, bulbs, bulblets and spikes per clump and cut flower yield as well as bulb production over the first year crop. Early spike emergence and flowering was also noted in ratoon crops compared to the first year crop. However, spike and flower quality was inferior to that of first year crop with regard to spike length and diameter, number of florets per spike, diameter of open flower and rachis length.

Key words: Tuberose, spacing, crop duration, growth and flowering

INTRODUCTION

Gujarat is endowed with a diverse agroclimate conducive for growing different flower crops throughout the year. Gujarat has made rapid strides in floriculture, evident from 61% increase in area from 7,118 ha (2005-06) to 11,473 ha (2008-09) and over 100% increase in flower production from 42,182 tonnes in 2005-06 to 85,216 tonnes in 2008-09 (Anon., 2009). Major flowers grown in the state are roses, spider lily, marigold, jasmine and tuberose. Among these, tuberose is valued by the aesthetic world for its beauty, elegance and pleasant fragrance. Long flower spikes are excellent cut flowers for table decoration. Individual florets are much in demand for preparation of artistic garlands, floral ornaments, bouquets and for button holes. The 'concrete' and 'absolute' prepared from tuberose florets are valuable perfumery products. In fact, India is the second largest producer and exporter of tuberose concrete to the world market. Though tuberose is cultivated on a commercial scale in Gujarat, there are no standard recommended packages of practices available for the Saurashtra region of Gujarat. It is well established that flower and bulb production in tuberose is strongly influenced by planting density and crop duration, besides other factors. Keeping this in mind, the present experiment was undertaken to evaluate the response of spacing and crop duration on growth, flowering, cut flower yield and bulb production in tuberose.

MATERIAL AND METHODS

The present investigation was undertaken at the Jambuvadi Fruit Research Station, Department of Horticulture, Junagadh Agricultural University, Junagadh. The effect of five different spacings (45 x 45, 45 x 30, 45 x 15, 30 x 30 and 30 cm x 15 cm) on growth, cut flower yield and bulb production in tuberose was evaluated in a Randomized Block Design with four replications. Medium sized bulbs (1.8 to 2.4 cm in diameter) of tuberose cultivar 'Double' were planted in the first week of June 2002 and retained for the next three years [first year crop, second year crop (first ratoon) and third year crop (second ratoon)]. The experimental field was brought to a fine tilth by ploughing and harrowing. The gross plot size was 2.70 m x 2.70 m. However, net plot size varied with the spacing employed (Table1).

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Well decomposed farm yard manure @15 t ha^{-1} was uniformly applied and thoroughly mixed with the soil. The crop was fertilized with 150 kg N, 25 kg P_2O_5 and 25 kg K_2O ha^{-1}. Half dose of nitrogen as urea, and full doses of phosphorus as single super phosphate and potassium as muriate of potash, were applied at the time of planting. The remaining half of nitrogen was applied in two equal splits at 45 and 90 days after planting. The same dose of fertilizers was repeated for ratoon crops, as well. Five plants were selected randomly from the net plot in each treatment and replication and tagged for recording observations. Plants were grown under uniform cultural practices. Observations were recorded on fourteen plant characters, viz., plant height, number of leaves per clump at first flower emergence, number of bulbs, bulblets and spikes per clump at final harvest, days to first spike emergence and first flower opening from the date of planting, spike length and diameter (cm), number of florets per spike, diameter of open flower (cm) and rachis length (cm). Cut flower and bulb yield (lakh ha^{-1}) were calculated on per hectare basis to reflect the yield per unit area. Plant height (cm) was measured from the ground level to the tip of the longest leaf at harvest. Data obtained were tested for critical difference (CD) among various treatments (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

Vegetative growth parameters

Data presented in Table 2 reveal that different spacings and crop duration had significant influence on growth of tuberose in the first year crop and ratoon crops.

Table 1. Planting density and net plot size under different spacings

Sl. No.	Spacing (cm)	Planting density (number of plants/ha)	Net plot size (m)
S_1	45 x 45	49383	1.80 x 1.80
S_2	45 x 30	74074	2.10 x 1.80
S_3	45 x 15	148148	2.40 x 1.80
S_4	30 x 30	111111	2.10 x 1.80
S_5	30 x 15	222222	2.40 x 2.10

Table 2. Influence of spacing and crop duration on vegetative growth parameters in tuberose cv. 'Double'

Spacing(cm)	Plant height (cm)			No. of leaves per clump		
	First year crop	Ratoon crop		First Second ratoon	Ratoon crop	
		Firs ratoon	year crop		First ratoon	Second ratoon
S_1 (45 x 45)	46.18	52.63	58.82	67.25	82.70	98.00
S_2 (45 x 30)	44.15	49.30	56.82	60.75	74.95	91.70
S_3 (45 x 15)	35.76	42.86	43.74	46.95	60.00	76.10
S_4 (30 x 30)	41.72	48.99	55.79	57.80	69.60	87.80
S_5 (30 x 15)	30.09	33.66	33.45	36.60	46.15	57.50
CD *(P = 0.05)*	5.36	5.40	4.53	8.36	12.75	11.91

Growth parameters, viz., plant height and number of leaves per clump increased with increase in spacing. The widest spacing of 45 cm x 45 cm was at par with 45 cm x 30 cm and recorded the highest values for plant height (46.18 cm) and number of leaves per clump (67.25). This may be due to greater available space to every plant for availing sufficient nutrients, soil moisture and solar radiation, factors which may have restricted the plants in closer spacings. This is in accordance with findings of Malik *et al* (2009). These attributes progressively increased in ratoon crops compared to the first year crop. This could be ascribed to formation of bulblets in the first year crop that could develop into bulbs in the ratoon crop which, in turn promoted plant growth during the ratoons.

Flowering traits

Flowering traits also differed significantly under various plant spacings and crop durations in the first year crop as well as in ratoon crops (Table 3). However, days to spike emergence, days to first flower opening and spike diameter were not affected by spacing in the second ratoon crop. It was also observed that days to spike emergence and days to first flower opening decreased with increase in spacing. On the other hand, spike length, spike diameter, diameter of open flower and rachis length increased with increase in spacing. Wider spacings, viz., 45 cm x 45 cm and 45 cm x 30 cm were on par and induced early spike emergence with higher spike length and diameter as compared to closer spacings (45 cm x 15 cm and 30 x 15 cm). Earliest spike emergence (205.75 days) and maximum spike length (89.64 cm) and diameter (0.95 cm) were observed under a spacing of 45 cm x 45 cm. Better growth and subsequent differentiation may have contributed to improved spike characters under wider spacing. Similar results were obtained by Tyagi *et al* (2008). Early spike emergence was recorded in ratoon crops compared to the first year crop. This might be due the well established root system in ratoon crops. Nevertheless, spikes had smaller diameter in both ratoon crops compared to the first year crop.

Wider spacings also resulted in early opening of flowers, with higher diameter of open flower and rachis length. The spacing of 45 cm x 45 cm registered minimum days to flower opening (229.2 days) and maximum diameter of open flowers (4.6 cm) and rachis length (34.8 cm). Better leaf growth under wider spacing may have accelerated photosynthesis during the vegetative period and its translocation of photosynthesis to various metabolic sinks during the reproductive period could be responsible for

improvement in floral attributes. These results are in line with those reported by Kumar and Singh (1998). Early flowering was observed in ratoon crops compared to that in the first year crop. However, flower quality (with regard to diameter of open flowers and rachis length) was inferior to the first year crop.

Cut flower yield and Bulb production

An appraisal of data furnished in Table 4 indicates that cut flower yield and associated traits were significantly affected by planting distance and crop duration in the first year crop and ratoon crops. Number of spikes per clump and number of florets per spike increased with increase in spacing. Maximum number of spikes per clump (4.1) and number of florets per spike (48.2) were obtained in bulbs planted at a spacing of 45 cm x 45 cm. Ratoon crops recorded higher number of spikes per clump than the first year crop, whereas, the first year crop registered higher number of florets compared to the ratoon crops. Cut flower yield increased with decrease in spacing and highest cut flower yield (2.72 lakh/ha) was recorded under a spacing of 30 x 30 cm, which was at par with 45 cm x 15 cm spacing in both the first year crop and in ratoon crops. These results are in agreement with earlier findings of Kadam *et al* (2005). Higher cut flower yield was observed in ratoon crops as compared to the first year crop.

Bulb production also varied significantly with different spacings and crop durations in the first year crop and ratoon crops (Table 5). Number of bulbs and bulblets per clump increased with increase in spacing. The widest spacing of 45 x 45 cm was at par with 45 cm x 30 cm and the highest number of bulbs (18.40) and bulblets (31.60) per clump. Number of bulbs and bulblets per clump under each spacing were correspondingly higher in ratoon crops compared to the first year crop. Bulb yield increased with decrease in spacing. Highest bulb yield (22.00 lakh/ha) was obtained with a spacing of 30 cm x 15 cm. This is in close conformity with observations of Singh (2003). Ratoon crops showed a progressive increase in bulb yield for each successive spacing compared to the first year crop. This may be ascribed to the fact that well established clumps had higher number of daughter bulbs which in turn produced more number of spikes thereby resulting in higher cut flower yield and bulb yield compared to the first year crop.

Table 3. Influence of spacing and crop duration on flowering traits in tuberose cv. 'Double'

Trait	Spacing (cm)					
	S_1 (45 x 45)	S_2 (45 x 30)	S_3 (45 x 15)	S_4 (30 x 30)	S_5 (30 x 15)	CD (P = 0.05)
1. Days to spike emergence						
First year crop	205.7	210.2	225.0	215.2	234.7	19.71
First ratoon crop	189.0	194.0	208.5	198.7	216.2	18.67
Second ratoon crop	178.7	183.7	193.7	189.7	200.7	NS
2. Days to first flower opening						
First year crop	229.2	235.0	257.0	240.2	268.7	25.01
First ratoon crop	216.2	222.0	239.2	228.2	249.2	14.79
Second ratoon crop	203.0	210.5	221.7	216.2	228.5	NS
3. Spike length (cm)						
First year crop	89.6	87.0	74.9	84.2	66.4	7.08
First ratoon crop	82.4	77.2	62.3	70.8	53.0	7.15
Second ratoon crop	68.4	64.5	50.9	62.1	39.7	6.72
4. Spike diameter (cm)						
First year crop	0.95	0.92	0.87	0.90	0.82	0.06
First ratoon crop	0.83	0.78	0.72	0.75	0.65	0.04
Second ratoon crop	0.67	0.66	0.64	0.65	0.61	NS
5. Diameter of open flower (cm)						
First year crop	4.6	4.4	4.0	4.3	3.9	0.6
First ratoon crop	3.3	3.0	2.4	2.7	2.2	0.18
Second ratoon crop	2.6	2.8	2.1	2.5	1.5	0.14
6. Rachis length (cm)						
First year crop	34.8	33.0	30.4	32.1	28.9	3.2
First ratoon crop	27.2	24.9	19.5	21.7	17.8	1.58
Second ratoon crop	21.7	21.0	17.3	20.5	11.2	1.32

Table 4. Influence of spacing and crop duration on yield parameters in tuberose cv. 'Double'

