MICROPROPAGATION AND NUTRITIONAL STUDIES OF TISSUE CULTURED BANANA VAR. GRANDE NAINE

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DIVISION OF HORTICULTURE
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MICROPROPAGATION AND NUTRITIONAL STUDIES OF TISSUE CULTURED BANANA VAR. GRANDE NAINE

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CERTIFICATE

This is to certify that the thesis entitled **"Micropropagation and nutritional studies of tissue cultured Banana Var. 'Grande Naine' "** submitted in partial fulfilment of the requirements for the degree of **Doctor of Philosophy** in Horticulture to the University of Agricultural Sciences, Bangalore, is a bonafide record of research work carried out by **Mr. Raju Besagarahally** under my guidance and supervision and that no part of this thesis has been submitted for the award of any other degree, diploma associateship, fellowship or other similar titles.

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INTRODUCTION

I INTRODUCTION

Banana is one of the important fruit crops in International trade and is grown in all tropical and subtropical regions of the world. The total production is estimated to be 76.40 m. tonnes (Anon., 1992). It ranks third in importance among the fruits of the world and is consumed all over the world. It is rather a staple food in many African countries.

Though, India inherits immense wealth of banana germplasm, it is still the second most important fruit crop of the country both in area and in production, accounting for nearly 12 per cent (40 lakh ha) of the total area under fruit crops and for over 30 per cent (10.4 m. tonnes) of the total fruit production. In spite of being one of the largest producer of banana the export of bananas is very meager accounting to Rs. 4.48 million during 1993-94 (Chadha, 1995).

Conventional clonal multiplication of banana, which is through suckers is seriously limited by its low rate of multiplication which ranges between 5-10 per year (Vuylsteke and De Langhe, 1985). Apart from the slow rate of sucker production, multiplication by suckers also encourages the spread of many pathogens which results in a significant loss of productivity (Cronauer and Krikorian, 1987). Bananas are susceptible to a broad range of diseases like Panama wilt, Sigatoka leaf spot, cucumber mosaic virus, bunchy top virus and root burrowing nematodes.

The main advantages of in vitro propagation techniques are rapid multiplication of plants with known desirable characters, free from pests and diseases, high survival rate during field establishment, vigorous growth, retention of healthy leaves, uniform flowering, shortened harvesting period and higher yields.

The successful regeneration of shoots and/or multiple shoots, plantlet formation of different cultivars of banana belonging to different genomic groups *in vitro* have been reported by several research workers (Doreswamy *et al.*, 1983; Laxmikanth and Nataraja, 1988). The *in vitro* establishment of shoot tips and morphogenesis appears to be profoundly influenced by genotype than by any other factors. Therefore, media composition and cultural environment often need to be varied from one species to the other. Even, closely related varieties differ in their requirement for optimum and quicker morphogenetic response (Krikorian, 1989).

According to Cronauer and Krikorian (1986), variations in *in vitro* establishment of shoot tip culture of banana can be due to differences in vigor, cell division activities and apical dominance in different genomic groups. However, only a limited number of banana cultivars have been tried for *in vitro* multiplication. Information on multiplication rates, behaviour of tissue cultured banana plants in the field and its performance, varietal response to tissue culture is very scanty.

Banana being a gross feeder, requires high amount of nutrients for proper growth and production. It is estimated that a crop of fifty tonnes of banana in one hectare removes 320 kg N, 32 kg P_2O_5 and 925 kg K_2O every year (Lahav and Turner, 1983). As such, banana crop requires more of nitrogen and potash for its growth and production compared to phosphorus. It is estimated that the expenditure on fertilizers alone amounts to 25 to 30 per cent of the total cost of production. Hence, to ensure high yield of quality banana judicious and timely application of adequate nutrients is of paramount importance.

Nitrogen being a chief promoter of growth, its supply largely affects the growth and fruiting through controlling the utilization of carbohydrates (Claypool, 1936). Deficiency of nitrogen in the initial stages of growth results in poor meristem development, and ultimately in lower yields. A total deficiency of nitrogen would seriously impair growth beyond flowering stage (Charpentier and Martin-Prevel, 1965).

Indian soils are generally known for their high potassium content (Joseph, 1969). In spite of it, banana requires high amount of potassium mainly to maintain adequate fertility in the soil as potash does not move easily in the soil. Potassium plays an important role in all metabolic activities, improves quality and shelf life of fruits and helps the plant to tolerate adverse effects of drought, pests and diseases (Huber, 1985; Mengel, 1985). The potassium status of the plant during floral initiation and subsequent floral differentiation contributes to overall bunch development (Alexandrowicze, 1955; and Obiefuna, 1984).

Adequate information on banana nutrition can be found in the literature, but most of this concerns plants raised from suckers. Very little or no information can be found on the growth dynamics, culture and nutrition of banana plants obtained through tissue culture. The frequency of application and the amount of fertilizer has to be modified for tissue cultured banana compared to suckers as the former have a well developed root system. This clearly highlights the need for research on these aspects and is expected to provide vital information of practical benefit to banana growers. With this background, a study was taken to standardize *in vitro* propagation of Grande Naine

cultivar of banana and its nutrient requirement in the field for higher yields of superior quality fruits, with the following specific objectives.

1. To standardize the culture media and growing conditions for propagation of Grande Naine cultivar under *in vitro* conditions.

2. To develop hardening techniques for *in vitro* propagated banana plantlets.

3. To study the influence of varying levels of N and K on growth dynamics, yield and quality of fruits.

4. To study the influence of nutrients on dry matter accumulation, its distribution,total nutrient uptake and benefit-cost ratio.

REVIEW OF LITERATURE

II REVIEW OF LITERATURE

The available literature on propagation on banana through tissue culture and the nutritional requirements of tissue cultured banana is presented in this chapter.

2.1 Tissue culture studies

2.2 Nutritional studies

2.1 Tissue culture studies

2.1.1 Role of tissue culture in banana

The commercial application of tissue culture techniques for vegetative propagation of cultivated plants started more than 20 years ago. Conventional plant perpetuation has been replaced by large scale micro propagation in various horticultural crops. The primary objectives are higher productivity, elimination of pathogens, rapid and large scale multiplication of identical plants and cloning of hybrids, selected individuals/mutants and of parent plants for F1 hybrid seed production, selection of somaclonal variants and induced variants, and preservation of germplasm for long term storage. The clonal propagation provided a true-to-type large scale production of selected hybrids, only by culturing explanted organs or small fragments of tissue or single cells under *in vitro* conditions. Currently, 180-200 million plants are produced worldwide every year through micropropagation (Reuther, 1990).

Commercial micropropagation is based on separate shoot and root generation system. The physiological state of the donor plants, culture conditions, greenhouse and

field growth are of high relevance to microplant production. The influence of light and temperature, nutrition conditions and of the developmental stage of the donor plants on the survival rate of the primary explants and their regeneration potential are important factors for micro plant propagation. The limiting factor for commercial micropropagation is the difficulty in establishing various crops in the greenhouse. *In vitro* plantlets reveal morphological, anatomical and physiological aberration like vitrification, which is characterized by an abnormally high water content of the leaf and stem tissues, reduced epicuticular wax formation and a disturbed mechanism of stomatal movement. The low survival rate of vitrified plantlets is due to the reduced lignification of the shoot, twisted and chlorotic leaves and uncontrolled water release. The water vapour saturation and the accumulation of ethylene and CO_2 in the atmosphere of densely sealed containers are considered to be responsible for these aberrations (Morel and Martin, 1952; Morel, 1960; Murashige, 1974; Litz, 1984; 1985).

Multiplication of banana is usually by vegetative means through buds, suckers, portion of corm or bits, since, the edible clones of banana are parthenogenetic and hence seedless or seed sterile. Sword suckers are most desirable, although, they frequently carry pathogens and pests, especially nematodes, such as *Radopholus similis*, the borer, *Cosmopolites sordides*, Race 4 of *Fusarium oxysporum* f.sp. *cubense* and viruses such as cucumber mosaic virus (CMV) and bunchy top. Apart from these, they have lower survival rate, are less vigorous and non-uniform in growth, which result in loss of productivity (Vuylsteke and De Langhe, 1985). Although the conventional methods of vegetative propagation in banana have reached commercial

acceptability, for want of better alternatives, tissue culture techniques have been shown to have definite and indispensable advantages over the former. Also, an increased area under banana cultivation has increased the demand for planting materials. Tissue culture ensures an extremely rapid rate of multiplication of disease free plants. The main advantages of *in vitro* propagation techniques for banana as reported by many workers are as follows:

1. The plants can be rapidly multiplied from a mother plant of known desirable characters.
2. Plantlets show high survival rate during field establishment.
3. Plantlets are cheaper and easier to propagate and also to transport.
4. Plants are more uniform in height and retain more healthy leaves.
5. Plants exhibit more vigorous growth and high fruit yields.
6. Plants are uniform in flowering with a short harvesting period.
7. Plants are free from pests and diseases.

2.1.2 Feasibility and methods of *in vitro* micropropagation techniques in *Musa*

Banana and plantain improvement through *in vitro* techniques provide wider scope to overcome the various problems faced. Shoot tip or meristem culture plays a vital role in facilitating rapid multiplication of the desired clones. Damasco and Barba (1985) reported that in Saba banana (BBB), an average of ten new shoots per subculture were regenerated from shoot tip culture at two month intervals. An increase in multiplication from one sucker to two lakh plantlets in ten months was projected,

assuming 80 per cent survival. The plants rooted easily *in vitro* and readily survived when transplanted to field. A maximum multiplication rate was obtained *in vitro* in Dwarf cavendish and plantain cv. Agbagba, producing more than 45,000 and ten lakh plants per year with an average proliferation rate of 3.5 and 15 tips per plant, respectively. After the first successful "Shoot tip culture" attempt in *Musa* by Ma and Shii (1972 and 1974), De Guzman *et al.*(1980), Doreswamy *et al.* (1983) Cronauer and Krikorian (1984a, 1986 and 1988), Jarret *et al.* (1985a) and Banerjee *et al.* (1986b) have all reported success in shoot-tip and /or meristem-tip culture techniques and have developed aseptic culture procedures in a wide variety of banana and plantain cultivars. Further, there are reports on successful callus/cell/protoplast culture (Cronauer and Krikorian 1986), embryo culture (Cox *et al.*, 1960) inflorescence tip culture (Cronauer and Krikorian, 1985; Banerjee *et al.*, 1986a, Doreswamy and Sahijram, 1989) and somatic embryogenesis (Jarret *et al* 1985a; Rao *et al.*, 1982; Novak *et al.*, 1987; Escalant and Teisson, 1988) of banana; these techniques, however, are mostly useful in crop improvement programmes.