Spacing(cm)	No. of spikes/clump			Number of florets/spike			Cut flower yield (lakh/ha)		
	First year crop	Ratoon crop		First year crop	Ratoon crop		First year crop	Ratoon crop	
		First ratoon	Second ratoon		First ratoon	Second ratoon		First ratoon	Second ratoon
S_1 (45 x 45)	4.1	4.6	5.0	48.2	40.7	35.0	1.99	2.21	2.46
S_2 (45 x 30)	3.1	3.5	4.0	46.2	37.1	32.5	2.27	2.57	2.94
S_3 (45 x 15)	1.5	2.0	2.1	40.7	30.9	25.8	2.26	2.93	3.05
S_4 (30 x 30)	2.9	2.9	3.0	44.3	33.3	31.7	2.72	3.24	3.25
S_5 (30 x 15)	0.9	1.0	1.1	34.0	26.3	17.6	1.94	2.16	2.38
CD *(P = 0.05)*	0.33	0.31	0.39	5.01	3.10	2.84	0.52	0.38	0.51

Table 5. Influence of spacing and crop duration on bulb production in tuberose cv. 'Double'

Spacing(cm)	No. of bulbs/ clump			No. of bulblets/clump			Bulb yield (lakh/ha)		
	First year crop	Ratoon crop		First year crop	Ratoon crop		First year crop	Ratoon crop	
		First ratoon	Second ratoon		First ratoon	Second ratoon		First ratoon	Second ratoon
S_1 (45 x 45)	18.40	24.05	28.10	31.60	38.80	44.00	09.09	11.88	13.88
S_2 (45 x 30)	16.55	21.95	25.10	28.15	34.90	39.40	12.26	16.26	18.59
S_3 (45 x 15)	12.05	15.65	17.65	23.20	28.10	30.40	17.85	23.19	26.15
S_4 (30 x 30)	14.20	18.85	21.00	26.00	31.85	34.85	15.78	20.94	23.33
S_5 (30 x 15)	9.90	12.45	14.15	18.50	21.95	23.90	22.00	27.67	31.44
CD *(P = 0.05)*	1.89	2.78	3.68	5.67	3.97	5.75	2.74	4.45	5.66

Ratoon crops registered higher cut flower yield and bulb production over the first year crop owing to better vegetative growth and higher number of spikes per clump. However, flower quality was inferior which can hinder better price realization in the increasingly quality conscious markets. Tuberose plants, when spaced at a distance of 30 cm x 30 cm, yielded maximum number of cut-flowers without any detrimental effect on flower quality.

From the present investigation it may be therefore inferred that for higher cut flower yield, planting distance of 30 cm x 30 cm and for higher bulb yield 30 cm x 15 cm be adopted for planting tuberose cv. 'Double' in the Saurashtra region of Gujarat. It is further recommended that fresh crop be planted for ensuring superior quality cut flowers.

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(MS Received 3 February 2010, Revised 6 September 2010)

J. Hortl. Sci.
Vol. 5 (2): 138-139, 2010

Short communication

Effect of cultivars and season on grafting success in sapota under Paschim Midnapur conditions of West Bengal

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ABSTRACT

Two sets of experiments were carried out during 2007-08 to assess incompatibility of sapota cultivars to softwood grafting, and to find out the best time for softwood grafting, in a private orchard at Jhargram of Paschim Midnapore, West Bengal. Considerable variation in success of softwood grafting among sapota cultivars was observed. Among ten cultivars studied, CO-2 showed highest compatibility with Khirnee rootstock to softwood grafting, followed by Cricket Ball and DSH-2. There was a total failure in graft-take in cultivars CO-1, DSH-1 and Guthi. Softwood grafting success was highest in sapota when carried out on 1st July (72%) followed by 15th August (70%), 5th June (62%) and 15th June (56%).

Key words : Sapota, softwood grafting, Khirnee, cultivars, incompatibility, season

The sapota or Chiku (*Achras sapota* L.) is one of the most delicious, sweet, pulpy fruits, grown extensively in 24-Parganas (North and South) and Purba Midnapur districts of West Bengal. Due to its tropical nature, the crop is also found to grow well in the drier tracts of West Bengal, like Paschim Midnapore. There is a good demand for planting material of this crop not only in West Bengal, but also in the neighboring states of Jharkhand, Bihar and Orissa. The crop is mainly propagated by grafting on to Khirnee (*Manilkara hexandra* L.) rootstock. Although inarch grafting or approach grafting is universally practised, the method is laborious, time-consuming and also expensive. Currently, an alternative to approach grafting, softwood method of grafting in polythene bags, is becoming very popular. However, its success depends mainly on season of operation and varietal reaction to this method, which need to be standardized for West Bengal conditions. This information is particularly lacking for the western part of West Bengal where the weather is somewhat different from that in other parts of the state.

The study was undertaken in the nursery of MPS farm at Paschim Midnapore where adequate nursery facilities and mother plants are available. The investigation was conducted in 2007 and 2008 following Randomized Block Design using 'Cricket Ball' as the scion. To identify the best time of operation for large-scale production of sapota grafts, grafting was made on 1-year old Khirnee rootstock seedlings, during June to October. Fifty grafts with three replications were made each time. To study varietal response to softwood grafting, scions of ten cultivars, *viz.*, Cricket Ball, CO-1, CO-2, CO3, DSH-1, DSH-2, Guthi, H-7/1, Kalipati and PKM-2 were grafted on Khirnee rootstock on 1st July of 2007 and 2008. Fifty grafts with three replications in each combination were made. The terminal portion of sapota shoot having grayish coloured wood (6-8 mm thick and 6-8 cm in length) was used as scion. Each graft was tied and covered with white polythene (Pepsi-cap) for creating higher humidity around the scion. Grafted plants were kept under partial shade for better success. Plant growth was recorded 90 days after grafting.

Data presented in Table 1 reveal that sapota cultivars responded significantly to softwood grafting, with different degree of success. The highest successful grafts were obtained in CO-2 (85%) variety, followed by 'Cricket Ball' and 'DSH-2' (65%). But, there was total failure of graft in CO-1, DSH-1 and Guthi varieties. Other cultivars like CO-3, H-7/1, Kalipatti and PKM-2 also showed poor response to softwood grafting. The results clearly indicates

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Table 1. Response of sapota cultivars to softwood grafting after three months

Cultivar	Success (%)	Plant height (cm) (extended new growth)	Number of leaves/graft
Cricket Ball	65 (53.73)	2.8	15.4
CO-1	0 (0.00)	0.0	0.0
CO-2	85 (67.21)	6.0	19.0
CO-3	25 (30.00)	1.4	10.0
DSH-1	0 (0.00)	0.0	0.0
DSH-2	65 (53.73)	3.2	17.6
Guthi	0 (0.00)	0.0	0.0
H-7/1	30 (33.21)	2.6	8.8
Kalipatti	25 (30.00)	1.1	8.2
PKM-2	20 (26.57)	2.4	8.0
SEm ±	1.5	0.3	0.7
CD *(P=0.05)*	4.4	0.8	2.2

Figures in parantheses indicate angular transformed values

that graft-incompatibility phenomenon exists between stock and scion of sapota cultivars, which may be attributed to varied woody nature of tissues, differential active-movement of sap, presence of growth promoting/inhibiting factors at the site of graft union hampering cambial activity between stock and scion. Differential response of sapota cultivars to softwood grafting has also been reported by Kulwal *et al* (1988) and Shirol *et al* (2005). Incompatibility in softwood grafting in cultivars was also reported in fruit crops like cashew (Ghosh, 1995) and custard apple (Ghosh and Tarai, 2005). Another interesting observation in this experiment was that cultivars, CO-2, Cricket Ball and DSH-2 [that gave the highest percentage of success (85 to 65%) under Paschim Midnapore, West Bengal Condition], showed less success in softwood grafting under Dharward conditions of Karnataka (Shirol *et al*., 2005). This finding indicates that propagation technique needs to be standardized in each variety for each locality. Growth of the grafted plants in respect of height and leaf production was better in cultivars with higher grafting success compared to that cultivars those performed poorly in softwood grafting.

It is clear from data in Table 2 that success in softwood grafting is significantly influenced by time of grafting. Highest success (70 to 72%) was recorded when grafting was carried out on 1st July and 15th August, followed by 5th and 15th June. Higher grafting success during the early part of monsoon (5th June to 1st July) was mainly due to favourable weather conditions (high humidity and atmospheric temperature) which could have resulted in maximum cambial activity in both stock and scion. Besides,

Table 2. Effect of season on success of softwood grafting in sapota after three months

Date of operation	Success (%)	Number of leaves/graft
5th June	62 (51.94)	13.8
15th June	56 (48.45)	14.0
1st July	72 (58.05)	12.0
15th July	45 (42.13)	14.8
1st August	15 (22.79)	8.0
15th August	70 (56.79)	9.6
1st September	20 (26.57)	7.0
15th September	10 (18.43)	6.0
1st October	0 (0.00)	
SEm ±	1.6	-
CD *(P=0.05)*	4.8	1.6

Figures in parantheses indicate angular transformed values

the scion seemed to be in a physiologically active condition for better sap flow at that time. Although early and middle part of the rainy season (15th August) was found to be better under Paschim Midnapore condition of West Bengal, in Dharwad (Karnataka), the months of April and May were the best suited for softwood grafting in sapota with graft success of 47 to 62% (Sulikeri *et al*, 1997). In Navsari (Gujarat) January and February were congenial for approach-grafting (Bhuva *et al*, 1990) in sapota. Growth of grafts in terms of leaf production was higher in grafts prepared during the early part of rainy season (5th June to 15th July) and leaf number progressively decreased in grafts prepared after 15th July.

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(MS Received 19 August 2009, Revised 15 July 2010)

J. Hortl. Sci.
Vol. 5 (2): 140-143, 2010

Short communication

Effect of drip irrigation and polythene mulch on the fruit yield and quality parameters of mango (*Mangifera indica* L.)

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ABSTRACT

A field experiment was carried out at Horticultural Research Farm, Precision Farming Development Centre, Department of Horticulture, Indira Gandhi Krishi Vishwavidyalaya, Raipur, Chhattisgarh during the year 2009-2010 in Randomized Block Design with three replications and ten treatment combinations (100%, 80%, 60%, and 40% water through drip irrigation system with and without polythene mulch + Basin irrigation with and without mulch). Fruits characters, yield and yield attributing parameter were higher under drip irrigation with 0.6 V volume of water + polythene mulch (T8) and the same characters were lowest under control (Basin irrigation with V- volume of water). Application of black plastic mulch with drip irrigation system can conserve moisture, check the growth of weeds and improve the fruit yield and quality. Water use efficiency was higher under drip irrigation with 0.6 V volume of water + polythene mulch and low under basin irrigation with V volume of water. The net income and benefit cost ratio was also higher under the treatment T_8 as compared to surface method of irrigation.

Key words: Mango, drip irrigation, mulching

Mango (*Mangifera indica* L.) is one of the most important tropical fruits of India. It is known as king of fruits. It is the premier and choicest fruit of India. In Mango production, India ranks first in the world with respect to area (2.20 m.ha) and production (13.79 m.t) with productivity of 6.3 t/ha (Indian horticulture Database, 2008). Mango shares 38 % in area and 21.7% in production of total fruit production of India and this offers bright prospects for boosting the exports.

Chhattisgarh is one of the important mango growing States of India. Most of the area of Chhattisgarh is rainfed and has an immense potential to improve the mango production. Under Chhattisgarh conditions, North Indian varieties mature 15 to 20 days earlier, which results in better market price. Most of the areas are under mango grown as rainfed; it is therefore proposed to find out the optimum water requirement under drip irrigation for mango and to evaluate its effect on fruit yield & quality. Increasing demand for highly efficient irrigation system calls for the use of drip irrigation, which has also been found suitable under adverse conditions of climate, soil and irrigation water (Singh *et al.*, 1989). Keeping the above in mind, the study was carried out to understand the response of mango to drip irrigation with and without polythene mulch.

Layout of Experiment

The study was a part of field experiment designed to compare drip and conventional method of basin irrigation at Precision Farming Development Centre, Department of Horticulture, Indira Gandhi Krishi Vishwavidyalaya, Raipur, Chhattisgarh during the year 2009-2010. The experiment consisted of using treatments *i.e.*, 100%, 80%, 60% and 40% of water (percentage in respect to water requirement of crop) through drip irrigation system having with and without plastic mulch (100 micron) and a control. The distance between lateral-to-lateral was fixed as 10 m and four emitters of different LPH in each plant is placed according to recommended spacing of mango plants (10 m row to row and plant to plant spacing). Experiment was conducted on fifteen years old trees of mango cultivar Dashehari. The treatments were replicated three times in randomized block design. The soil of experimental field was clay-loam which is locally known as *Dorsa* in the region in which available N, P & K were 321.27, 30.83 and 200.02 kg/ha and soil pH was 7.31. The fertilizer doses of N: P: K 200:200:200 (g/tree/year) was applied through irrigation water (fertigation) in two split doses whereas, for surface irrigation system the fertilizer was sprayed after mixing with water in two split doses. Standard cultural practices were

also followed for mango cultivation. The observations on yield and physico-chemical parameters of mango were recorded to know the effect of drip irrigation and mulch. The details of ten treatments are given below:

T_1: Basin irrigation with 1.0 V-volume of water (control)

T_2: Basin irrigation with 1.0 V-volume of water + polythene mulch

T_3: Drip irrigation with 1.0 V-volume of water

T_4: Drip irrigation with 1.0 V-volume of water + polythene mulch

T_5: Drip irrigation with 0.8 V-volume of water

T_6: Drip irrigation with 0.8 V-volume of water + polythene mulch

T_7: Drip irrigation with 0.6 V-volume of water

T_8: Drip irrigation with 0.6 V-volume of water + polythene mulch

T_9: Drip irrigation with 0.4 V-volume of water

T_{10}: Drip irrigation with 0.4 V-volume of water + polythene mulch

Where V = Irrigation water requirement

Estimation of Emission Uniformity

Field emission uniformity takes into account the uniformity of emitter discharge through the system. Keller and Karmeli (1975) defined the emission uniformity as:

$$\text{Emission Uniformity} = \frac{\text{Average of lowest ¼ flow}}{\text{Average of all emitter flow}} \times 100$$

Estimation of Irrigation Water Requirement (V)

The depth of irrigation water for different treatments was calculated depending on the potential evaporation. Reference crop evapotranspiration (ET_0) was calculated using Modified Penman Method (Doorenbos, and Pruitt, 1977). The crop co-efficient (Kc) for different growth stages of mango was selected. The actual crop evapotranspiration was estimated by multiplying the reference crop evapotranspiration, crop co-efficient, area under each plant and wetting fraction.

The quantity of water to be applied was estimated by using the following equation:

$$V = ETo \times Kc \times Ap - (Ap \times Re)$$

Where,

V = Net depth of irrigation (litre/day/plant)

ETo = Reference crop evapotranspiration (mm/day)

Kc = Crop co-efficient

Ap = A x W = Effective area to be irrigated (Sq.m)

A = Area allocated to each plant, 36 sqm apprx.

W = Wetting fraction (0.3-0.5 for fruit crop)

Re = Effective rainfall (mm/day).

Drip irrigation was scheduled on alternate days; hence total quantity of water delivered was cumulative water requirement of two days minus effective rainfall (if rain occurred). The duration of delivery of water to each treatment was controlled with the help of gate valves provided at the inlet of each lateral. In case of basin irrigation, irrigation was scheduled at weekly interval. The cumulative depth of water required for seven days was estimated and supplied to each plant. The water (through surface method of irrigation) was directly applied in the basin with the help of PVC pipes.

Benefit-Cost Analysis

Benefit-cost analysis was carried out to determine the economic feasibility of using the drip irrigation. The interest rate and repair and maintenance cost of the system were 12% and 1% per annum of the fixed cost respectively. The useful life of drip system was considered to be 8 years. The cost of cultivation includes expenses incurred in field preparation, cost of grafted plants, fertilizer, weeding, crop protection, irrigation water and harvesting charges. The income from produce was estimated using prevailing average market price as Rs. 2000 /quintal for drip irrigated with polythene mulch, Rs. 1500/ quintal for drip irrigated without mulch and Rs. 1200/ quintal for surface irrigated, the difference in rates was due to better quality of produce found through drip with mulch as compared to without mulch and surface irrigation. The benefit–cost ratio, from mango cultivation over 1 ha was estimated. The data were analysed statistically as per standard procedure.

Fruit yield and quality

Data on yield with different irrigation treatments are presented in Table –1. Drip irrigation with 60 % V-volume of water + mulch (T_8) recorded the maximum yield (59.92 q/ha) as compared to other treatments and the yield was lowest in control (26.95 q/ha). The yield

Table 1. Effect of irrigation levels on the yield and yield parameters of mango

Treatments	Water applied (cm)	Length of fruits (cm)	Breadth of fruits (cm)	No. of fruits/ plant	Av. Fruit Weight(g)	Yield (q/ha)	Increase in yield (%)	Water use efficiency (q/ha-cm)	Emission Uniformity(%)
T_1	27.50	6.81	4.53	194.31	138.70	26.95	-	0.98	85.10
T_2	27.28	6.95	4.95	222.67	142.15	31.65	17.43	1.16	85.25
T_3	26.32	6.98	4.04	360.27	125.68	45.27	67.99	1.72	87.24
T_4	25.67	8.71	5.13	293.14	153.25	44.92	66.60	1.75	90.25
T_5	23.95	7.07	4.68	278.71	146.12	40.72	51.12	1.70	90.80
T_6	23.12	8.82	5.24	328.53	161.18	52.95	96.47	2.29	93.12
T_7	23.32	8.01	4.90	239.30	148.19	35.46	31.53	1.52	92.72
T_8	18.67	8.89	5.82	366.17	163.65	59.92	122.26	3.21	95.35
T_9	25.70	8.14	4.45	225.23	141.55	31.88	18.27	1.24	91.43
T_{10}	23.09	8.04	4.85	262.08	145.39	38.10	41.37	1.65	93.40
CD at *(P=0.05)*	0.954	0.378	0.210	27.96	4.41	1.12	-	0.102	0.962

Table 2. Effect of irrigation levels on physico-chemical composition of fruits

Treatments	Pulp (%)	TSS (Brix)	Peel (%)	Stone (%)	Acidity (%)	Weed Control (%)
T_1	64.25	17.50	19.25	20.32	0.268	13.67
T_2	65.98	19.50	14.68	19.31	0.210	54.35
T_3	67.98	18.50	12.98	19.04	0.230	34.56
T_4	70.33	20.25	14.08	19.00	0.209	65.39
T_5	68.24	19.98	14.70	19.10	0.228	29.62
T_6	71.58	22.65	13.10	15.32	0.190	85.98
T_7	67.95	21.05	13.02	17.06	0.223	32.10
T_8	72.60	23.35	12.95	14.38	0.178	90.20
T_9	64.00	18.98	15.68	16.50	0.216	30.73
T_{10}	61.72	20.98	15.68	15.54	0.226	68.32
CD at *(P=0.05)*	1.42	0.840	0.903	0.945	NS	12.29

increase was 122.26% over control. This could be due to the water stress the plant has to undergo before the next irrigation. But in case of drip irrigation water is made available in the root zone there by reducing the water stress pressure directly near (Bankar *et al*, 1993).

The variation in water applied for different treatments was due to the variation in pan evaporation and rainfall pattern, as the quantity of water applied was based on pan evaporation. It was observed from the Table-1, that drip irrigation treatments with replenishing 60% of water requirement or the depth of water (18.67cm) given to the plant was optimum for the growth and fruit yield as compared to the surface irrigation. Water required for drip irrigation was lower than that of surface irrigation.

Water Use Efficiency

The irrigation water use efficiency for different treatments was computed from fruit yield and water applied (Table–1). The irrigation water use efficiency in drip irrigation treatments with 0.6 V-volume of water with polythene mulch was maximum (3.21 q/ha-cm) followed by drip irrigation with 0.4 V (1.75 q/ha-cm), 0.6V (2.29 q/ha-cm) and 0.8 V (1.65 q/ha-cm) volume of water. The water use efficiency was lowest in control treatment (0.98 q/ha-cm). The irrigation water use efficiencies of 60% water through drip with black polythene mulch was nearly 3.27 times the water use efficiencies of surface irrigation treatment. Srivastava *et al* (1999) reported that with the highest water application it recorded the lowest water use efficiency. The emission uniformity was highest under drip irrigation with 0.6 V-volume of water + polythene mulch (95.35%) and lowest in basin irrigation with V-volume of water (85.10%).

Fruit quality attributes

The TSS, pulp and moisture content were highest under drip irrigated treatment of 0.6 V volume of water with black polythene mulch and lowest in control. But the peel, stone and acidity were lowest in the same treatment and highest in control, which is better in reference to quality for any fruit crop. Patel and Patel (1998) reported that the increase in yield was mainly because of better growth, bigger size and more juice content in the fruits under drip-irrigated plants. Similarly the weed control percentage was higher under treatment T_8 (90.20%) and lowest in control (13.67%).

Economic-Feasibility

Maximum net returns of Rs. 88,709/ha with B: C ratio of 2.84 was recorded when mango crop were irrigated with 0.6 V-volume of water through drip irrigation + polythene mulch (Table–3). However, in drip irrigated polythene mulch treatments T_4, T_6 and T_{10}, the net returns of Rs. 58,709/ha, Rs. 74,769/ha and Rs. 45,069/ha were obtained with B: C ratio of 1.88, 2.40 and 1.45 respectively. While in case of surface irrigation without mulch the net return of Rs. 12,340/ha was lowest with B: C ratio of 0.61.

Table 3. Cost analysis of mango

S.No.	Particular/Treatment	T1	T2	T3	T4	T5	T6	T7	T8	T9	T10
1.	Fixed costa. Cost of system	-	-	27,000	27,000	27,000	27,000	27,000	27,000	27,000	27,000
	b. Life (yrs.)	-	-	8	8	8	8	8	8	8	8
	c. Depreciation	-	-	3375	3375	3375	3375	3375	3375	3375	3375
	d. Interest cost@ 12 %	-	-	3240	3240	3240	3240	3240	3240	3240	3240
2.	Operation coste. Repair & Maintenance @ 1 %	-	-	2700	2700	2700	2700	2700	2700	2700	2700
3.	Total operational cost (Rs.)	-	-	9315	9315	9315	9315	9315	9315	9315	9315
4.	a. Cost of mulching	-	7816	-	7816	-	7816	-	7816	-	7816
	b. Cost of cultivation	14,000+6,000*	14,000	14,000	14,000	14,000	14,000	14,000	14,000	14,000	14,000
5.	Total cost of cultivation (Rs.) 3+4(a+b)	20,000	21,816	23,315	31,131	23,315	31,131	23,315	31,131	23,315	31,131
6.	Yield (q/ha)	26.95	31.65	45.27	44.92	40.72	52.95	35.46	59.92	31.88	38.10
7.	Selling price (Rs./q.)	1200	1200	1500	2000	1500	2000	1500	2000	1500	2000
8.	Income from produce (Rs.)	32,340	37,980	67,905	89,840	61,080	1,05900	53,190	1,19,840	47,820	76,200
9.	Net Return (Rs.)	12,340	16,164	44,590	58,709	37,765	74,769	29,875	88,709	24,505	45,069
10.	B: C Ratio	0.61	0.74	1.91	1.88	1.62	2.40	1.28	2.84	1.05	1.45

* In surface irrigation without mulch labour charges are extra for weeding, fertilizer application etc.