2.1.2.1 Shoot-tip culture

The apical meristem and/or shoot-tip culture technique offers an efficient tool for rapid clonal micropropagation and large scale production of pathogen free plant material. A meristem culture technique first developed by Ma and Shii (1972) and Doreswamy *et al.* (1983) suggested that decapitation of terminal buds was necessary to release the axillary buds from apical dominance used for initiating lateral shoot proliferation. Ma and Shii (1972) opined that shoot proliferation was dependent on the

proliferation. Ma and Shii (1972) opined that shoot proliferation was dependent on the presence of 340 mg/l adenine sulphate, 2.0 mg KI/l and 100 mg/l of tyrosine/l in the medium. A high rate of shoot proliferation can also be obtained in liquid cultures even in the absence of growth regulators. Ma and Shii (1974) and De Guzman *et al.* (1980) demonstrated that rapid proliferation rates were dependent on a high concentration of cytokinins (15 % coconut water (CW) and 5.5 mg/L BA). Doreswamy *et al.* (1983) cultured axillary buds on MS medium containing 15% CW and 10.00 mg/l BA. Krikorian and Cronauer (1984) and Cronauer and Krikorian (1984a) also used similar medium. The rooting of regenerated plants has been reported to occur on medium containing 5 mg/l of IBA with 0.25% (W/V) activated charcoal. Subsequently, several research workers from various parts of the world have reported success in shoot/meristem-tip culture technique in a number of cultivars of bananas and plantains which further helped in standardization of *in vitro* micropropagation procedures. A study conducted by Banerjee and De Langhe (1985) revealed a high proliferation rate in the variety Asamiensa(24.4) followed by Bluggoe (BBB)(20.4), Silk (AAB) (16.00) and a low rate in Dwarf Cavendish (AAA) (9.4). The cultivar Saba (AAA) recorded proliferation rate of 31.1 (Jarret *et al.*,1985a). The experiment conducted by Vuylsteke and De Langhe (1985) indicated proliferation rates of 19.5, 17.4, 16.7, and 5.7 with Jackson Agbagba, French Sambre of AAB genome and Zizi of BBB genome, respectively. Among the cultivars studied by Wong (1986) cv. Mysore (AAB), IC-2 (AAAA) and Lacatan (AAA) showed proliferation rates of 6.6, 3.3 and 2.6, respectively, in shoot tip culture. Zamora *et al.* (1986) recorded a proliferation rate of 3.00 with Genome (ABB) of Cv.Cardada in shoot tip culture. In the studies carried

out by Shanmugavelu *et al.* (1987), proliferation rates of 7.35 and 5.70 in genomes cv. Matti (AA) and Genome (AAB) cv. hybrid CO-1 through shoot tip culture were observed.

2.1.2.2 Inflorescence tip culture

Ma *et al.* (1978) from Taiwan were the first to report on the induced reversion of floral apices to vegetative shoots in banana (*Musa cavendishi* Lamb). Further, Cronauer and Krikorian (1985, 1988) and Bakry (1985) have also reported induction of vegetative shoots from floral apices in Grande Naine and Poyo 901 of Cavendish group, respectively. Jarret (1986) and Cronauer and Krikorian (1985) reported that isolated terminal indeterminate floral apices of Dwarf Cavendish could be induced to reinitiate into vegetative growth mode directly without the formation of callus.

2.1.2.3 Cell/Callus/Protoplast culture:

Mohan Ram and Steward (1964), De Guzman *et al.* (1980), Vuylsteke and De Langhe (1985), Banerjee *et al.* (1986a) and Cronauer and Krikorian (1988) have all reported the induction of callus in banana. Similar observations were reported by Rao *et al.* (1982), Cronauer and Krikorian (1985), Jarret *et al.* (1985a) and Banerjee *et al.* (1986b). However, difficulty in maintenance of proliferating callus has been a major constraint. Chen and Ku (1985), Matsumoto (1986); Fitchet (1987); Cronauer and Krikorian 1987); Hwang and Chi (1988) observed adventitious shoot production from calloid of triploid desert banana "Highgate". The cell/callus/protoplast cultures are very

useful in breeding and recombinant technology , but not exploited for commercial propagation in banana.

2.1.2.4 Somatic embryogenesis

Rao *et al.* (1982) reported the occurrence of pro-embryo like structures in callus derived from floral apices of banana cultured in the presence of 2,4,5-trichloro phenoxy acetic acid. Cronauer and Krikorian (1983) reported the occurrence of somatic embryos from callus in liquid medium. Jarret *et al.* (1985a), however, suggested that the mature structures observed were roots and not embryos. Banerjee *et al.* (1986a) obtained pro- embryos when thin layers of meristamatic tissue excised from proliferating shoot tips were cultured in liquid MS medium supplemented with 2,4,5-T. Cronauer and Krikorian (1988) reported the production of calloid structures and regeneration of adventitious shoot-tips. Sannasgala *et al.* (1987) also reported mass production of proembryo-like structures in *Musa* [ABB] Cv.Bluggoe.

2.1.2.5 Embryo culture

In vitro culture of *M. balbisiana* embryo was reported by Cox *et al.* (1960). Intact seeds were surface sterilized, embryos dissected, and cultured. Rowe and Richardson (1975) reported upto 50 per cent germination of excised banana embryos.

2.1.2.6 *In vitro* germplasm storage:

In vitro methods are assuming an increasingly important role in the conservation of plant genetic resources, particularly for clonally propagated material, and for species which are difficult to conserve as seeds. Conventional germplasm conservation of a vegetatively propagated crop like *Musa* is replete with many problems (De Langhe, 1984). The use of shoot tip culture techniques to maintain such collections has considerable advantage. *In vitro* cultures occupy a relatively small amount of space and they are suitable for international exchange of germplasm.

Recently, a general method has been described for the establishment of a regenerable embryogenic cell suspension from proliferating meristems (Dhed'a *et al.*, 1991), and has been successfully applied to several genetically distinct cultivars. Protoplasts have also been isolated from embryogenic cell suspensions and plants have been regenerated through somatic embryogenesis at high frequency (Panis *et al.*, 1993).

2.1.3 Stages in *in vitro* clonal micropropagation of *Musa*

2.1.3.1. Type of explant

The type of explant, its size, and the manner in which it is cultured, can all affect the initiation of cultures and further morphogenetic responses (Murashige, 1974). There is an optimum size for explants used for initiating cultures. Very small size of explants, whether they are shoot or meristem or tip fragments of whole plant tissues or pieces of callus, do not survive well in culture. Similarly, large explants may be difficult to easily manipulate and decontaminate effectively. When the tissues are cut,

difficult to easily manipulate and decontaminate effectively. When the tissues are cut, the cut surface turns brown due to oxidation of polyphenols into quinones which equally damage the cells and affect the survival of explants *in vitro*. The uptake of mineral salts by cultured plant tissues is likely to vary according to the size of explant irrespective of the solid or liquid media (George and Sherrington, 1984).

2.1.3.2 Use of growth regulators:

Selection of appropriate combinations of plant growth regulators is the most important aspect in developing a successful protocol for tissue culture. Ma and Shii (1972) observed the beneficial effects of adenine sulphate in shoot proliferation of banana cultures when added to Smith and Murashige's (1970) basal media. De Guzman *et al.* (1980) reported that high proliferation rates in banana depends on a high concentration of cytokinins and coconut water. The inclusion of cytokinins and auxins are essential for proliferation of cultures (Banerjee and De Langhe, 1985). For *Musa* shoot tip culture, only cytokinins and auxins are required for inducing proliferation of shoots (Doreswamy *et al.*, 1983). The most commonly used auxins are IAA, NAA and IBA. Obviously, BA is the cytokinin of choice for induction of shoot bud proliferation *in vitro*, and BA has been found to be superior to kinetin (Wong, 1986 ; Zamora *et al.*, 1986).

2.1.3.3 Shoot /Multiple shoot development:

Stimulation of axillary branching and/or adventitious bud formation is accomplished by placing an intact or fragmented shoot apex onto a basal medium supplemented with 0.2 to 10 mg/l of cytokinin, appears to be the primary factor affecting the rate of proliferation (Vuylsteke and De Langhe, 1985). The shoot multiplication rate is in turn a result of cytokinin concentration in the culture medium (Damasco and Barba, 1985). Banerjee *et al.* (1986b) who studied the regeneration pattern in *Musa* shoot tip cultures in the presence of 1 uM IAA and 1 uM BA, observed no proliferation. In the presence of higher concentrations of BA, however, multiple shoot buds were produced, and the entire rooted plantlets were readily recovered and transferred to soil.

2.1.3.4 Rooting of explants

Root formation can be readily induced on cultured *Musa* shoots. The individual shoots from a shoot cluster are usually separated first. Ma and Shii (1972) transferred shoots to sphagnum moss and allowed rooting to occur naturally. Other investigators have employed a number of auxins. Berg and Bustamante (1974) found that root formation was a slow process and NAA was effective in inducing rooting. Rooting in *Musa* cultures was easily induced when individual shootlets were transferred to basal medium (Sandoval, 1985). However, addition of auxins to the basal medium may stimulate further root growth. Apart from NAA, IBA is also very effective in inducing rooting at 1 mg/l (Banerjee and De Langhe, 1985).

Cronauer and Krikorian (1984a and 1984b) reported no differences in the root inducing effects of IAA, NAA or IBA. In the presence of 0.025 per cent activated

Cronauer and Krikorian (1984a and 1984b) reported no differences in the root inducing effects of IAA, NAA or IBA. In the presence of 0.025 per cent activated charcoal, MS basal medium supplemented with 1 mg/l IBA or NAA was found suitable for rapid root initiation. When rooting individual shoots, care should be taken to ensure that they are free of developing secondary axillary buds, the occurrence of which inhibits root formation and enhances multiple shoot development (Jarret *et al.*, 1985b).

Studies on the effects of activated charcoal (AC) have shown that when shoots are grown on a medium containing coconut water (CW) the number of roots and overall root length can be enhanced. The use of auxins such as NAA, IBA or IAA with extremely low levels of AC (0.025%) gave the most effective root inducing medium. When shoots are placed on such a medium, a sufficient number of roots are formed within 3-4 weeks to support the young plantlet (Cronauer and Krikorian, 1984b).

2.1.3.5 Hardening

One of the major obstacles in the application of tissue culture methods for plant propagation has been the difficulty in successful transfer of plantlets from the laboratory to the field (Wardle *et al.*, 1983). The reasons for such a difficulty appears to be related to the dramatic change in the environmental conditions. The environment of the culture vessel is one of low light intensity, with very high humidity (generally 100 %) and poor root growth, while the green house and/or field conditions are typified by very high light intensity, low humidity, and mycroflora (Desjardins *et al.*, 1987). Several workers have developed protocols to overcome some of these constraints.

The plantlets obtained through tissue culture techniques should have proportionate roots that are capable of supporting further growth and development. They are usually transplanted into compost and kept in partial shade at a high ambient humidity for several days. A suitable environment is often created by covering the plantlets either with glass or clear polyethylene or by subjecting them to intermittent misting. Plants are finally hardened by gradually reducing the humidity and increasing the light intensity.

The term media is sometimes used to describe the mixtures of materials such as peat, perlite, vermiculite, pumice, rock wool, sand and soil used for transferring the plantlets from *in vitro* conditions. Compost commonly used for rooting conventional cuttings is suitable for transferring these plantlets, but there may be marked differences in root growth and plantlet survival with different media (Rodriguez *et al.*, 1987). Peat may prove to be too acidic substrate for some species and some kinds of vermiculite are too alkaline. Apart from having a neutral or only slightly acidic pH, compost or other substrates need to have high water holding capacity and still provide good aeration.

Since there is a close relationship between humidity, illumination and temperature, the use of humidity tends to reduce desiccation of leaves. Gradual increase in illumination and decrease in humidity can help in successful transplanting of tissue cultured plantlets in a green house and subsequently in the field.

2.2 Nutritional studies

Banana is one of the most important tropical fruit crops, being a gross feeder it requires liberal supply of nutrients, specially nitrogen and potash, which determine the growth and productivity. Hence, application of adequate quantity of nutrients is of paramount importance to ensure high yield of quality bananas.

Considerable research work has been done to standardize not only the quantity of different nutrients but also the appropriate time and method of application to obtain the maximum fertilizer use efficiency under varying agro-climatic conditions in different parts of the country. The work done on application and uptake of various nutrients to banana plants has been reviewed under the following headings.

2.2.1 Role of nitrogen on growth, yield and quality of banana.

2.2.2 Role of potash on growth, yield and quality of banana.

2.2.3 Role of NPK on growth, yield and quality of banana.

2.2.4 Role of split application of nutrients on growth and yield of banana.

2.2.5 Role of nutrients, on their uptake and distribution.

2.2.1 Role of Nitrogen on growth, yield and quality of banana

Nitrogen is considered as the most important nutrient element for the growth of banana, being almost universally in short supply, even in very fertile soils. A healthy and robust vegetative growth, with proper orientation and size of leaves, are essential pre-requisites for high yields and nitrogen is mainly responsible for such a vegetative frame. This was clearly emphasized as early as 1937 by Tanaka, in a sand culture experiment. Through monitoring the use of carbohydrates, he observed that absence

of nitrogen resulted in poor growth and that nitrogen was largely responsible for controlling growth and fruiting . Significant increase in growth and yield due to nitrogen application was also observed by Croucher and Mitchell, (1940).

Bhan and Majumdar (1956) observed that application of 220 g of nitrogen per plant of banana(cv. Martamon) resulted in early growth and maturity, and higher number of hands, number of fingers per bunch and yield. Application of nitrogen promoted vegetative growth, increased the length of bunch and number of hands, resulting in increased yields (Steinhausen, 1957).

Champion *et al.* (1958) observed an increase of 18-25 tonnes/ha in Robusta banana by application of nitrogen at the rate of 100-200 kg/ha. Deficiency of nitrogen resulted in stunted growth, reduction in the rate of leaf production, size of leaf and suckering (Murray, 1959). Butler (1960) reported that nitrogen increased banana production by 10 to 12 per cent besides reducing the period of maturity. Lack of nitrogen caused reduction in leaf number with a long interval between the emergence of two consecutive leaves. The size of individual bunch and fruits were largely determined by the amount of nitrogen application several months before the emergence of inflorescence (Ochse *et al.*, 1961). Katyal and Chadha (1961) reported that nitrogen had a positive influence on periodicity of the plant, induced the growth of leaves and increased the number of fruits per bunch.

Battikhah and Khalidy (1962) reported that with increase in nitrogen application, there was a corresponding increase in plant height, leaf production, leaf area and leaf nitrogen content, but application beyond 200 g N per plant did not substantially

increase plant growth. Higher amount of nitrogen levels, improved the quality and shortened the period of maturity, but failed to increase yields (Jagirdar *et al.*, 1963).

A total deficiency of nitrogen would seriously impair the growth beyond flowering stage. If differentiation concided with a period of nitrogen deficiency, considerable reduction in yield and quality occurs (Charpentier and Martin-Prevel, 1965). Venkatesam *et al.* (1965) reported that nitrogen induced better growth and improved effective leaf area. Martin-Prevel (1966) observed that excess or deficiency of nitrogen had marked effects on fruit composition. He was of the opinion that earliness of the crop (Dwarf Cavendish) was due to N supply. Martin-Prevel and Montagut (1966) outlined the importance of nitrogen in banana plant growth and indicated a two-way relationship between uptake and dry matter production. Since no nitrogen storage occurred within the plant, additional nitrogen promoted growth. The uptake of nitrogen was directly correlated with increase in dry matter production irrespective of varieties, soils and environmental conditions. Simmonds (1966) reported, hastening of shooting and increased bunch weight due to application of nitrogen. Leigh (1969) stated that application of 227 kg nitrogen per hectare was the optimum dosage for banana, when applied in 3-4 splits. The application of nitrogen hastened maturity and increased yields (Singh, 1969). Application of judicious amount of nitrogen, resulted in improved grades, earliness, bunch weight and yield (Champion, 1970; Melin, 1970).

Arunachalam (1972) reported the early maturity effect of nitrogen on dwarf Cavendish, Giant Cavendish, Robusta and Lacaton banana with reference to plant height, girth, leaf area, number of leaves, cropping period and yield. The sucker production was also enhanced in all the varieties with application of 170 g nitrogen per

plant with a reduction in cropping period besides better fruit development and its chemical composition. Shanmugam and Velayutham (1972) reported that banana plants supplied with adequate nitrogen produced 17 leaves , while seven leaves were produced in plants which did not receive nitrogen at all. However, Ramanathan *et al.*(1973) found that Nitrogen applied in any form did not affect yields.

Ramaswamy and Muthukrishnan (1974) reported that application of nitroger at 170 g/plant resulted in better response in terms of number of hands and fruits, weight of hands and fruits. Application of 300 g/plant/year resulted in higher bunch weight and more number of hands and fingers per bunch (Monica *et al.*, 1978). Singh *et al.* (1979) reported that 150 g nitrogen per plant gave higher yields and better fruit quality in Basrai banana.

Chattopadhyay *et al.* (1980) reported that with increasing nitrogen upto 240 g/plant/year increased the yield in cv. Giant Governor banana upto 31.2 and 30.88 t/ha. for the main and ratoon crops respectively. Irizarry *et al.* (1981) recommended an application of 338 kg N per ha for obtaining optimum yields in the cv. Maricongo, while Kohli *et al.* (1981) reported that an application of 180 g N/plant gave the highest yield (44 t/ha) in Robusta banana. Application of 150 g nitrogen/plant has resulted in higher yields of Robusta banana (Mustaffa, 1983).

Anjorin and Obligbesan (1984) noticed that the beneficial effect of nitrogen on early growth and development was evident only when moisture was not a limiting factor. Application of nitrogen upto 300 g/plant significantly increased plant height, pseudostem girth, pseudostem weight and leaf weight, while higher nitrogen rates (400 g/plant) depressed all these parameters.

Kohli *et al.* (1984) reported that flowering was considerably delayed in Robusta banana, when no nitrogen was given, while application of 150-300 g nitrogen plant resulted in higher dry matter production and higher yields. Kohli *et al.* (1985) reported that an application of 150 g N/plant gave the highest yield in Robusta banana. Plants receiving no nitrogen took more number of days to reach flowering (378 days), While nitrogen fertilized plants took 315-329 days to flower.

Sharma (1985) in a trial with cv. Basrai found that the plants that received nitrogen at the rate of 187.5 g/plant applied to the soil in June-July plus 187.5 g/plant applied in 12 sprays at two week intervals starting from October/November gave the greatest plant height, pseudostem girth, number of fruits per bunch and bunch weight. Obiefuna and Onycle (1987) reported that yearly application of 200 g N/plant resulted in production of heaviest bunches. Application of 225-425 g N/plant in the banana cv. Gaint Cavendish increased plant height, girth and yield components (Oubahou and Dafiri, 1987).

Hazarika and Mohan (1991) reported that application of 160 g nitrogen gave the highest yield, number of fingers per bunch and bunch weight, when applied in 3 split doses. Prabhuram (1992) stated that the application of 200 g nitrogen in the form of urea resulted in increased bunch weight, number of fingers per hand and hands per bunch in cv. Rasthali. Higher average bunch weight and fruit yield per hectare were obtained with 200 g nitrogen applied in four splits in cv. Robusta (Reddy, 1992). Singh and Kashyap (1992) reported that application of 400 g N/plant in cv. Robusta gave the highest yield (69.32 t/ha), number of hands/bunch and number of

fingers/bunch. Highest growth rate of stem and fingers were obtained with application of 320 g N/plant in banana cv. Basrai (Kohli *et al.* 1984).

2.2.2 Role of potash on growth, yield and quality of banana

Potassium is a key element in banana nutrition. It plays a vital role in the physiological and biochemical functions of the plant. Way back in 1807, Fourcroy and Vauquelin (in Lacoeuilhe, 1973), while analyzing a banana plant, found a high concentration of potassium in the plant sap. Since then, potash was considered as a major nutrient in banana cultivation, and many research workers have shown that adequate supply of potash fertilizer not only increased the yield of banana but also improved quality and induced tolerance to biotic and abiotic stresses.

Banana requires potassium in large quantities throughout its normal life upto flowering, i.e., during the potential growth of the plant and again during fruit filling stage. The need for potash is substantial (Lacoeuilhe, 1973).

The most universal potassium deficiency is appearance of orange-yellow chlorosis of the older leaves and their subsequent rapid death with significant reduction in the life span of the leaf (Murray, 1960; Lahav, 1972). Potassium deficiency causes choking, reduced leaf size, delay in flower initiation, reduced fruit number/bunch, hand number/bunch and fruit size. (Martin-Prevel and Charpentier, 1966; Murray 1959; Lahav, 1972).

The study conducted by Wood (1939) indicated that application of farm yard manure and potash resulted in increased yield. The finger length and circumference, number of fingers, fruit weight, bunch weight and total number of bunches/ha were

very high. Potassium deficiency impaired protein synthesis, since free aminoacids and soluble forms of nitrogen increased in low potassium plants. The fruit growth was restricted by low potash supply in two ways, the translocation of carbon compounds from the leaves to the fruit was reduced and even when sugars reached the fruit, their conversion to starch was restricted, which ultimately lead to production of thin and fragile bunches (Martin-Prevel, 1973). In Taiwan a study conducted by Chu (1960; 1961) found that the application of potash significantly increased pseudostem growth, fruit yield, quality and storage as well as disease resistance. Potassium application to banana plants helped in increased bunch weight and also robust growth of plants. Potassium application also increased resistance power of plants to diseases (Katyal and Chadha, 1961).

Hewitt and Osborne (1962) observed that the application of 204 kg K_2O/acre/annum in 3 to 4 split doses increased bunch and finger length and weight, rind thickness and circumference. It was observed that potash application also markedly improved the keeping quality of fruits when examined after 20 days of storage.

The total weight of fruit/acre as well as average bunch weight in potassium treated plants have shown appreciable increase (Osborne and Hewitt, 1963). According to Ho (1968) potash application at the early stages recorded maximum height, girth, number of leaves, leaf area and increased sucker growth. The study also revealed a close relationship between pseudostem height and girth with yield and this was confirmed by him later (Ho, 1969).

Turner and Bull (1971) suggested the application of potash at 1500 lb/ac for reducing leaf deficiency symptoms. Higher rates of potash (360 g/plant) significantly increased the pseudostem height, girth, leaf area and sucker production and resulted in early flowering and maturity with good graded quality bunches of Robusta banana. Reduction in bunch size, in finger length and circumference were observed due to low potassium content (Lahav, 1972).

Low potassium supply resulted in reduced respiration but produced large variation in photosynthesis (Martin-Prevel, 1973). The total dry matter was the balance between gross photosynthesis and respiration. If respiration was lower in potash deficient plants then the main effect on dry matter production would be through reduction of photosynthesis. Turner (1979), reported that low supply of potash restricted the transfer of N, P, Ca, Mg, Mn, Cu, and Zn across the xylem. Pillai *et al.* (1977) reported that the higher yields were obtained by application of 114 g/K_2O/per plant of nendran.

In Cuba, application of 750 g K_2O/plant produced highest yield with an average bunch weight of 38.7 kg and 164 bananas/bunch (Garcia *et al.* 1979). Inadequate potassium supply reduced the total dry matter production of banana plants and distribution of dry matter within the plant. The organ most drastically affected is the bunch, which signifies the importance of potassium in banana production. Turner and Barkus (1981) found that while low potassium supply halved the total dry matter produced, the bunch size was reduced by 80 per cent. Low potassium supply reduced specific root weight and relative growth rate (Turner and Barkus, 1981).