ACKNOWLEDGEMENT

Authors are thankful to the National Committee on the Use of Plasticulture Application in Horticulture (NCPAH), Department of Agriculture and Cooperation, Ministry of Agriculture, Government of India for providing the necessary funds to conduct this research.

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(MS Received 10 August 2010, Revised 22 December 2010)

J. Hortl. Sci.
Vol. 5 (2): 144-147, 2010

Short communication

Interspecific hybrid developed in *Epidendrum* orchid from the cross *E. radicans* Pav. ex. Lindl. x *E. xanthinum* Lindl.

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ABSTRACT

An interspecific *Epidendrum* hybrid was developed using *E. radicans* (known as 'fire star orchid', 'ground-rooted orchid') as female parent and *E. xanthinum* known as 'yellow orchid' as male parent. The selected line (NRCO-Epidendrum cross/2005-01) was characterized for morphological and floral traits. Flower size (3.5 cm x 3.4 cm) of selected line was bigger than both parents, with bright saffron-orange colour (RHS 44A). Dorsal sepal size (1.8 cm x 0.6 cm), lateral sepal size (1.9 cm x 0.7 cm), petal size (1.8 cm x 0.6 cm), lip size (2.3 cm x 2 cm) and column size (1.1 cm x 0.2 cm) were bigger than in parents. Shape and fimbriated side lobes of lip with deep cleft of anterior margins was similar to the male parent (*E. xanthinum*), except colour. The F_1 progeny of 'NRCO-Epidendrum cross/2005-01' flowered with different red-orange to yellow shades is categorized broadly into three types: Red-orange, Orange-yellow and Yellow. Epidendrums are popularly known as 'Crucifix orchid' and 'Poor man's orchid', have a long flowering period with 2-3 flowerings in a year, and are easy to multiply. These attributes are ideal for popularizing this plant in India as a potted plant as well garden plant.

Key words: Epidendrum hybrids, interspecific hybridization, epiphytes, fimbriated lip, clefted anterior lobe

The genus *Epidendrum* was named so by Carolus Linnaeus in the year 1763, referring to its epiphytic growth habit (meaning derived from the Greek words, *Epi*-"on" and *dendron*-"tree"). *Epidendrum* is often considered a mega-genus consisting of around 1500 species from the neotropical. origin (Hagsater and Arenas, 2005), similar to the genus *Dendrobium* from the old world tropical origin Asia and largely spreading from Carolina, North Louisiana to South Argentina, Mexico, throughout West Indies, Andes and Brazil. However, many species synonymous with *Epidendrum* have been segregated out and resurrected into more than seventeen genera. These species are generally characterized by their reed-stem, growing like tufts, floriferous, bearing flowers with free and spreading sepals, slit rostellum, fringed lip adnate to the column with colour ranging from white, red, orange, green to yellow. This genus, exceptionally, also consists of a few terrestrials and lithophytes by habitat. Epidendrums are popularly called 'Crucifix orchid' and also 'Poor man's orchid', as they are one of the easiest growing orchids and need little attention, unlike the popular *Cymbidium* and *Phalaenopsis* hybrids.

Need for interspecific hybrids in Epidendrum orchids:

Orchid breeding is carried out mainly by commercial firms and is still in its infancy in India. Acclimatization and introductions do not suffice for improving plant wealth in India (Randhawa and Mukhopadhyaya, 1986). Epidendrums are easy to multiply, have a long flowering period with 2-3 flowering spells in a year, suited to tropical & sub-tropical conditions. These attributes are ideal for popularizing these orchids in India as potted garden plants. Synthesis of hybrids using rare and endangered species for commercial purposes will reduce the pressure on their wild relatives (Kishor and Sharma, 2009). Orchids can also be introduced from other countries for commercial use for developing hybrids, as there is no restriction on this at present as per *'Convention on International Trade in Endangered of Wild Flora and Fauna'* (CITES).

Variability in commercial Epidendrum varieties is very low. With an objective to create variability, hybridization was carried out using *E. radicans* Pav. ex. Lindl. and *E. xanthinum* Lindl. as parents, in 1999-2000. The exact origin and collection details of these species were not recorded at this center and there are no scientific reports on introduction of these species in to India, except for a report on *E. radicans* as an alien species by Rao and Mohanan (1983). This species, *E. radicans*, is grown for cut flower and as a potted plant (Teob, 1989). Hence, attempts have been made

earlier to develop an efficient micropropagation method (Chen *et al*, 2002).

Hybridization and *in-vitro* programme:

Epidendrum radicans, popularly known as the 'fire star orchid' and 'ground-root' orchid, was used as the female parent (Fig. 1) and *E. xanthinum* (Syn. *E. secundum*, now called *E. ellipticum* var. flavum Lindl.) (Fig. 2) known as the yellow orchid, was used as the male parent. Hybridization was done by emasculating flowers of the female parent by removing the anther cap and pollinia (that are four in number, with two clusters). Then, fresh pollinia collected from the male parent were attached to the stigma of the column for pollination. Even though the stigmatic surface is highly sticky pollen bags were used for covering the inflorescence to avoid cross pollination by insects. Flower colour turned dark and the floral lip dried up in 3-4 days, when pollination was successful. Mature, ellipsoid capsules harvested at 4-5 months. Seedlings were raised *in-vitro* from seeds contained in capsules, and, flowering was observed after two years planting. Observations on flower colour variations among the progeny and clones selected are described below and presented inTable 1. Morphological characters were recorded at the full bloom stage and colour of flowers was recorded with the help of 'Royal Horticultural Society colour chart'.

Fig 1. Flower of *Epidendrum radicans*

Fig 2. Flower of *Epidendrum xanthinum*

Fig 3. Flower of NRCO-*Epidendrum cross-2005-01*

Description of selected F_1 progeny of 'NRCO-Epidendrum cross/2005-01':

The F_1 progeny of 'NRCO-Epidendrum cross/2005' has flowers of red-orange to yellow shades (Fig. 3). Flower colour variation was categorized broadly into three types: Red-orange, Orange-yellow and Yellow (Fig 6 & 7). The data of the selected F_1 line (NRCO-Epidendrum cross/2005-01) along with it parents are presented in Table 1. Flower size (3.5 cm x 3.4 cm) of selected line was larger than in both parents, with bright saffron-orange colour (RHS 44A). Floral characters like dorsal sepal size (1.8 cm x 0.6 cm), lateral sepal size (1.9 cm x 0.7 cm), petal size (1.8 cm x 0.6 cm), lip size (2.3 cm x 2 cm) and column size (1.1 cm x 0.2 cm) were relatively larger than in either parent, except the width of the dorsal sepal and petal. However, the shape and fimbriated side lobes of the lip and deep cleft of the anterior margins of selected F_1 line were similar to that of the male parent (*E. xanthinum*) excepting colour. Flower colour of the F_1 selected line fell in between colour of the female parent (*E. radicans*) with orange (RHS 28A/25A) and red colour (RHS 53B) being that of the male parent (*E. xanthinum*). The mid lobe and disc colour of F_1 hybrid line was similar to that of the female parent with yellow colour (RHS N25D). But, in the male parent, the disc was of the same colour as sepals and petals, except the colour of crested teeth. In the selected line and *E. radicans*, inflorescence was observed to be a corymbose racemose (Fig. 4) and flowers were closely paniculated, whereas in *E. xanthinum*, the peduncle was as long as the stem recurving and pendulous with sparse flowers (Fig. 5).

Fig 4. Corymbose racemose inflorescence of *Epidendrum radicans*

A hybrid, *E. xobrienianum* (natural cross), derived from *E. evectum* x *E. radicans* reported by John Veich and Sons, (1884-1894) has been recognized by the Royal Horticultural Society, UK, an internationally recognized orchid registration authority. But, *E. evectum*, shown as *E.*

Fig 5. Inflorescence of *Epidendrum xanthinum*

Fig 6. Inflorescence of NRCO-*Epidendrum cross-2005-01*

Fig 7. Flower colour variation among F_1 progeny of NRCO-*Epidendrum cross-2005* (From right: 1-Maroon-red group, 2 & 3–Orange group, 4-Yellow group)

Table 1. Morphological characters* of E. radicans, E. xanthinum and their hybrid (selected clone)

E. radicans (Female parent)	
Plant	*Height :* 24-58 cm, *Leaves* : green, flat & concave, 6-11, 11 cm x 2.7 cm, ovate-oblong, acute-emarginate, less pigmented
Flower	*Peduncle:*slender, terminating into corymbose racemose inflorescence, pedicel straight, yellow in colour; *Flowers:* 20-25, size 3.4 x 3.15 cm, resupinate, *Dorsal sepal*:1.8 x 0.65 cm, orange (RHS 28A/25A), *Lateral sepals* : orange (RHS 28A/25A), 1.75 x 0.68 cm, *Petals* – smaller than sepals, 1.3 x 0.7 cm, orange; *Lip* : 3 lobed, 1.8 x 1.6 cm, yellow (RHS N25D), side lobes fimbriated & slightly darker at margins, mid lobe disc crested with 03 bright yellow teeth, anterior lob moderately clefted, *Column* : short, 2 auricles, semi-terete, 0.8 x 0.2 cm, *Anthers* : 4, yellow & cap yellowish green
E. xanthinum (Male parent)	
Plant	*Height* 41:74 cm; *Leaves* : medium green, 8-12, 8.7 cm x 2.6 cm, oblong-lanceolate, obtuse tip
Flower	*Peduncle* : as long as the stem, curving, terminating into curving and pendulous racemose & loosely paniculated, *Flowers:* 13-15, size 3.5 x 3.3 cm, red (RHS 46B), *Dorsal sepal*:1.8 x 0.6 cm, red (RHS 46B); *Lateral sepals*: 1.6 x 0.6 cm, red (RHS 46B); *Petals* : 1.6 x 0.8 cm, red (RHS 53B); *Lip* : 1.9 x 1.8 cm, flat, 3 lobed, side lobes deeply fimbriated & red, mid lobe crested with 03 bright prominent bright yellow teeth, anterior lob deeply clefted & reflexed; *Column* : moderately long, 0.9 x 0.2 cm;, *Anthers* : 4, yellow & cap yellowish green
NRCO-Epidendrum cross/2005-01	
Plant	*Height:* 32-51 cm; *Leaves* : 9.2 cm x 2.4 cm, dark green colour, more pigmented, ovate oblong, acute-emarginate,
Flower	*Peduncle*:slender, terminating into corymbose racemose inflorescence (Fig. 6), pedicel straight, yellow colour; *Flowers:* 15-23, size 3.5 x 3.4 cm, resupinate; *Dorsal sepal*-1.8 x 0.6 cm, orange (RHS 44A); *Lateral sepals* : 1.9 x 0.7 cm, orange (RHS 44A); *Petals* : smaller than sepals, 1.8 x 0.6 cm, orange (RHS 44A); *Lip* : 3 lobed, 2.3 x 2 cm, orange (RHS 44A), side lobes tripartite, fimbriated & colour similar to sepal colour, mid lobe disc yellow (RHS N25D) crested with 03 bright yellow teeth, anterior lobe deeply clefted; *Column* : long with auricles, semi-terete, 1.1 x 0.2 cm, darker orange (RHS 47 B); *Anthers:* – 4, yellow & cap yellowish green

* at the time of flowering and based on two years' data (2005-06 & 2008-09)

jamiesonis, is a synonym for the former (RHS, UK). However, Epidendrum hybrids developed through systematic breeding were not reported after this and efforts in this direction are lacking. Hence, this new line developed by us can be useful as germplasm stock, and further improvement can be made through mutation breeding, introgression and by hybridization with its close relatives like *Cattleya*, *Oncidium* etc.