In Costa Rica, Garita and Jaramillo (1984) reported that the application of 750 kg K_2O/hectare gave the highest bunch weight and yield (66424 kg/ha/year). Obiefuna (1984) concluded that the application of 300 g of K_2O/plant at 19th/20th leaf stage resulted in increased bunch weight (73.9%), number of fingers (33.7%) and finger weight (44.2%) over control. Application of potash at the rate of 250 g/plant resulted in increased plant height , girth of the pseudostem, number of leaves per plant and fruit yield (35.3 t/ha), total sugars, reducing sugars and non-reducing sugars of the fruit, while the titrable acidity was lower (Baruah and Mohan, 1985).

Mustaffa (1987; 1988) observed that application of muriate of potash at the rate of 400 g/plant significantly increased the height and girth of pseudostem, number of leaves and leaf area in Robusta banana under rainfed conditions. Yadav *et al.*(1988) concluded that optimum doses of potash for Dwarf Cavendish was 200 g/plant for maximum fruit yield. Baruah and Mohan (1991) recorded the highest phyllochron (7.3) and number of leaves (30.3), when 344g K/plant was applied at 3rd and 5th month after planting. EL-Khoreiby and Salem (1991) observed that highest potash application of 500 g/plant resulted in a positive response in height, basal circumference, growth and leaf production of Dwarf Cavendish.

Delayed floral initiation, reduction in leaf size, fruit number, hands/bunch and size of the fruit were observed in plants that received no potash compared to the plants given 250 g potash which produced highest fruit yield of 35.3 t/ha (Baruah and Mohan, 1992). Ray *et al.* (1993) concluded that application of potash at the rate of 300 g/plant/year was optimum to produce higher yield of 74.9 t/ha in main crop and 76.4 and 70.9 t/ha in I and II ratoon crops respectively. Application of potassium to

banana had direct effect on fruit quality. With increase in potash levels, the total soluble solids, reducing, non-reducing and total sugars, ascorbic acid and sugar acid ratio of the fruits increased with decrease in acidity (Croucher and Mitchell, 1940; Singh *et al.*, 1973; Jambulingam et al., 1975; Lahav and Turner, 1983).

Baruah and Mohan (1986) conducted an experiment with different levels of potash upto 300 g/plant and they observed that TSS, total sugars, reducing, non/reducing sugars in the fruit increased with increasing levels of potassium, while titrable acidity was low. The TSS was increased from 15.9 to 17 per cent with increasing potassium levels from 0 to 240 g/plant in Cavendish banana (Chattopadhyay and Bose, 1986). Ram and Prasad (1988) observed an increase in TSS upto 21.12% when potash was applied at the rate of 200 g/plant in banana cv.Campiergang local.

2.2.3 Role of NPK on growth, yield and quality of banana

There was a greater response to nutrients when applied together than separately. Increase or decrease of one nutrient element may substantially increase or decrease the other nutrient uptake. Antagonism and synergism among the nutrient elements are reported to have affected growth and development of banana. Norris and Ayyar (1942) reported that banana plants require large quantity of potash, moderate quantity of nitrogen and relatively little amount of phosphorus for optimum production. Summerville (1944) observed considerable response to nitrogen when applied in combination with potassium but the effect of phosphorus was not very pronounced.

Bhan and Majumdar (1956) reported poor growth response of banana cv. Martaman to nitrogen, when applied at low level along with heavy application of

phosphorus with or without potash. A study conducted in Trinidad for a population of 1500 plants by Monsterin (1956) recommended application of 50:80:250 kg NPK/ha. Champion *et al.* (1958) observed that in Robusta banana, nitrogen and potash when applied at the rate of 160 and 240 g respectively per plant gave an yield of 35.20 t/ha. While other treatments did not show such effect. Fertilizer experiments conducted in Jamaica over 13 years period revealed that economic response to fertilization can be expected from combined use of nitrogen, phosphorous and potash (Butler 1960).

Bhangoo *et al.* (1962) reported from Honduras that, application of 350-160-180 lb/ha of nitrogen, phosphorus and potash greatly improved the yield, average bunch weight, average number of hands/bunch and marketable quality of bunches. Funaioli (1962) applied NPK at 200:100:100 kg/ha to Dwarf cavendish banana and found that yield responses were higher with nitrogen alone as compared with combined application of nitrogen, phosphorous and potash. Lin *et al.* (1962) reported that nitrogen, phosphorous and potash applied particularly in combination increased the yield significantly at a dosage of 200:100:300 kg/ha. However, phosphorous had less effect compared to nitrogen and potash.

Martin-Prevel (1964) in Guinea, recommended 250:60:1000 kg N P K/ha for better yields. While Champion (1970) recommended a dosage of 560:224:672 kg NPK/ha in Puerto Rico for higher yields. The fertilizer recommendation in different states of India was reviewed by Shanmugam and Velayutham (1972).

The study conducted by Singh (1972) on the effect of nitrogen and potash on Robusta banana revealed that the length of fruit was more under nitrogen in presence of potash. The effect of nitrogen in the presence of phosphorous and potash increased

the number of hands, fruits and weight of the bunch significantly. The reducing sugar content was higher in treatments with K combination. A fertilizer mixture composing of 900 N, 480 P_2O_5 and 480 K_2O kg/ha enhanced vegetative growth which resulted in higher yield in Jahajee variety of Dwarf cavendish group (Sharma and Roy, 1973).

Arunachalam *et al.* (1976) stated that the application of 170g N/plant with P_2O_5 and K_2O at 85 and 340 g respectively in 2 split doses on 3rd, 5th months after planting for Cavendish clones resulted in higher yield. In a five year trial with Robusta banana the best results were obtained with nitrogen at 180 g/plant in 5 splits, P_2O_5 at 15.5g/plant in 3 splits and K_2O at 186.75g/plant in 5 splits/year (Kohli *et al.*, 1976).

Pillai *et al.* (1977) concluded that nitrogen and potash significantly increased fruit number and bunch weight when applied at the rate of 191 g and 301 g/plant respectively. Randhawa and Iyer (1978) reported that an application of 180:100:225 g NPK/plant in combination gave highest yield of 45 tonnes/ha in banana cultivar Robusta. Nambiar *et al.* (1979) reported that a combination of 225:250:450 g NPK/plant gave the greatest bunch weight in banana cv. Nendran.

Nanjan *et al.* (1980) stated that 100 kg of nitrogen in combination with 40 kg P_2O_5 and 400 kg K_2O/ac produced heaviest bunches in Robusta banana. Chundawat *et al.* (1983) reported from South Gujarath that the yield of Basrai banana increased by the application of NPK fertilizer at the rate of 180:180:180 g respectively in three split doses within six months of planting. Turner (1985) recommended a fertilizer dose of 200 to 250 kg N, 50 kg of P_2O_5 and 400 kg of K_2O/ha/year to produce 50 tonnes of fruits/ha. The application of 200g N, 80g P_2O_5 and 200g K_2O/plant resulted in highest TSS content (21.2%) in banana cv. Campeirgani local (Ram and Prasad, 1988).

Upadhyay (1989) found that application of higher rate of phosphorous and potash increased the yield and fruit quality. Pandit *et al.* (1992) reported that the highest yield (35 t/ha) and number of hands/bunch were obtained by applying 400g of Ammonium sulphate, 300g of Super phosphate, 250g of Muriate of potash/plant. The treatment also produced fruits with highest total sugar content with the lowest acidity. Application of 300 g each of N and K_2O was most effective in increasing the size, weight of bunch and fingers, number of hands and fingers per bunch in banana cv. Harichal (Pathak *et al.* 1992). Optimum requirement level of nitrogen, phosphorous and potash varied with varieties; Poovan required 160 g/plant, where as Rasthali and Nendran 210 g/plant (Selvaraj and Azhakiamanavalan, 1992).

The study conducted by Natesh *et al.* (1993) in banana cv. Nendran revealed that increased yields were obtained with the application of 190 g N, 115 g P_2O_5, 300 g K_2O/plant/year (in four split doses at 2nd, 4th, 6th and 8th months after planting). The application of 200:100:300 g NPK/plant resulted in high fruit yield (74.9 t/ha) in cv. Basrai banana, (Ray *et al.*, 1993). Parida *et al.* (1994) reported that combined application of NPK resulted in increased plant height, stem girth and number of leaves/plant and significantly reduced the time to shoot in cv. Robusta.

2.2.4 The role of split application of nutrients on growth and yield of banana.

Nitrogen is subjected to high losses when applied to the soil through the process of nitrification and leaching. In order to ensure sustained availability of nitrogen throughout the growth period and to minimize wastage nitrogen should be applied in

split doses at shorter intervals. Generally, early application before shooting decides the future bunch size and number of fingers and the total yield. Regarding frequency of application of potash and phosphorus, Croucher and Mitchell (1940) considered that twice yearly was often enough, but there is a tendency to split the dose of these fertilizers further.

The study conducted by Summerville (1944) and Alexandrovicze (1955) reported that the time of application of fertilizer was obviously important and for better results it should be applied during early stages of the plant growth. Twyford (1967) reported that in Robusta, potash supply in the first two months had a greater influence on the number of hands produced while potash applied after shooting did not influence the finger size. Earlier application of potash was most beneficial for plant growth and fruit yield (Ho, 1968). Leigh (1969) recommended the dressing of N, P_2O_5 and K_2O in three to four split application during the year. In Puerto Rico, fertilizers were applied in these split doses as stated by Champion (1970). It has been reported from Mozambique that at least 200 kg N, 50-150 kg P_2O_5 and 100-600 kg K_2O/ha should be applied in three to four splits (Marques and Monteiro, 1971).

Shanmugam and Velayutham (1972) stated that potash could be applied in three split doses viz., first, third and fifth month after planting along with nitrogen. Veeraraghavan (1972) working with manurial-cum-liming experiment on Nendran followed two equal split applications in 2nd and 4th month after planting. Lahav (1973) followed four splits per year in the northern coastal plain of Israel.

In Assam, three split applications of fertilizers were given to Dwarf Cavendish (Jahajee) (Sharma and Roy, 1973). Ramaswamy and Muthukrishnan (1974)

recommended two split application of fertilizers in the third and fifth months after planting in Robusta. Jaramillo and Bazan (1976) applied nitrogen at intervals of 4, 6, 8, 10, 12 and 14 weeks for the cv.Gaint Cavendish and obtained highest yields with 47 kg N/ha applied once in three months. Arunachalam *et al.* (1976) stated that it was advisable to give 170 g of N with 85 g P_2O_5 and 340 g K_2O/plant in two split doses on third and fifth month after planting for Cavendish clones. In a five year trial with Robusta banana best results were obtained with nitrogen at 180 g/plant in five splits, phosphorous at 15.50 g/plant in three splits and potash at 186.75 g/plant in five splits/year (Kohli *et al.* 1976). Pillai *et al.* (1977) followed two equal split applications of NPK, at 70-80 days and 110-120 days after planting in a manurial study with Nendran.

Gopimony *et al.* (1980) reported that top dressing of banana cv. Zanzibar (AAB) with urea, in five weekly applications each at 100 g/plant during fifth or sixth month after planting increased bunch weight, hand numbers and finger numbers per bunch. Pillai and Khader (1980) adopted three split applications during 50, 80 and 120 days after planting in Robusta. Kohli *et al.* (1985) followed four split applications of nitrogen with P_2O_5 and K_2O. Nair *et al.* (1990) reported that the application of nitrogen and potash at 400 g and 600 g/Plant in six split doses at monthly intervals gave the highest yield in Nendran banana. Rajeevan and Mohan Kumaran (1992) conducted a trial to study the influence of split application of nutrients in banana cv. Mysore with a fertilizer dose of 200:200:440 g NPK/plant/year applied in different splits in the second, fourth, sixth and eight months of planting. Application of 3/4th of the dose in the second month and 1/4th of the dose in the six month, improved the

yield upto 17.76 per cent over control where the above dose was applied in two equal splits, in the second and fourth month of planting.

Natesh *et al.* (1993) concluded that the maximum bunch weight, hand weight, finger weight, pulp weight, TSS, total sugars and sugar-acid ratio were obtained when 190:115:300 g NPK/plant/year was applied in four splits (2nd, 4th, 6th and 8th months after planting) as compared with the same rates applied in two splits.

2.2.5 Role of nutrients on their uptake and distribution.

Estimating the removal of nutrients by the plants is an important aspect to compute the requirements of nutrients to be added to the soil for sustaining higher yields. The studies on nutrient requirements, distribution and uptake have been reported by many workers in banana (Baillon *et al.*, 1933; Norris and Ayyar 1942; Hewitt, 1955). Baillon *et al.*(1933) estimated that approximately 116:35:772 g of NPK were removed from the soil by Cavendish banana and hence recommended application of high doses of nitrogen in Canary Islands. Norris and Ayyar (1942) analysed the composition of different parts of the banana plants and worked on the mineral uptake of banana and found that potash and calcium were the chief substances and phosphorous was utilized in only a small quantity. Hewitt (1955) studied leaf sampling technique in Lacatan banana. He reported that the third leaf in the succession of leaves from the top at the time of shooting was the best for leaf analysis to determine the nutrient status of the plant. The critical level of leaf nutrients as reported by him was 2.60 (range 2.3 to 3.8), 0.45 (range 0.53 to 0.81) and 3.30 (range 2.0 to 5.3) per cent NPK on dry weight basis respectively. Besides, he reported that application of nitrogen promoted leaf

nitrogen and decreased leaf potassium. However, the leaf phosphorous was not altered by the application of fertilizers containing P_2O_5. He further stated that increased yields could be correlated with increased nitrogen content in the leaf.

Simmonds (1959) reported a range of 0.3 to 2.2% N, 0.06 to 0.5% P and 2.7 to 6.5% K in banana leaves on dry weight basis. Jacob and von Vexkull (1960) recorded that the nutrient removal for a 30 ton crop was 50 to 75 kg N, 15 to 20 kg P_2O_5 and 175 to 220 kg K_2O. Rishbeth (1960) observed that the severity of panama disease symptoms was related to potash uptake and he suggested that improvement in potash uptake by mature plants might significantly induce wilt resistance. Hewitt and Osborne (1962) observed that for securing higher yields the leaf tissues should have 2.6, 0.4 and 4.0 per cent of NPK respectively. They further indicated that the time of application of fertilizers had no effect on the nutrient composition of the leaf tissues. Increased rate of potash application resulted in decreased leaf nitrogen level.

Battikhah and Khalidy (1962) observed that plant height, leaf production rate and leaf nitrogen content increased with nitrogen application. Third leaf contained the maximum nitrogen while phosphorus and potash levels in the leaf decreased with age. Bhangoo *et al.* (1962) recorded increased nitrogen content of banana leaf tissue with increasing application of nitrogen by 17.2, 17.6 and 21.1 per cent over control from 200, 350 and 650 lb nitrogen/acre respectively. However the nitrogen content of the leaf tissue from the highest yielding treatment 350:160:180 lb NPK was only 9.4 per cent higher than the control.

Martin-Prevel (1962) reported that apparently 100 per cent of applied potash and at the most 40 per cent of the applied nitrogen are utilized. The K:N ratio was

normally 3 in the bunch and increased from 3.6 in the whole plant at the fifth leaf stage to 6.4 at harvest. Martin-Prevel (1964) reported the nutrient uptake by Dwarf cavendish banana as 50:12.5:150 NPK kg/ha for the production of 25 tons of fruits.

Twyford and Coulter (1965) reported that the adequacy level of NPK was found to be 2.9, 0.29 to 0.48 and 3.8 per cent respectively. The nitrogen and potash contents of leaf were more or less constant over a wide range of soils. Montagut and Martin-Prevel (1965) found a progressive increase in nutrient requirement from 75 days after planting till 285 days. The requirement of nitrogen was higher than phosphorous. They further reported that the total annual uptake from the soil was 250 kg N, 60kg P_2O_5, 1000kg K_2O, 200kg Ca and 100kg Mg/ha with a plant density of 2500. Twyford (1967) observed the gross uptake of potash was always the highest, being 3.73 times as high as nitrogen and that of phosphorus was the lowest. He also reported that the uptake of nutrients was slow upto 60 days and then increased rapidly till shooting.

Joseph (1971) recorded a total nutrient uptake of 33kg N, 8kg P_2O_5 , 285kg K_2O , 30kg Ca and 49kg Mg/ha in Malaysia. Ramaswamy (1971) reported the level of leaf nutrients as 3.29, 0.44 and 3.11 per cent N, P and K respectively, with the soil application of 425 kg N per hectare. Arunachalam (1972) reported that in general, the N, P, K, Ca and Mg content of leaves recorded at bull stage, in N treatment (170 g plant-1) ranged from 3.18 to 3.43, 0.462 to 0.545, 5.36 to 3.76, 2.3 to 2.4 and 0.25 to 0.28 per cent respectively.

Randhawa *et al.* (1973) found that potash content of leaf tissues increased with potash levels in Robusta and was 4.88 to 5.71 per cent with 675 g K_2O /plant. The optimum level of nitrogen was 206.1 g/plant resulting in 3.5 percent dry matter and an

yield of 44.4 t/ha. Ramaswamy and Muthukrishnan (1974) reported that the nitrogen content in leaf of Robusta banana was found to reach a maximum of 3.29 per cent after 7 months when nitrogen was applied at the rate of 170 g/plant. At this level, inflorescence emergence was accelerated and the period from planting to harvest was found to be shortened by 29 days. The concentration of NPK in different parts of Basrai banana leaf during May to October was studied by Shawkey *et al.*(1974). Maximum nitrogen and phosphorous content was in lamina, but potash content was maximum in the midrib while in petiole it was twice as high as in fruits.

In an investigation on Robusta, Jambulingam *et al.* (1975) concluded that leaf potash should be above 4.30 per cent for optimum production. Increase in potassium levels in leaves after soil application of 360 g K_2O/plant was found to be highly significant. Veeraraghavan (1972) reported that an average crop of banana removes 300 kg N, 80 kg P_2O_5 and 800 kg K_2O/ha. Bhavanisanker (1980) reported a critical leaf concentration in Nendran banana as 2.26 to 3.39 per cent nitrogen, 0.23 to 0.43 per cent phosphorous, 3.18 to 3.47 per cent potash, 0.94 to 1.35 per cent Calcium and 0.43 to 0.80 per cent Magnesium at shooting stage.

In a fertilizer trial with Dwarf Cavendish banana Chattopadhyay *et al.*(1980) studied the concentration of phosphorous and potassium in the third leaf at vegetative (180 days after planting) flowering and harvesting stages. They observed a gradual decline in the concentration of all the three nutrients as the plants advanced in age. At a level of 120:45:240 g NPK/plant, they found nitrogen content to decrease from 2.56 to 1.10 per cent during this period where as, the level of phosphorous and potash declined from 0.60 to 0.29 and 3.38 to 2.82 per cent, respectively. Irizarry *et al.*

(1981) observed that during the crop cycle, the nutrient contents of roots, corms, pseudostems and leaves increased upto harvest and they suggested application of 338:58:780:100 kg NPK & Mg/ha for the main crop, and 114:35.5:353:43 kg NPK & Mg/ha for the ratoon crop to obtain optimum yield.

Kohli *et al.* (1981) worked out the adequacy levels of nitrogen, phosphorous and potassium in leaf tissues in relation to the optimum yield as 2.85, 0.20 and 4.69 per cent NPK, respectively. Mathew and Aravindakshan (1981) recorded a steady increase in the uptake of nitrogen upto shooting which declined at harvest. Application of nitrogen was found to enhance the uptake of phosphorous, while uptake of potash increased with the stages of growth upto shooting, but did not show any corresponding increase with applied nitrogen. The uptake of nitrogen and phosphorous was found maximum in lamina during the vegetative phase and in fruits during fruiting phase, where as maximum potash uptake was noticed in the pseudostem both in vegetative and reproductive phases. Turner (1985) also reported that an increase in potash reduced the proportion of all nutrients (except K_2O) in the roots and increased the proportion in fruits. Large proportion of 20 to 36 per cent NPK were located in the fruits. Kotur and Mustaffa (1984) found that the critical level of leaf nitrogen concentration to be 3.51 per cent in Robusta banana.

Buragohain and Shanmugavelu (1986) conducted elaborate studies on the nutrient uptake of the banana `Vayal Vazhai' (ABB). The results revealed that there was a sharp increase in the uptake of nitrogen from 16.47 kg/ha at sucker stage to 310.82 kg/ha at the shooting stage, and thereafter, there was a gradual decline in the nitrogen uptake. The difference in nitrogen uptake between various stages was highly significant

and rose from 2.50 kg/ha at sucker stage to 60.68 kg/ha at the shooting stage and thereafter, it declined to 55.60 kg/ha at harvest. The uptake of potash was 28.29 kg/ha at sucker stage which, then increased tremendously up to 879.21 kg/ha at harvest.

Baruah and Mohan (1991) showed that increase in potash levels considerably increased the accumulation of nutrients in the leaf, while nitrogen concentration decreased. Dave *et al.* (1991) observed greater association of leaf contents of N, P, K, Mg, Fe, Mn and Zn with fruit yield. Hazarika and Mohan (1992) reported an increase in nitrogen content of the leaves with the increased levels of nitrogen application. The potassium content of the leaves increased with the increase in nitrogen content. Shawkey *et al.* (1993) reported that with increased nitrogen application rates there was an increase in leaf nitrogen content while leaf Calcium content decreased. They reported a leaf nitrogen content of 1.99 to 2.62 per cent, at standard nitrogen rate.

MATERIAL AND METHODS

III MATERIAL AND METHODS

The present investigation was conducted to standardize the protocol for micropropagation of banana cv. Grande Naine, and to determine the requirement of nitrogen and potassium for banana plants raised through tissue culture.

3.1 Micropropagation of Banana (cv. Grande Naine)

The present investigation was conducted in the Plant Tissue Culture Laboratory (UAS/CTD) at the Division of Horticulture, University of Agricultural sciences, G.K.V.K. Bangalore, during 1992-93.

3.1.1. Plant material

Healthy and vigorous banana plants (raised from sword suckers) of cv. Grande Naine (3-4 month age and in active growth phase), free from viruses and diseases, were selected as a source of explant (Plate-1). The selection of these plants was also based on the performance of the mother plants with respect to their yield potential.

3.1.2. Preparation of explants

The procedure described by Doreswamy *et al.* (1983) and Cronauer and Krikorian (1984a) was followed for explant preparation with certain modifications. The plant material obtained from the field was thoroughly washed in running tap water followed by washing with a detergent solution (Teepol 0.5%) to remove adhering soil particles.

by washing with a detergent solution (Teepol 0.5%) to remove adhering soil particles. Later, the rhizomes were kept immersed in a fungicide solution (Bavistin 1% w/v) for half an hour, to further cleanse the planting material. The outer leaves, leaf base and corm tissue were trimmed using a sterilized stainless steel knife until the length of the explants was 4-6 cms and the diameter, 3-4 cms. These trimmed suckers enclosing the shoot tip were washed with double distilled water. After trimming one more outer layer, they were soaked in a solution of Bavistin(0.5%) and streptocycline (0.05%) for eight hours. After thoroughly washing with double distilled water, they were trimmed again, so that the trimmed suckers were of 2-3 cm in length and 2-2.5 cm in diameter. These shoot tips were soaked in cetrimide solution for 30 minutes. After removing one more outer layer, the shoot tips were surface sterilized with chlorine water (prepared in TDW) in a closed container for 15-20 minutes. Further operations such as washing several times with sterile distilled water to remove all traces of chlorine and trimming the explant (about 2-3 mm size) were carried out under a laminar flow chamber. The different stages of preparation of explant are shown in Plate 2.