ACKNOWLEDGEMENT

The authors thank Ms. Geetamani Chhetri, Technical Person (under 'DUS Testing on Orchids') and Shri. K.B. Gupta (NRC for Orchids, Sikkim) for field assistance.

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(MS Received 22 September 2009, Revised 8 September 2010)

J. Hortl. Sci.
Vol. 5 (2): 148-150, 2010

Short communication

Influence of de-navelling and stalk-end nutrient application on nutrient composition of 'Robusta' banana fruits

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ABSTRACT

The contents of N, P, Mg, S, Fe and Mn in banana fruit increased significantly due to denavelling from 0.32%, 0.086%, 0.12%, 0.024%, 52 ppm and 4.8 ppm, under 'control' to 0.37%, 0.085%, 0.13%, 0.027%, 59 ppm and 6.7 ppm, respectively. Dipping stalk end of the bunch in fresh cow dung enhanced these above nutrients to 0.40%, 0.086%, 0.14%. 0.028%, 63 ppm and 7.6 ppm, respectively. When cow dung was enriched with ammonium sulphate, the fruits showed 0.50-0.51% of N, 0.081-0.090% of P, 0.16-0.23% of Mg, 0.032-0.040% of S, 59-111 ppm of Fe and 8.1-17.8 ppm of Mn. Addition of potassium sulphate further enhanced this effect in respect of K (2.11-2.44%) and Fe (74-115 ppm) in fruit. Increasing level of ammonium sulphate in the blend significantly decreased Ca content of the fruit from 0.24% at 5 g to 0.10% at 25 g. When potassium sulphate was included in the blend, Ca content showed further reduction (0.19% at 5 g to 0.10% at 25g). At 15 g of ammonium sulphate and 7.5 g of potassium sulphate the maximum bunch weight of 27.993 kg was obtained (as against 16.724kg under retention of male bud throughout) corresponding to the enhanced nutrient composition of 2.44% of K, 0.12% of Ca, 0.18% of Mg, 0.033% of S, 115ppm of Fe and 14.9ppm of Mn that may have nutraceutical implications.

Key words: De-navelling, external feeding, nitrogen, potassium, sulphur, 'Robusta' banana, *Musa* sp. (AAA), composition of fruit

Manipulation of bunch size of banana to suit market demands is pratcised in South East Asian countries. Banana plant is supplied with nutrients through soil and foliage, de-navelling (removal of male inflorescence for nutrient diversion) and post-shooting feeding nutrients through the distal stalk-end of rachis (Venkatarayappa *et al*, 1976, Prasanna Kumari Amma *et al*, 1986, Ancy *et al*, 1998 and Ancy and Kurien, 2000) to achieve high yields. De-navelling serves dual purposes of saving mobilization of food into unwanted sink of banana plant as well as earning additional income when excised male bud is used as a vegetable (Singh, 2001). Therefore, an attempt was made to enhance the bunch yield by feeding N, K and S through the excised distal stalk-end of rachis after de-navelling and to determine influence of treatments on composition of mineral nutrients in fruits of "Robusta" banana.

A field experiment was taken-up during 1998-2002 on healthy 'Robusta' banana (*Musa* sp. AAA) plants at flowering stage. The crop was raised on a red clay loam having pH of 6.5, electrical conductivity of 0.3 dS/m, organic carbon of 1.2%, cation exchange capacity of 21.4 cmol (p+)/kg, exchangeable K of 1.3 cmol (p+)/kg, 1*N* KCl exchangeable acidity of 0.5 cmol (p+)/kg and available S of 38 ppm. Rachis at distal end of the bunch was excised along with male bud giving a slanted cut immediately after all the pistillate (female) flowers had set fruits and after 4 bracts were shed (about 15 days of flower emergence). Half a kilogram aliquots of fresh cow dung were blended to form slurry with required quantity of fertilizer [5-25 g of ammonium sulphate (AS) / 2.5 to 12.5 g of potassium sulphate (SOP)] and 100 ml of water. Cow dung contained about 1.4% of N, 0.5% of P, 0.9% of K, 1.8% of Ca, 0.8% of Mg, 0.4% of S, 250 ppm of Fe, 80 ppm of Mn, 64 ppm of Zn and 38 ppm of Cu. The blend was placed in a polythene bag and tied securely to dip the excised rachis into the slurry. The treatments were: control-1 (the male bud retained till harvest along with the male bud); control-2 (de-navelling by excision of rachis 10 cm after the last hand); control-3 (de-navelling and dipping excised distal end of rachis in the slurry of cow dung and 100 ml water); other 5 treatments receiving 5, 10, 15, 20 and 25 g of AS blended with cow dung (applied as in control-3); and

another 5 treatments receiving 2.5, 5.0, 7.5, 10.0 and 12.5 g SOP in addition to 5 to 25 g of AS blended in cow dung as above (applied as in control-3), respectively. The treatments were arranged in a completely randomized design with 3 replications. The cow dung applied was retained till harvest. Uniform bunches carrying 10 hands (having an average number of fingers = 122 ± 2.57) were selected to receive treatments. Harvesting was taken-up at maturity (about 100 days after flowering). At harvest the fruit was sampled, cut into pieces, dried in oven at 70°C and powdered for N analysis by Kjeldahl method. Contents of other nutrients in the digest of the fruit sample in the di-acid (9:4 nitric: perchloric acid) were determined using standard analytical methods (Jackson, 1967).

The contents of N, P, Mg, S, Fe and Mn increased significantly due to denavelling ('control 2') from 0.32%, 0.086%, 0.12%, 0.024%, 53ppm and 4.8ppm, under 'control 1' to 0.37%, 0.085%, 0.13%, 0.027%, 59 ppm and 6.7 ppm, respectively (Table 1). Dipping the stalk end of the bunch in cow dung ('control 3') enhanced the contents of these nutrients to 0.40%, 0.086%, 0.14%. 0.028%, 63 ppm and 7.6 ppm, respectively. This effect was pronounced when the cow dung was enriched with ammonium sulphate and the fruits showed 0.50-0.51% of N, 0.080-0.090% of P, 0.16-0.23% of Mg, 0.032-0.040% of S, 59-111ppm of Fe and 8.1-17.8ppm of Mn. Addition of SOP further enhanced this effect in respect of K (2.11-2.44%) and Fe by showing 74-115ppm in fruit. Increasing level of ammonium sulphate in the blend significantly decreased Ca content of the fruit from 0.24% at 5g to 0.10% at 25 g. Between enrichment of cow dung with ammonium sulphate and ammonium sulphate + potassium sulphate, the latter appeared to enhance the composition of P, K, Ca, Fe and Mn. In the case of P and Ca the differences were not significant. No changes were discernible for Cu and Zn as the content of these nutrients in fruit was in traces.

In respect of N content of fruit, the improvement was fairly uniform at 0.50-0.51% when ammonium sulphate was blended in the cow dung and at 0.49-0.50% when ammonium sulphate + potassium sulphate were used for enrichment of the cow dung in the entire range of these additions. The highest P content of 0.090% was observed when 10g of ammonium sulphate or 5 g of ammonium sulphate + 5 g of potassium sulphate were added to cow dung. In both Ca and Mg, the addition of 5 g of ammonium sulphate to cow dung produced the maximum increase of 0.24% and 0.23%, respectively, while these contents were reduced to 0.15% and 0.19% in the presence of potassium sulphate. Sulphur content was the highest at 20 g of ammonium sulphate (0.04%) followed by 20 g of ammonium sulphate + 10 g of potassium sulphate (0.038%) addition to cow dung. Similarly, the contents of Fe and Mn peaked at 15 g of ammonium sulphate and/or potassium sulphate (111 and 115 ppm), respectively. No changes were discernible for Cu and Zn as the content of these nutrients in fruit was in traces. Substantial enhancement of bunch weight resulted by de-navelling (19.041 kg) and dipping the stalk end in cow dung only (19.904 kg) and enriched cow dung (21.948-27.993 kg). When 15 g of ammonium sulphate and 7.5 g of potassium sulphate were blended in cow dung and applied the maximum bunch weight of 27.993kg was obtained (as against 16.724 kg under 'control 1').

Table 1. Effect of de-navelling and feeding ammonium sulphate and potassium sulphate on composition of 'Robusta' banana fruit

Treatment	N	P	K	Ca	Mg	S	Fe	Mn	Bunch
	%						mg/kg		yield (kg)
Control 1	0.32	0.086	1.73	0.09	0.12	0.024	53	4.8	16.724
Control 2	0.37	0.085	1.87	0.10	0.13	0.027	59	6.7	19.041
Control 3	0.40	0.086	1.98	0.10	0.14	0.028	63	7.6	19.904
5g AS*	0.51	0.081	2.00	0.24	0.23	0.031	87	8.1	23.600
10g AS	0.51	0.090	2.19	0.10	0.16	0.034	74	10.4	25.403
15g AS	0.51	0.080	2.30	0.14	0.19	0.032	111	17.8	25.791
20g AS	0.50	0.082	2.12	0.13	0.17	0.040	73	9.6	24.166
25g AS	0.51	0.086	2.10	0.10	0.16	0.034	59	8.2	22.484
5g AS + 2.5g SOP**	0.49	0.090	2.11	0.15	0.19	0.034	103	13.1	25.545
10 g AS + 5.0g SOP	0.50	0.081	2.17	0.12	0.15	0.036	86	12.1	25.824
15 g AS + 7.5g SOP	0.49	0.086	2.44	0.12	0.18	0.033	115	14.9	27.993
20 g AS + 10.0g SOP	0.50	0.080	2.14	0.10	0.16	0.038	84	12.1	24.837
25 g AS + 12.5g SOP	0.49	0.085	2.13	0.10	0.15	0.029	74	10.6	21.948
SEm (±)	0.005	0.0011	0.028	0.003	0.002	0.0004	1.6	0.021	0.5492
CD (p = 0.05)	0.015	0.0030	0.082	0.010	0.005	0.0012	4.6	0.060	1.6027

*AS – Ammonium sulphate; ** SOP – Sulphate of potash

Removal of male bud caused an increase in the nutrient composition of fruits as also of the weight of the bunch because of: (i) conservation and utilization of energy of nutrients for finger development which would be otherwise lost for opening of the remainder of the flower and (ii) removal of a strong and active competing sink for photosynthates and mineral nutrients despite its smaller size relative to the bunch (Kurien *et al*, 2000; Ancy and Kurien, 2000; Singh, 2001). Further, the translocation of nutrients into the infructescence from such exogenous feeding in 'Poovan (AB)', 'Monthan (AAB)' and 'Nendran (AAB)' varieties has been reported by Buragohain and Shanmugavelu (1986), Sobhana and Arvindakshan (1989). Substantial response of yield of banana fruits as well as the composition of the fruit may be attributed to the presence of other mineral and bio-chemical ingredients of cow dung. The ripening of the fruits after harvest was normal and the fruit quality was not affected by the treatments.

The results indicate that de-navelling and feeding nutrients using enriched cow dung enhanced the mineral composition of banana fruits which can have nutraceutical implications. There is also scope to manipulate the nutrient composition of the fruit further by appropriately modifying the composition of the cow dung blend since the nutrient translocation into the fruits has been demonstrated in this study.

ACKNOWLEDGEMENT

The authors are grateful to the Director, Indian Institute of Horticultural Research, Bangalore for encouragement and facilities provided and to Shri N.K. Kacker, Technical Officer for technical support rendered.