EXPERIMENT I

3.1.3 Effect of size of explant and culture media on *in vitro* establishment of shoot-tips

In this experiment three different sizes of shoot-tip explants were incubated in eleven liquid culture media for two weeks maintaining standard culture conditions (Doreswamy *et al.*, 1983),to study the effects on establishment.

Treatment details

1. **Explant size:**

   (a) 2-3 mm

   (b) 4-5 mm

   (c) 6-7 mm

2. **Culture media:**

Murashige and Skoog's (MS) liquid media (Appendix I) was supplemented with Benzylaminopurine (BAP) and adenine sulphate (Ads) as given below at different concentrations.

1. BAP 20 µm + Ads 100 µm
2. BAP 20 µm + Ads 200 µm
3. BAP 20 µm + Ads 300 µm
4. BAP 40 µm + Ads 100 µm
5. BAP 40 µm + Ads 200 µm
6. BAP 40 µm + Ads 300 µm
7. BAP 60 µm + Ads 100 µm
8. BAP 60 µm + Ads 200 µm
9. BAP 60 µm + Ads 300 µm
10. 10% coconut water
11. Control (only MS)

Replications: Six

Number of Explants/replication: Twelve

Design: Factorial CRD

Observations: After two weeks of incubation, all the explants were evaluated for their ability to establish in different liquid media. Greening of the explant and swelling were utilized as important criteria for assessing the success in establishment. Shoot tips that had turned dark brown/black and which did not swell were considered as non-established.

EXPERIMENT II

3.1.4 Effect of culture media on percentage establishment of explants on semi-solid media

Here, healthy and contaminant free explants (2-3 mm) were tried on the best six semi-solid media combinations which proved better in Experiment I. The explants were excised by removing discoloured tissue and transferred to the semi-solid media and incubated for four weeks maintaining standard culture conditions.

Treatment details:

1. **Culture media:**
    1. BAP 40 µm + Ads 100 µm
    2. BAP 40 µm + Ads 200 µm
    3. BAP 40 µm + Ads 300 µm
    4. BAP 60 µm + Ads 100 µm
    5. BAP 60 µm + Ads 200 µm
    6. BAP 60 µm + Ads 300 µm

Replications: Five

Number of explants/replication: Eight

Design: Factorial CRD

Observations: The explants were observed for their bulging in the tip and morphogenetic activity. Such explants were counted and expressed in terms of per cent.

EXPERIMENT III

3.1.5 Effect of culture media on multiple shoot induction

The successfully established explants from the previous experiments were used in this experiment. The explants were excised by trimming the discoloured tissues and

then 4-6 vertical cuts were made at the tip of each explant. They were cultured in the same media as it was in the previous experiments and incubated for eight weeks (the cultures were transferred to fresh media after 4 weeks) maintaining standard culture conditions.

Treatment details:

1. Culture media:

1. BAP 40 µm + Ads 100 µm
2. BAP 40 µm + Ads 200 µm
3. BAP 40 µm + Ads 300 µm
4. BAP 60 µm + Ads 100 µm
5. BAP 60 µm + Ads 200 µm
6. BAP 60 µm + Ads 300 µm

Replications: Five

Number of explants/replication: Five

Design: Factorial CRD

Observations:
1. Number of multiple shoots
2. Average length of multiple shoots
3. Average number of leaves

EXPERIMENT IV

3.1.6 Influence of auxins on *in vitro* production of roots

In this experiment the influence of three auxins at various concentrations in MS basal media, on the production of adventitious roots, was studied. The explants were incubated for four weeks maintaining standard culture conditions.

Treatment details:

1. IAA 0 4 8 12 16 µm
2. IBA 0 5 10 15 20 µm
3. NAA 0 5 10 15 20 µm

Replications: Five

Number of explants/replication: Five

Design: CRD

Observations: Number of roots produced/shoot

EXPERIMENT V

3.1.7 Influence of rooting media on establishment of *in vitro* plantlets

The *in vitro* rooted plantlets were removed from the culture media and washed with water. They were given a quick dip in 0.5% Bavistin solution and transferred to thumb pots containing media and placed in the green house, where the temperature ranged between 25 and 27°C and relative humidity between 80 to 90 per cent.

Treatment details:

1. Soilrite
2. Soil
3. Sand
4. Soilrite + Soil + Sand 1:1:1 v/v
5. Soilrite + Soil + Sand 2:1:1 v/v
6. Soilrite + Soil + Sand 2:1:2 v/v

Replications: Ten

Number of plantlets/replication: Ten

Design: CRD

Observations: Percentage survival of plantlets in greenhouse recorded after 45 days.

3.2 Nutritional studies on tissue cultured banana

Studies on requirement of nitrogen and potassium for tissue cultured banana cv. Grande Naine was conducted at Rashmi Farm, Channasandra village, Bangalore South Taluk. The experimental field is located at on altitude of 890 M above mean sea level (13° N; 77°38' E).

3.2.1 Soil characteristics

The physical and chemical properties of the soil are furnished here under.

a) **Physical properties**

1. Soil type : Red sandy loam
2. Coarse sand : 34.30%
3. Fine sand : 18.10%
4. Silt : 22.20%
5. Clay : 25.40%

b) **Chemical properties**

1. pH : 5.72
2. EC : 0.025 ds/m
3. Organic carbon : 0.51%
4. Available N : 177.75 kg/ha
5. Available P_2O_5 : 21.65 kg/ha
6. Available K_2O : 143.25 kg/ha

3.2.2 Experimental details

3.2.2.1 Design and layout

The experiment was laid out in a Randomised Block Design with 9 treatments in three replications. Under each treatment there were sixteen plants, provided with guard rows on all sides of the plot. Four plants from each replications were used for recording biometric observations till harvest.

3.2.2.2 Treatment details

Nitrogen and potash were applied at three levels, while phosphorus was kept constant at 100 g plant^{-1}.

T_1 N_1K_1 = 150 g N: 200 g K_2O/plant

T_2 N_1K_2 = 150 g N: 300 g K_20/plant

T_3 N_1K_3 = 150 g N: 400 g K_2O/plant

T_4 N_2K_1 = 225 g N: 200 g K_2O/plant

T_5 N_2K_2 = 225 g N: 300 g K_2O/plant

T_6 N_2K_3 = 225 g N: 400 g K_2O/plant

T_7 N_3K_1 = 300 g N: 200 g K_2O/plant

T_8 N_3K_2 = 300 g N: 300 g K_2O/plant

T_9 N_3K_3 = 300 g N: 400 g K_2O/plant

Nitrogen was applied in five split doses at the rate of 10%, 15%, 25%, 30% and 20% after 30th, 60th, 90th, 120th and 150th days after planting respectively.

Potash was also applied in five split doses at the rate of 10%, 15%, 20%, 25% and 30% after 30th, 60th, 90th, 120th and 150th days after planting respectively.

Phosphorus was kept constant for all the treatments and was applied at the rate of 100g per plant and applied with nitrogen and potash in five equal split doses on 30th, 60th, 90th, 120th and 150th days after planting.

Gross plot size : 10.8m x 10.8m.

Net plot size : 7.2m x 7.2m.

3.2.3 Field preparation and planting

The experimental site was thoroughly ploughed to a fine tilth, harrowed and levelled. Pits of $45cm^3$ size were dug at a spacing of 1.8 x 1.8m. The pits were filled with top soil and 40 g of furadan granules were applied to each pit prior to planting. Uniform sized Grande Naine tissue cultured plants (Plants cultured on the same day) of three months age were planted in the pits, provided with drip irrigation. Other recommended cultural practices were carried out regularly and uniformly for all the treatments.

3.2.4 Morphological characters

Field observations on growth characters of the four tagged plants and bunches under each treatment replicationwise were done following the methods of Yang and Rao (1962). Data on growth characters were recorded on 30th, 60th, 90th, 120th, 150th days after planting, at shooting and at harvest.

3.2.4.1 Growth of the plant

3.2.4.1.1 Plant height

The height of the plant was measured 30 cm above the ground level up to the angle between the youngest first and second leaf axils and finally this 30 cm was added and the total height was expressed in cm.

3.2.4.1.2 Pseudostem girth

The girth of the pseudostem was measured at 30 cm above the ground level and expressed in cm.

3.2.4.1.3 Number of functional leaves

The number of photosynthetically active leaves were counted and recorded.

3.2.4.1.4 Leaf area

The leaf area was calculated by multiplying leaf length and breadth with a constant 0.80 and expressed in m^2 (Hewitt, 1955).

3.2.4.1.5 Leaf Area Index (LAI)

The LAI of functional leaves was calculated by employing the formula suggested by Williams (1946).

$$LAI = \frac{\text{Leaf area per plant}}{\text{Land area occupied by the plant}}$$

Leaf area was calculated by adding the leaf area of the functional leaves at shooting.

3.2.4.1.6 Phyllachron

The number and date of emergence (only fully emerged leaves considered) of each leaf were recorded, and the phyllachron (the day interval between the emergence of successive leaf) was calculated.

3.2.4.2 Time taken for shooting and harvesting

The number of days required from planting to shooting, shooting to harvest (emergence to harvest interval) and planting to harvest (duration) were counted and recorded.

3.2.4.3 Dry matter production (DMP) and distribution

The dry matter production (DMP) and distribution in the different parts of the plant, was carried out five times during and crop growth period, once at the time of planting, 60 days after planting (DAP), 150 DAP, at shooting and at harvest.

For estimating the DMP, the whole plant was pulled out with little root damage. After recording the fresh weight of each part like Corms (including roots), Pseudostem, leaf, inflorescence, stalk and fruits, the representative samples from each part were taken and dried at 60°+/-1°c for 48 hours. Based on the moisture content, the DMP of different parts of the plant was compared and expressed as kg plant^{-1}. These samples were utilized for biochemical analysis of the plant.

3.2.5 Yield and yield components

3.2.5.1 Weight of the bunch

While harvesting the fully matured bunch, peduncle was cut, leaving 22.5 cm above the first hand and 5 cm below the last hand and the weight was expressed in kg.

3.2.5.2 Number of hands per bunch

Total number of hand per bunch was counted and expressed in number.

3.2.5.3 Number of fruits

The total number of fruits in the bunch was counted.

3.2.5.4 Average fruit weight

The middle finger in the top and bottom rows of the second hand were selected as representative fingers (Gottreich *et al.*, 1964) to record the average fruit weight.

3.2.5.5 Estimated yield

This was calculated by multiplying the yield plant^{-1} with the total number of plants hectare^{-1}.

3.2.5.6 Harvest Index

Harvest index was calculated by using the following formula and expressed in percentage.

$$HI = \frac{\text{Economic yield}}{\text{Biological yield}} \times 100$$

Economic yield indicates the bunch weight (dry) and the biological yield is the total biomass at harvest.

3.2.6 Fruit quality

The representative fingers were allowed for natural and uniform ripening (all yellow stage). These fruits were utilized for determining the following quality parameters.

3.2.6.1 Pulp/peel ratio

It was calculated by dividing the mean pulp weight by mean peel weight.

3.2.6.2 Total soluble solids. (T.S.S.)

T.S.S. was recorded with the extracted juice using a `Zeiss' hand refractometer at room temperature and expressed in percentage.