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(MS Received 5 July 2010, Revised 29 November 2010)

J. Hortl. Sci.
Vol. 5 (2): 151-154, 2010

Short communication

Studies on effect of chemical preservatives on physico-chemical changes of beverages in lime and ginger juice with their combinations

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ABSTRACT

The physico-chemical character of lime and ginger RTS and blended RTS were evaluated after addition of potassium meta-bi-sulphite (KMS) and sodium benzoate stored at ambient temperature up to 150 days. Lime and ginger RTS preserved with KMS (0.1%) retained more ascorbic acid and acidity as compared to other treatments. During storage, total soluble solids, reduction and total sugars showed an increasing trend with increasing period of storage under ambient condition in KMS (0.1%) as compared to other treatments. Among the various treatment RTS prepared from ginger juice with KMS 0.1% could be stored for extended period of time for sensory characteristics.

Key words: Lime, ginger, potassium, meta-bi-sulphite, sodium benzoate, RTS, storage

Lime (*Citrus aurantifolia* Swingle) is one of the important fruits of citrus group, acidic in nature and excellent source of vitamin C. India produces 15.42 thousand tonnes of lime per year, raw fruit is freshly consumed and also utilized in preparation of value added products like squash, cordial, syrup, marmalade, pickle, salted lime and dried peel. However, very less work has been done on preservation of lime juice for long duration. Since ancient times, ginger (*Zingiber officinale Rosc.)* has been used as a spice and medicine in India. The total production of ginger is 359 thousand tonnes during 2004-05 (Anonymous, 2005). Ginger can be used in ginger ale, ginger beer, dried pickle, paste and candied ginger.

As lime and ginger juices are health benefitting and refreshing, the ready-to-serve juice of lime, ginger and their blends are very important. Blending not only improves quality and nutrition of basic raw material, but also offers for development of newer product (Nath and Yadav, 2005). Very little work has been done on lime and ginger RTS as well as blended RTS of lime and ginger. Therefore, the present investigation was carried out at post-harvest laboratory of Department of Horticulture, I.G.K.V., Raipur, during the year 2007-08.

Lime and ginger juices were extracted from mature well-ripened lime and fresh ginger procured from local market. Healthy lime fruits and ginger rhizomes were selected and washed thoroughly in running tap water to remove dirt and dust particles. Lime juice was extracted by lime squeezer and filtered with muslin cloth to obtain clear fruit juice free from juice vesicles. In case of ginger, after removal of the peel, rhizomes were cut into pieces with the help of knife and ground in mixer and filtered through muslin cloth to obtain fiber-less juice. After the juice extraction 10% of blended juice of lime and ginger were used for RTS preparation. TSS of 17% and acidity of 0.3 % were maintained by addition of calculated amount of sugar, citric acid and water for all treatments. Fifteen treatments were prepared by combination of different concentration of lime juice (0%, 25%, 50%, 75%, and 100%), ginger juice (0%, 25%, 50%, 75%, and 100%), and chemical preservatives (sodium benzoate 0.1%, potassium meta bisulphate 0.1% and sodium benzoate 0.05% + potassium meta bisulphate 0.05%). The bottles of RTS beverages were kept at ambient condition for further studies up to 150 days. Stored RTS were evaluated at 0, 30, 60, 90, 120 and 150 days of storage for various physico-chemical parameters analysed by using completely randomized design.

Stored RTS were evaluated for ascorbic acid, acidity, TSS, reducing sugar, non-reducing sugar, total sugar, sugar: acid ratio and sensory characteristics. TSS was recorded by using hand refractometer. The ascorbic acid was determined by using 2-6 Dichlorophenol-indophenol dye. The acidity per cent was analysed by titrating the fruit and

rhizome juice with N/10 NaOH using phenolphthalein as an indicator. The reducing sugar, non reducing sugar and total sugar were also determined as per Ranganna (1997). Sensory evaluation of the RTS beverages was done by the panel of ten judges at 30 days interval following the Hedonic rating test as described by Ranganna (1997).

The ascorbic acid and acidity were decreased in lime and ginger RTS but TSS and sugar: acid ratio showed an increasing trend with increase in storage period (Table 1). Maximum ascorbic acid and acidity retention was observed in case of RTS preserved with KMS (0.1%). The loss in ascorbic acid might be attributed to the oxidation of irreversible conversion of L-ascorbic acid into dehydro-ascorbic acid oxidase caused by trapped or residual oxygen in the glass bottles. The decrease in acidity in RTS during storage might be attributed to the chemical interaction between organic constituents of the juice induced by temperature and action of enzymes. Deka (2000) and Deka *et al* (2004) reported similar finding with lime-aonla blended RTS and Nath and Yadav (2005b) with ginger-kinnow squash. The increase in TSS in RTS/ blended RTS, during storage was probably due to conversion of polysaccharides, like pectin, cellulose, starch etc., into simple sugars. Sugar: acid ratio in RTS/ blended RTS showed an increasing trend with increasing period of storage (Table 2). The finding of Singh *et al* (2005) for bael/ blended bael RTS beverages are in close conformity with these results.

Reducing sugar and total sugar increased with the increase in storage period in lime and ginger RTS/ blended

Table 1. Effect of different preservatives on ascorbic acid (mg/100ml) of stored lime and ginger RTS/ blended RTS beverages

Treatments	Storage period (in days)					
	0	30	60	90	120	150
T_1	27.63	27.53	25.30	20.30	10.50	9.50
T_2	27.50	25.16	22.41	18.23	8.43	6.43
T_3	27.60	25.85	24.46	19.23	9.43	7.96
T_4	3.25	3.21	2.30	1.46	1.15	1.03
T_5	3.13	2.58	2.20	1.16	0.95	0.89
T_6	3.16	2.86	2.23	1.36	1.00	0.93
T_7	26.50	26.33	24.30	18.36	9.43	8.86
T_8	26.13	25.43	22.35	15.36	7.56	6.46
T_9	26.36	26.06	24.25	17.36	9.10	8.43
T_{10}	25.36	25.30	23.21	17.36	9.30	8.76
T_{11}	25.33	25.10	22.26	15.46	8.03	7.06
T_{12}	25.40	25.26	23.16	16.70	9.16	8.50
T_{13}	27.36	27.10	25.05	17.36	10.36	9.50
T_{14}	27.20	25.06	22.10	14.60	7.50	6.43
T_{15}	27.30	25.76	24.36	17.26	9.36	8.60
SEm ±	0.07	0.12	0.16	0.13	0.12	0.14
CD*(P=0.05)*	0.19	0.32	0.46	0.36	0.31	0.39

Notation details-
T_1 - Lime juice + KMS 0.1%
T_2 - Lime juice + SB 0.1%
T_3 - Lime juice + KMS 0.05% + SB 0.05%
T_4 - Ginger juice + KMS 0.1%
T_5 - Ginger juice + SB 0.1
T_6 - Ginger juice + KMS 0.05% +SB 0.05%
T_7 - Lime juice 50% + ginger juice 50% + KMS 0.1%
T_8 - Lime juice 50% + ginger juice 50% SB 0.1%
T_9 - Lime juice 50% + ginger juice 50% + KMS 0.05% + SB 0.05%
T_{10} - Lime juice 75% + ginger juice 25% + KMS 0.1%
T_{11} - Lime juice 75% + ginger juice 25% + KMS 0.1%
T_{12} - Lime juice 75% + ginger juice 25% + KMS 0.05% + SB 0.05%
T_{13} - Lime juice 25% + ginger juice 75% + KMS 0.1%
T_{14} - Lime juice 25% + ginger juice 75% + KMS 0.1%
T_{15} - Lime juice 25% + ginger juice 75% + KMS 0.05% + SB 0.05%

Table 2. Effect of different preservatives on TSS (%), acidity (%) and sugar: acid ratio of stored lime and ginger RTS/ blended RTS

Treatment	TSS (%) Storage period (in days)						Acidity (%) Storage period (in days)						Sugar: acid ratio Storage period (in days)					
	0	30	60	90	120	150	0	30	60	90	120	150	0	30	60	90	120	150
T_1	17.00	17.33	17.33	17.47	17.54	17.60	0.30	0.29	0.28	0.26	0.24	0.22	56.60	59.31	61.89	67.19	73.08	80.00
T_2	17.00	17.20	17.30	17.38	17.47	17.56	0.30	0.29	0.26	0.22	0.20	0.16	56.60	59.31	66.53	79.00	87.35	109.75
T_3	17.00	17.20	17.31	17.40	17.48	17.58	0.30	0.28	0.27	0.24	0.22	0.17	56.60	61.42	64.11	72.50	79.45	103.41
T_4	17.00	17.10	17.14	17.21	17.30	17.34	0.30	0.29	0.29	0.28	0.26	0.24	56.60	58.96	59.00	61.46	66.53	75.25
T_5	17.00	17.10	17.12	17.21	17.25	17.30	0.30	0.29	0.28	0.28	0.26	0.22	56.60	58.96	61.14	61.46	66.34	78.36
T_6	17.00	17.10	17.12	17.20	17.29	17.33	0.30	0.29	0.29	0.28	0.26	0.23	56.60	58.96	59.03	61.42	66.50	75.34
T_7	17.00	17.20	17.34	17.38	17.45	17.52	0.30	0.29	0.28	0.27	0.25	0.22	56.60	59.31	61.92	64.37	69.80	79.63
T_8	17.00	17.20	17.30	17.38	17.38	17.45	0.30	0.29	0.27	0.25	0.23	0.18	56.60	59.31	64.07	69.52	75.56	96.94
T_9	17.00	17.20	17.31	17.32	17.43	17.48	0.30	0.28	0.27	0.26	0.24	0.19	56.60	61.42	64.11	66.61	72.02	92.00
T_{10}	17.00	17.20	17.34	17.46	17.54	17.56	0.30	0.28	0.27	0.26	0.24	0.20	56.60	61.42	64.22	67.15	73.08	87.80
T_{11}	17.00	17.20	17.30	17.39	17.47	17.54	0.30	0.27	0.25	0.23	0.21	0.17	56.60	63.70	69.20	75.60	83.19	103.17
T_{12}	17.00	17.20	17.31	17.42	17.50	17.55	0.30	0.29	0.27	0.26	0.24	0.22	56.60	59.31	64.11	66.96	72.19	87.75
T_{13}	17.00	17.20	17.34	17.42	17.50	17.56	0.30	0.29	0.28	0.27	0.25	0.22	56.60	59.31	61.92	64.51	70.00	79.81
T_{14}	17.00	17.20	17.31	17.33	17.40	17.50	0.30	0.27	0.25	0.23	0.21	0.17	56.60	63.70	61.92	75.34	82.85	102.94
T_{15}	17.00	17.20	17.30	17.42	17.47	17.52	0.30	0.28	0.27	0.25	0.23	0.21	56.60	61.42	69.24	69.68	75.95	83.42
SEm ±	-	-	0.05	0.05	0.05	0.05	-	-	0.01	0.01	0.01	0.01	-	-	0.15	0.09	0.15	0.26
CD *(P=0.05)*	-	-	0.16	0.16	0.15	0.15	-	-	0.03	0.03	0.03	0.04	-	-	0.43	0.26	0.44	0.77

Table 3. Effect of different preservatives on reducing sugar (%), non reducing sugar (%) and total sugar (%) of stored lime and ginger RTS/ blended RTS