3.2.6.3 Reducing sugars

The reducing sugar content of the fruits was estimated following the method of Ranganna (1977).

3.2.6.4 Non-reducing sugars

The percentage of non-reducing sugars was obtained by subtracting the percentage of reducing sugars from the total sugars.

3.2.6.5 Total sugars

The total sugar content of the fruit was estimated following the method of Ranganna (1977).

3.2.6.6 Acidity

The standard method of A.O.A.C. (1960) was followed to estimate the acidity. The acidity was expressed as percentage of citric acid on fresh weight of sample.

3.2.6.7 Sugar/acid ratio

The sugar/acid ratio of the pulp was arrived at by dividing the value of total sugars by that of acidity and this was reckoned as a measure of fruit quality.

3.2.7 Analysis of plant

3.2.7.1 Nitrogen

Microkjeldahl method of Humphries (1956) was followed in estimating the total nitrogen content in different parts of plant sample, during different growth stages.

3.2.7.2. Phosphorus and potash

The phosphorus and potassium content of the different plant parts at various growth stages were determined as per the procedure of Jackson (1973).

3.2.8 Analysis of soil

3.2.8.1 Soil Sampling

A 'V' shaped cut was made to a depth of 15 cm at each sampling spot. About 1.5 cm thick slices of soil from the sides were removed and collected in a clean container. Samples were thoroughly mixed and the quantity was reduced by quartering.

3.2.8.2 pH

The pH of 1:2.5 soil water suspension was determined using a pH meter.

3.2.8.3 Electrical conductivity

The EC of 1:2 soil water suspension was measured in Elico Conductivity Bridge and expressed as ds/m.

3.2.8.4 Available nitrogen

The available nitrogen in the soil was estimated by alkaline $KMnO_4$ method and the values were expressed as kg ha^{-1} (Jackson, 1973).

3.2.8.5 Available phosphorus

The available phosphorus in the soil was estimated using ammonium molybdate method as outlined by Jackson (1973).

3.2.8.6 Available potassium

The available potash in the soil was estimated using Flame Photometer (Jackson, 1973).

3.2.9 Benefit-Cost ratio.

The benefit-cost ratio for the different treatments was worked out based on the expenditure and return in order to study the economics of banana production.

3.2.10 Statistical analysis.

The data recorded were statistically analysed following the procedure given by Sunder Raj *et al.* (1972). Correlation coefficients between yield attributing characters and yield were worked out.

EXPERIMENTAL RESULTS

IV EXPERIMENTAL RESULTS

The results of the studies on in vitro propagation of banana cv. Grande Naine and its nutritional requirement in the field, are presented below.

4.1 Micropropagation of Banana.

4.1.1 Effect of size of explant and culture media on initial *in vitro* establishment of shoot tips

The influence of liquid MS media supplemented with various combinations of BAP and Adenine sulphate concentrations, on the initial *in vitro* establishment of the three sizes of explants of banana shoot-tips are presented in the Table 1 & Fig. 1. There were significant differences among the treatments with respect to the per cent establishment.

In case of small sized explants (2-3 mm) the maximum per cent initial establishment (78.50) was observed in liquid MS media supplemented with BAP at 40 µM and adenine sulphate at 200 µM, and this was closely followed by BAP at 40 µM and adenine sulphate at 300 µM (71.25 %). The least percent initial establishment (28.12) was observed in control (liquid MS basal media).

The medium sized explants (4-5 mm) performed better in terms of initial establishment in liquid MS media supplemented with various concentrations of cytokinin combination, compared to the big size explants (6-7 mm). Both medium sized explants and big sized explants recorded maximum percent initial establishment

sized explants and big sized explants recorded maximum percent initial establishment (81.26 and 69.25, respectively) in liquid MS media supplemented with BAP at 40 µM and adenine sulphate at 200 µM. Although, the medium sized explants performed slightly better than small sized explants in terms of initial establishment, small sized explants were preferred in other experiments because lesser the size of the explant, lesser were the chances of diseases in the explant multiplication.

4.1.2 Effect of culture media on establishment (%) of explants on semi-solid media

In this experiment, the small sized explants initially established in liquid MS media, were cultured on semi-solid MS media supplemented with BAP at 20,40 and 60 µM and adenine sulphate at 200 and 300 µM (Table 2).

The per cent establishment of explants on MS semi-solid media supplemented with various concentrations of cytokinins differed significantly among the treatments. The semi-solid MS media supplemented with BAP at 40 µM and adenine sulphate at 200 µM recorded the maximum per cent establishment (89.50), closely followed by BAP at 40 µM and adenine sulphate at 300 µM (85.25). Lowest per cent establishment (63.60) was observed with BAP at 20 µM and adenine sulphate at 200 µM.

4.1.3 Effect of culture media on multiple shoot induction

The response of multiple shoot development to different concentrations of cytokinins in the culture media differed significantly among the treatments (Table 3 & Fig. 2). From the data it is clear that the maximum number of multiple shoot

Table 1 Influence of explant size and liquid culture media on *in vitro* establishment of banana shoot tips cv. Grande Naine

Treatments	Establishment of shoot tip culture (%) Explant size		
	2-3mm (small)	4-5mm (medium)	6-7mm (big)
BAP 20 µM + Ads 100 µM	51.70	54.00	43.22
BAP 20 µM + Ads 200 µM	60.42	61.37	49.40
BAP 20 µM + Ads 300 µM	62.55	60.25	50.50
BAP 40 µM + Ads 100 µM	57.00	59.50	54.62
BAP 40 µM + Ads 200 µM	78.50	81.26	69.25
BAP 40 µM + Ads 300 µM	71.25	70.00	65.12
BAP 60 µM + Ads 100 µM	53.45	51.00	50.50
BAP 60 µM + Ads 200 µM	69.12	72.75	63.75
BAP 60 µM + Ads 300 µM	66.62	65.00	56.26
Coconut Water (10%)	56.25	59.50	49.00
ControL (only MS media)	28.12	30.00	32.50
C.D.@ 5%	9.22	6.32	5.03

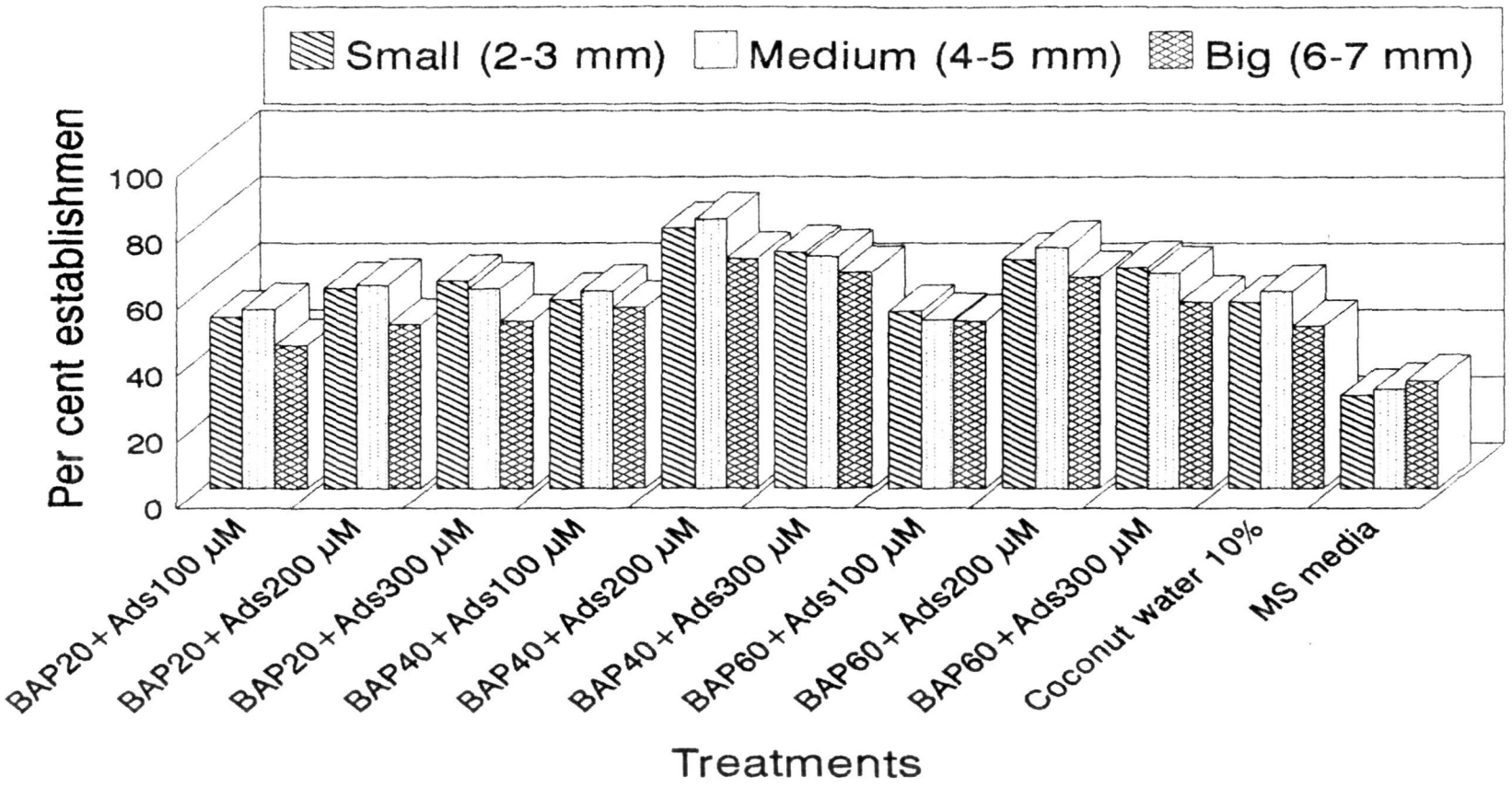


Fig. 1 Effect of explant size and MS liquid media on in vitro establishment of shoot-tips

development (18.25) and maximum average height of pseudostem (5.72) was observed in the culture media supplemented with BAP at 40 µM and adenine sulphate at 200 µM. The maximum number of leaves (4.20) were recorded when the culture media was supplemented with BAP at 40 µM and adenine sulphate at 300 µM. The least number of multiple shoots (6.16) and leaves (3.00) were recorded with BAP at 20 µM and adenine sulphate at 200 µM.

4.1.4. Influence of auxins on *in vitro* production of roots

In this experiment, the multiple shoots developed in the previous stage were subjected three different sources of auxins, at different concentrations in MS basal media to determine the best auxin and concentration for induction of adventitious roots (Table 4 & Fig. 3).

a) Indole acetic acid.

The data on the influence of IAA at different concentrations on the *in vitro* production of roots differed significantly among the treatments and the maximum number of roots (3.66) were produced when MS basal media was supplemented with IAA at 4 µM, followed by IAA at 12 µM. In case of control, where no auxin was supplemented, the average production of roots was 2.00.

b) 3-Indolebutyric acid

The influence of IBA on *in vitro* production of roots differed significantly between the treatments. MS basal media supplemented with IBA at 5 µM produced the highest number of roots (7.00) and this was followed by IBA at 10 µM (5.66). The lowest number of roots (2.66) was recorded in MS basal media without any auxins.