Treatment	Reducing sugar (%) Storage period (in days)						Non-reducing sugar (%) Storage period (in days)						Total sugar (%) Storage period (in days)					
	0	30	60	90	120	150	0	30	60	90	120	150	0	30	60	90	120	150
T_1	6.62	6.79	6.96	7.13	7.28	7.45	1.36	1.30	1.20	1.14	1.08	1.02	7.98	8.09	8.16	8.27	8.36	8.47
T_2	6.62	6.74	6.84	6.95	7.13	7.25	1.36	1.25	1.16	1.07	0.90	0.80	7.98	7.99	8.00	8.01	8.03	8.05
T_3	6.62	6.77	6.92	7.07	7.22	7.37	1.36	1.27	1.18	1.10	0.93	0.86	7.98	8.03	8.05	8.10	8.15	8.20
T_4	6.11	6.28	6.45	6.62	6.79	6.96	1.12	1.06	1.00	0.94	0.88	0.82	7.23	7.34	7.45	7.56	7.67	7.78
T_5	6.11	6.24	6.36	6.49	6.61	6.73	1.12	1.00	0.90	0.79	0.69	0.59	7.23	7.24	7.26	7.28	7.30	7.32
T_6	6.11	6.26	6.41	6.56	6.70	6.85	1.12	1.02	0.92	0.82	0.72	0.62	7.23	7.28	7.33	7.38	7.42	7.47
T_7	6.50	6.67	6.84	7.01	7.18	7.35	1.24	1.18	1.12	1.06	1.00	0.94	7.74	7.85	7.96	8.07	8.18	8.29
T_8	6.50	6.60	6.75	6.88	7.02	7.14	1.24	1.15	1.02	0.91	0.79	0.68	7.74	7.75	7.77	7.79	7.81	7.83
T_9	6.50	6.65	6.80	6.95	7.07	7.20	1.24	1.14	1.04	0.94	0.84	0.73	7.74	7.79	7.84	7.89	7.91	7.93
T_{10}	6.37	6.54	6.71	6.88	7.05	7.21	1.30	1.24	1.18	1.12	1.05	0.98	7.67	7.78	7.89	8.00	8.10	8.19
T_{11}	6.37	6.50	6.64	6.75	6.90	6.97	1.30	1.18	1.07	0.97	0.85	0.77	7.67	7.68	7.70	7.72	7.74	7.75
T_{12}	6.37	6.52	6.67	6.82	6.97	7.12	1.30	1.20	1.10	1.00	0.90	0.81	7.67	7.72	7.77	7.82	7.87	7.93
T_{13}	6.23	6.40	6.57	6.74	6.92	7.09	1.18	1.12	1.06	1.00	0.93	0.86	7.41	7.52	7.63	7.74	7.85	7.95
T_{14}	6.23	6.33	6.48	6.63	6.80	6.94	1.18	1.10	0.98	0.84	0.70	0.58	7.41	7.43	7.46	7.48	7.50	7.52
T_{15}	6.23	6.38	6.53	6.68	6.84	6.98	1.18	1.08	0.98	0.88	0.78	0.67	7.41	7.46	7.51	7.56	7.62	7.65
SEm ±	0.07	0.01	0.01	0.02	0.09	0.09	0.01	0.01	0.02	0.02	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.03
CD *(P=0.05)*	0.19	0.04	0.04	0.06	0.27	0.26	0.03	0.04	0.05	0.06	0.04	0.04	0.04	0.06	0.06	0.06	0.05	0.09

RTS but, non-reducing sugar decreased with increase in storage period (Table 3). Maximum change in sugar content in lime and ginger RTS/ blended RTS, was observed in RTS preserved with (KMS 0.1%) whereas, minimum change was recorded with RTS preserved with (SB 0.1%). The increase in reducing sugar as well as total sugar were related to the increase in total soluble solids and ultimate decrease in non-reducing sugar in the beverage during storage period. The variation in different fraction of sugar might be due to hydrolysis of polysaccharides like starch, pectin and inversion of non-reducing sugar into reducing. The increase level of total sugar was probably also due to conversion of starch and pectin into simple sugar. The similar findings reported by Deka (2000) and Deka *et al* (2004) for lime-aonla blended RTS and Tiwari (2000) for RTS beverages prepared from guava-papaya.

The organoleptic score reflects the acceptability of the produce to the consumer. The RTS/blended RTS showed decrease in overall acceptability score with increasing storage period up to 150 days under ambient condition (Table 4). The treatment T_4 (ginger juice 100% + KMS 0.1%) had a highest overall acceptability score followed by the T_6 (ginger juice 100% + KMS 0.05% + SB 0.05%) and T_5 (ginger juice 100% + SB 0.1%). However, the RTS of treatment T_9 (lime juice 50% + ginger juice 50% + SB 0.05%) and T_8 (lime juice 50% + ginger juice 50% + SB 0.1%) were least acceptable by the evaluators.

Table 4. Effect of different preservatives on overall acceptability scores of stored lime and ginger RTS/ blended RTS

Treatments	Overall acceptability Storage period (in days)					
	0	30	60	90	120	150
T_1	8.0	7.7	7.5	7.4	7.2	6.6
T_2	8.0	7.6	7.4	7.3	7.0	5.2
T_3	8.0	7.8	7.6	7.5	7.2	6.2
T_4	8.6	8.4	8.3	8.2	7.4	6.8
T_5	8.6	8.4	8.2	8.1	6.2	5.4
T_6	8.4	8.2	7.9	7.7	7.0	6.4
T_7	7.8	7.5	7.3	7.2	6.8	6.4
T_8	7.8	7.5	7.3	7.1	5.4	5.0
T_9	7.7	7.4	7.2	6.9	6.6	6.0
T_{10}	8.4	8.0	7.6	7.3	7.0	6.4
T_{11}	8.3	8.0	7.4	7.1	5.8	5.2
T_{12}	8.1	7.7	7.5	7.2	6.8	6.4
T_{13}	8.2	7.8	7.4	7.1	6.8	6.4
T_{14}	8.1	7.7	7.3	6.9	5.6	5.2
T_{15}	8.2	7.7	7.4	7.0	6.4	6.0

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(MS received 31 May 2010, Revised 26 October 2010)

Event Highlight

NATIONAL SYMPOSIUM ON
MOLECULAR APPROACHES FOR MANAGEMENT OF FUNGAL DISEASES OF CROP PLANTS

December 27-30, 2010

The National Symposium on "Molecular Approaches for Management of Fungal Diseases of Crop Plants" was organized during December 27-30, 2010 at Indian Institute of Horticultural Research, Bangalore jointly by Society for Promotion of Horticulture, Bangalore; Indian Institute of Horticultural Research, Bangalore; Confederation of Horticultural Associations of India, New Delhi and ALCOCERA, an Outreach Progamme on Alternaria, Colletotrichum and Cercospora diseases, with financial support from Department of Horticulture, Government of Karnataka, Bangalore; Department of Science and Technology, New Delhi; Department of Biotechnology, New Delhi; National Bank for Agriculture and Rural Development, Mumbai; Board of Research in Nuclear Sciences, Mumbai, and many others. More than 300 delegates consisting of policy makers, scientists, students and industrialists from across the country attended. The symposium provided a common platform for all stakeholders to share their experience and expertise for better understanding of molecular tools for disease diagnosis, resistance, genetic engineering and prospects of bioactive molecules in disease management. This also provided an excellent forum to researchers for interaction with experts, to take stock of the present status, generate new ideas and plan new strategies for management of fungal diseases in an event of emerging new pathotypes.

Dignitaries on the dais holding released publication Souvenir cum Abstracts on the inaugural day

The symposium comprised the following theme areas:

1. Genetic diversity and molecular diagnosis of emerging fungal pathogens
2. Secondary metabolite and proteome profiling of fungal pathogens
3. Molecular markers for fungal resistance
4. Genetic engineering for fungal resistance
5. Exploitation of bioactive molecules
6. Nanotechnology in fungal disease diagnostics and management
7. Seed and biotech industry perspective
8. Bioinfromatics
9. Student-Academic-Industry interaction

The symposium included three plenary lectures from eminent scientists, invited lectures from experts corporate houses, contributory oral pacers and poster presentations spread over 12 sessions.

Plenary lecture I : Plant bio-security, Trans-boundary movement of pathogens in the wake of WTO agreement ; role of molecular approaches

Dr. S. Nagarajan, Former Chairman, PPV& FRA, New Delhi talked on movement of plant pathogens in the wake of free trade of seed and planting material and cautioned against movement of exotic plant pathogens that could affect national food security. Quick and reliable diagnostic techniques need to be developed and applied for detection of fungal plant pathogens. He emphasized the need for establishment of National clean plant health project for production of disease free seed and planting material in horticultural crops.

Plenary lecture II : *Trichoderma* for plant disease management – a reality or myth?

Dr. A.N. Mukhopadhyay, former Vice chancellor, AAU, Jorhat focussed on *Trichoderma* as a biopesticide and its growth over the last 30 years in India. It is heartening to note that there are about 135 biopesticide units commercially producing different species of *Trichoderma*.

Today, *Trichoderma* formulations are well accepted for seed and soil treatment. Dr. Mukhopadhyay, in his talk, stressed that "*Trichoderma* is gift of god to human being" in plant disease management. *Trichoderma* is a versatile fungus very effective in management of soil-borne fungal plant pathogens such as Pythium, Phytophthora, Fusarium, Rhizoctonia, Sclerotium, Sclerotinia and Macrophomina spp., and in managing bacterial, viral and nematode diseases of crop plants. His talk also covered mass production of Trichodening on liquid and solid substrates, success stories at field level in crops like chickpea, lencil, tea, etc. in management of diseases, and its potential in organic production of several crops. Dr. Mukhopadyay brought out draw backs in the Technology such as non availability of strains tolerant to high temperature, high pH, high soil moisture, and the short shelf life of *Trichoderma*.

Plenary lecture III : Harnessing molecular approaches for diagnosis and management of plant diseases: need for a paradigm shift

Dr. B.L.Jalali highlighted the role of molecular approaches in management of fungal diseases and called upon young researchers to publish quality research papers in high impact journals. He stressed the importance of classical Plant Pathology in disease diagnosis and that in should be a part of identification systems in this era of Molecular Plant Pathology. As Mycologists are dwindling in number, there is a need to give special importance to basic Mycology.

Recommendations emerging from the twelve sessions were presented and discussed:

1) Basic Research

- Genetic diversity among major fungal pathogens in relation to host cultivar specificity, virulence, geographical identity and climatological factors, etc., should be worked out.
- Strains of *Trichoderma* tolerant to salinity and adoptable to a wide range of temperatures need to be identified/developed using recombinant DNA technologies.
- Chemical mechanisms /secondary metabolites / antibiotics and growth promoting compounds from antagonistic microbes responsible for disease suppression or growth promotion of host can be identified and used for development of products.
- There is a need to explore the possibility of developing simple identification system based on secondary metabolite fingerprints using thin layer chromatography for complex plant pathogenic and other fungal genera such as Alternaria, Aspergillus, Cercospora, Fusarium, Penicillium after detailed understanding by HPLC, MALDI TOF-MS and LC-MS.
- Identification of resistant gene analogues (RGAs) can help in the discovery of new resistance genes that can be deployed for improving disease resistance by genetic engineering.
- Use of nanoparticles of copper, silver and other metals for improving efficacy in disease management of fungal pathogens needs to be explored. Nanoparticles can also be employed to construct nano-lattices for antagonistic microbes to improve their bio-control efficiency.
- Identification and quantification of various PR proteins produced in plant defence mechanism and their application for disease management may be looked into.
- In depth study of signalling pathways that govern expression of defence in SAR need to be carried out.

2) Applied Research

- Simple, quick and reliable diagnostic kits need to be developed for seed borne and quiescent fungal pathogens, as mycologists are in short supply for rapid detection. While developing molecular diagnostic kits, data on taxonomy and symptomatology should also be included, along with molecular data.
- Biosensor development can be explored as a reliable and precise tool for detection of seed borne pathogens, to strengthen plant quarantine systems, to avoid introduction of exotic pathogens and to produce quality seed and planting material.
- Explore the possibility of using *Trichoderma* as pre-harvest spray for management of certain pre and post harvest diseases of horticultural crops. This practice will help reduce the residue of fungicide/pesticide in horticultural produce.
- Exploration of *Trichoderma* spp. for ISR against plant disease will further strengthen disease management strategies.
- Potential bio-control agents identified need to be converted into commercial products after generating toxicological data and conducting multi-locational trials.

- Seed treatment is recommended to control soil-borne plant pathogens in order to reduce the cost of application to soil.
- Techniques for selectable marker elimination facilitating successive transformation of transgenic plants with a second desirable trait (stacking of transgenes) may be practised in transgenic research to fulfill biosafety norms, at the same time achieving benefits of gene pyramiding for desirable traits. The public needs to be educated on GM technology.
- Strategies for developing fungus resistant transgenic plants need to analyzed critically before initiatory transgenic work. RNAi technology seems to be a promising area and needs to be explored further.
- Application of genomics together with conventional breeding may be explored wherever possible. Pyramiding of resistance genes in breeding using MAS may be exploited wherever possible.
- In view of development of resistance in pathogens against fungicides, greater emphasis should be laid on bio-control strategies.
- More attention should be paid to use of biotic and abiotic elicitors for management of field and horticultural crop diseases. Use of exogenic elicitors like potassium phosphate, beta aminobutyric acid and salicylic acid for control of foliar diseases needs to be exploited.

3) Future Directions

- National clean plant health management project needs to be initiated on the lines of USDA, USA. Indian Institute of Horticultural Research, Bangalore should take the lead. If required, one scientist may be deputed to USDA to get trained on project development.
- The concept of Indian microbial diversity information system (IMDIS) should be taken up on priority involving various organizations and plant health clinics. As a start, a pilot project may be taken up jointly by NBRI Bioinformatics and IIHR (Dr. P. Chowdappa) on applying IMDIS on ORP Leaf spot diseases. A couple of workshops/meetings may be organized to elaborate upon the IMDIS concept.

4) Policy Matters

- Removal of the term "biopesticide" is recommended for *Trichoderma* based formulations as it is known for its growth promoting effect in many crops. Therefore, these products should be named as "growth promoters". This is to facilitate removal of the tedious process of generating toxicological data for registration purpose in CIB. Alternative terms suggested are i) microbial antagonists, ii) bio-control microbes, iii) microbial supplements, iv) microbial antagonists, etc.
- Bio-fungicides are commercially produced by several companies, but not all are producing good quality products. Hence, the Government should appoint a technical committee to inspect infrastructural facilities and technical manpower competent to produce good quality commercial products before issuing licenses.
- Bio-informatics platform for Indian fungal diversity, disease diagnosis and management should be established at selected institutes which should serve as centres of excellence for rapid identification and monitoring of exotic pathogen threats.

Dr. H.P.Singh, Deputy Director General (Horticulture), ICAR, New Delhi and Chief Guest to the function said that feeding an evergrowing population was a major concern across the country as water, land and workforce engaged in agriculture is declining. Growth in horticulture has increased eight fold since the first Five Year Plan compared to a three-fold increase in field crops. He said that technologies were available to enhance farmers' profitability in horticulture even in wastelands. New diseases and climate change were affecting crops, hampering production. There have been several instances of crops being damaged by sudden and severe fungal diseases in India. In the light of sudden emergence of Late Blight in tomato in 2008, Dr. H.P. Singh said there is a need to keep vigil on imported crops to avoid dangers of introduction of exotic pathogens that may cause extinction of native vegetation. There is also a need to understand such sudden and severe outbreaks and identify factors responsible for the same employing molecular techniques. Dr. H.P.Singh remarked that we should be prepared to develop new crop varieties that resist new races in the event of emergence of new virulent biotypes as in the movement of wheat stem rust Ug 99 race. Field based diagnostic tools, particularly dip stick assays, which can be used at the farm level by farmers is the need of the hour, as PCR based assay is restricted to the laboratory. Disease forecasting models need to be developed to reduce fungicidal load and the cost of production as none of the available models are effective as these are based on regression equations using certain parameters .These models should be refined using leaf

wetness duration and temperature, and, should be web based. Management systems including acquired systemic resistance, nutrition, transferring resistance, silencing of genes, early detection and plant resistance etc., are the future challenges.

Dr. P. Chowdappa, Organizing Secretary, while proposing a vote of thanks remarked that the deliberations over four days ignited young minds and provided a platform for exchange of ideas on molecular technologies.

Awards

Life time achievement award

Dr. H.P.Singh was conferred with the Life Time achievement award by Society for Promotion of Horticulture, Bangalore, for his contributions to development of the horticulture industry in India.

Best student awards

- "Influence of *Trichoderma* and Bacillus based formulations on growth and induction of systemic acquired resistance in tomato"- Mohan Kumar, S.P., Jyothi Lakshmi, M., Shivashankar,S. and Chowdappa,P. [Best Student award – I]
- "Effect of BABA priming of jute against Macrophomina phaseolina infection" -Rudra Ray, Ghosh, A., Dutta, N., Chattopadhyay, C. and Chakrabarti, K. [Best student award – II]
- "Molecular detection and quantification of Phytophthora infestans in tomato using conventional and real-time PCR assays" - Nirmal Kumar,B.J., Madhura, S., Padma Priya, H.V., Reddi Bhargavi, B, Sandhya, H. and Chowdappa, P [Best student award – III]

Best oral presentation

- "Pyramiding of three blast resistant genes (Pi-1(t) + Pi-2(t) Pi-kh) using marker assisted selection into elite Indica cultivar Sambamahsuri" - Srinivas Prasad, M., Ratna Madhavi, K., Madan Mohan, K., Balachandran,S.M. and Viktamath, B.C. [Best Oral paper award – I]
- "Species-specific sequence characterized amplified region (SCAR) markers delineating Colletotrichum gloeosporioides and C.acutatum causing leaf disease in rubber (Hevea brasiliensis)"- Bindu Roy, C., Ravindran, M. and Saha, T. [Best Oral paper award – II]
- "Identification of differentially expressed proteins in chickpea upon Fusarium oxysporum infection" – Yashawant Kumar, Gayatri Gurjar, Vidya S. Gupta and Ashok P. Giri [Best Oral paper award –III]

Best poster presentation

- "QTL mapping of durable and race-specific stem rust resistance in wheat"-Zwart, R.S., Banarjee. R., Shah, N., Bansal, U.K., Sivasamy, M., Singh, D., Miah, H., Martin, P., Raman, H., Bariana, H.S and Gupta, V.S.[Best Poster presentation- I]
- "Development of seed coating formulations of bio-control agents and their effects on seeding growth and blights of tomato" Chowdappa, P., Mohan Kumar, S.P., Bhanuprakash K. and Yogeesha, H.S. [Best poster presentation- II]
- "Extracellular biosynthesis of protein capped CdTe nanoparticles using the fungus Fusarium oxysporum"- Asad S. Syed, Ankita Bedi, Sana Moeez and Absar Ahmad [Best Poster presentation- III]

Three laboratory manuals were released:

1. Laboratory Manual on Biological Control of Plant Diseases
2. Laboratory Manual for Molecular Identification of Plant Pathogenic Fungi
3. Short and Long Term Storage of Fungal Cultures

The following technical bulletins (published under ORP on Leaf spots) were also released:

1. Alternaria Leaf Blight of Cruciferous Vegetables
2. Alternaria Blight of Rape Seed Mustard
3. Alternaria Blight of Sunflower
4. Anthracnose of Grapes
5. Anthracnose of Mango
6. Downy and Powdery Mildew of Cucurbits
7. Phytophthora Bilght of Capsicum
8. Purple Leaf Blotch and Stemphylium Blight of Onion and Garlic
9. Red Rot of Sugarcane
10. Leaf Spot of Mungbean and Urdbean

FORTHCOMING EVENTS

Event	Contact details
I International Symposium on Sustainable Vegetable Production in South-East Asia March 14-17, 2011, Salatiga (Central Java), Indonesia	Prof. Dr. Stefaan De Neve, University of Glent, Coupure Links 653, 9000 Glent, Belgium. E-mail: stefaan.deneve@ugent.be or Dr. Sri Rochayati, Indonesian Soil Research Institute, Jl. Juanda, Bogor, Indonesia E-mail: sri.rochayati@gmail.com E-mail symposium: VegSEA2011@ugent.be
International Symposium on Organic Matter Management and Compost Use in Horticulture April 4-7, 2011, Adelaide, Australia	Mr. Johannes Biala, P.O. Box 74 Wynnum Queensland 4178, Australia Phone: (61)7-39011152, Fax: (61)7-33962511 E-mail: biala@optusnet.com.au Web: http://compost-for-horticulture.com/
National Symposium on Harnessing biodiversity for biological control of crop pests May 25-26, 2011, Bangalore, India	Dr. T. Venkatesan, Organizing Secretary, Society for Biocontrol Advancement, National Bureau of Agriculturally Important Insects, P.B. no. 2491, Bangalore – 560 024, India Phone: 080-23511982/998 Extn. 337 Fax 080-23411961 E-mail: chysopa1@yahoo.com Website: http://www.nbaii.res.in
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Published by Society for Promotion of Horticulture, IIHR, Bangalore-560 089. E-mail: sph@iihr.ernet.in
Chief Editor: Dr. A. Krishnamoorthy, E-mail: jhs@iihr.ernet.in
Printed at Jwalamukhi Mudranalaya Pvt. Ltd., 44/1, K.R. Road, Basavanagudi, Bangalore-560 004, Ph: 080-26601064
E-mail: jwalmuki@gmail.com

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SUBJECT INDEX - VOL. 5 (1 & 2) 2010

ACKNOWLEDGEMENT

The undersigned acknowledges with thanks services rendered by the following external referees during 2010 for critically going through the manuscript of research papers submitted by authors to *Journal of Horticultural Sciences*, with their comments:

Dr. Aghora, T.S. Bangalore
Dr. Awasthi, M.D. Bangalore
Dr. Atul Grover Meerut
Dr. Ashok Halepyatti Bangalore
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JOURNAL OF HORTICULTURAL SCIENCES

(Form IV)

Place of Publication	:	Bangalore – 560 089
Periodicity of publication	:	Half yearly
Printer's Name	:	Mr. C.S. Shivakumar
Nationality	:	Indian
Address	:	Jwalamukhi Mudranalaya Pvt. Ltd. 44/1, K. R. Road, Basavanagudi Bangalore – 560 004
Publisher's Name	:	Society for Promotion of Horticulture
Address Hessaraghatta Lake P.O. Bangalore – 560 089	:	Indian Institute of Horticultural Research
Editor's Name	:	Dr. A. Krishnamoorthy
Nationality	:	Indian
Address Hessaraghatta Lake P.O. Bangalore – 560 089	:	Indian Institute of Horticultural Research
Names and addresses of individuals who own the journal and partners or share - holders holding more than one per cent of the total capital	:	Society for Promotion of Horticulture Indian Institute of Horticultural Research Hessaraghatta Lake P.O. Bangalore – 560 089

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Krishnamoorthy, A. and Mani, M. 2000. Biological Control of Pests of Vegetable Crops.p367-78. In: Biocontrol Potential and its exploitation in sustainable Agriculture. Vol. 2: Insect Pests. Upadhyaay, R. K. Mukerji, K. G. and Chamola, B.P. (ed.). Kluwer Academic / Plenum Publishers, New York

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