Table 2 Influence of cytokinins with MS media on *in vitro* establishment of explants

Treatments	% establishment
BAP 20 µM + Ads 200 µM	63.60
BAP 20 µM + Ads 300 µM	68.65
BAP 40 µM + Ads 200 µM	89.50
BAP 40 µM + Ads 300 µM	85.25
BAP 60 µM + Ads 200 µM	79.36
BAP 60 µM + Ads 300 µM	80.00
C.D. @ 5%	3.32

Table 3 Influence of cytokinins with MS media on *in vitro* development of multiple shoots

Treatments	No. of Multiple shoots	Av. height of Pseudostem	No. of leaves
BAP 20 µM + Ads 200 µM	6.16	3.10	3.00
BPP 20 µM + Ads 300 µM	7.00	3.00	3.00
BAP 40 µM + Ads 200 µM	18.25	5.72	3.90
BAP 40 µM + Ads 300 µM	16.10	5.16	4.22
BPP 60 µM + Ads 200 µM	10.50	4.26	3.10
BAP 60 µM + Ads 300 µM	12.00	4.56	3.00
C.D. @ 5%	4.03	1.06	0.27

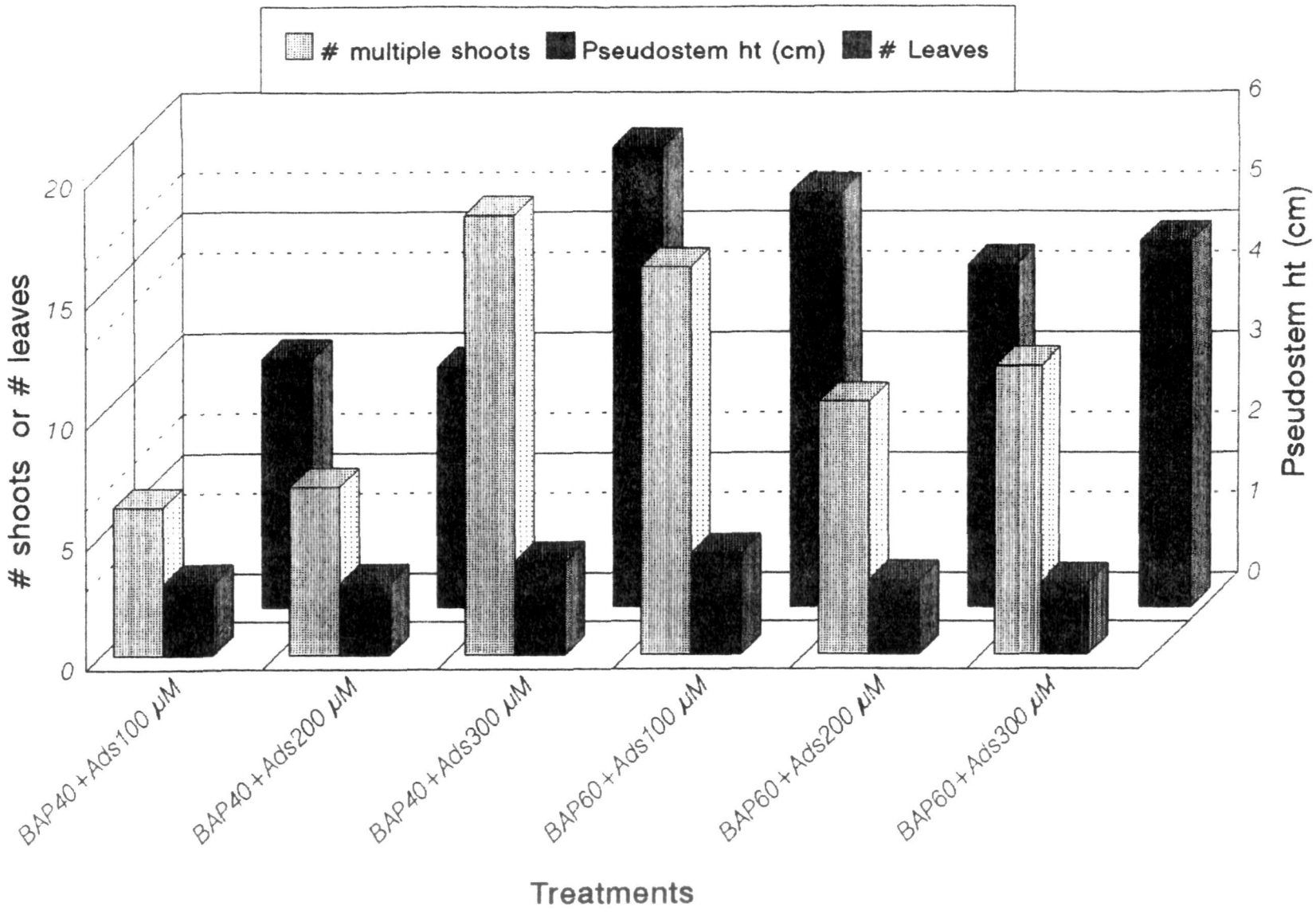


Fig 2 : Influence of cytokinins with MS media on *in vitro* development of multiple shoots

c) 1-Naphthalene acetic acid.

The mean number of roots produced due to various NAA treatments varied from 2.33 to 7.66 and differed significantly among the treatments. The least number of roots (2.33) was observed in MS basal media without auxins. The MS basal media supplemented with NAA at 5 µM produced the maximum number of roots (7.66), followed by NAA at 10 µM (6.00).

4.1.5 Influence of rooting media on establishment of *in vitro* plantlets in green house.

Three different rooting media, in various combinations, were used to harden the *in vitro* banana plantlets in the green house for 45 days. The effect of rooting media on per cent survival of banana plantlets in the green house differed significantly among the treatments (Table 5). The rooting media combination of perlite, soil and sand in the ratio of 2:1:2 had the highest survival per cent of plantlets (96.50). This was on par with other rooting media like perlite, sand and soil in the ratio of 1:1:1 (93.20), 2:1:1 (93.00) and perlite alone (90.40). Soil and sand used for hardening the plantlets in the green house were found to be moderate in per cent plantlet survival (73.80 and 70.00, respectively).

Table 4 Influence of auxins on *in vitro* production of roots

Auxin & Conc. (µM)	Average no. of roots
IAA	
0 (MS basal)	2.00
4	3.66
8	2.32
12	3.00
16	2.66
C.D.@ 5%	1.44
IBA	
0 (MS Basal)	2.66
5	7.00
10	5.66
15	5.00
20	5.33
C.D. @ 5 %	1.15
NAA	
0 (Ms Basal)	2.33
5	7.66
10	6.00
15	5.66
20	4.66
C.D. @ 5%	1.32

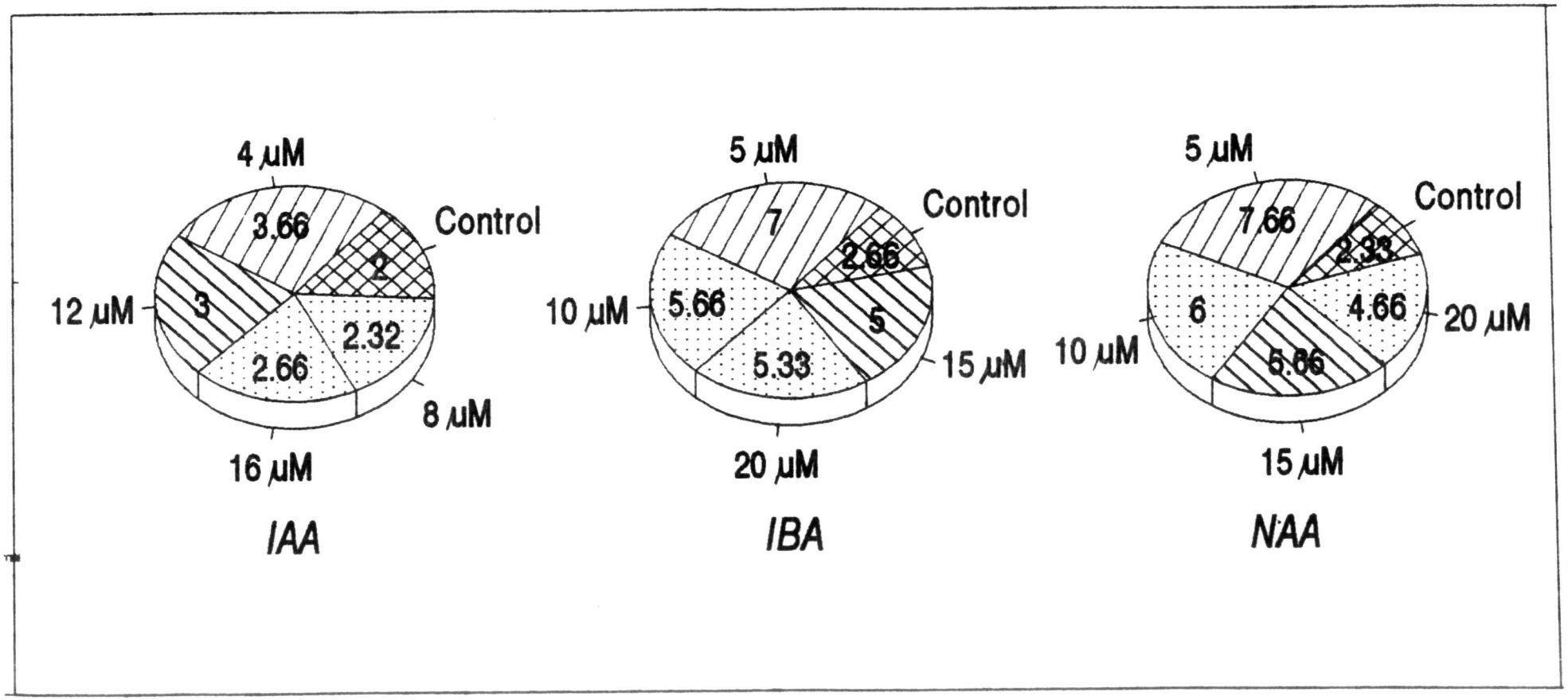


Fig 3 : Influence of auxins on *in vitro* development of roots

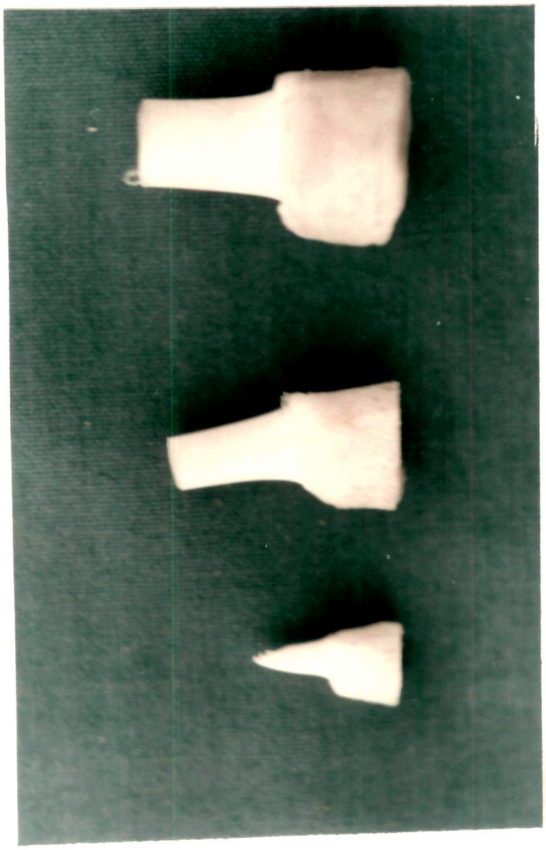
PLATE 1

A. Different sizes of banana explants used in the experiment

B. Development of multiple shoots in banana under *in vitro* conditions

C. Rooting of banana plantlets under *in vitro* conditions

D. Hardening of banana plantlets at various stages



A

B

C

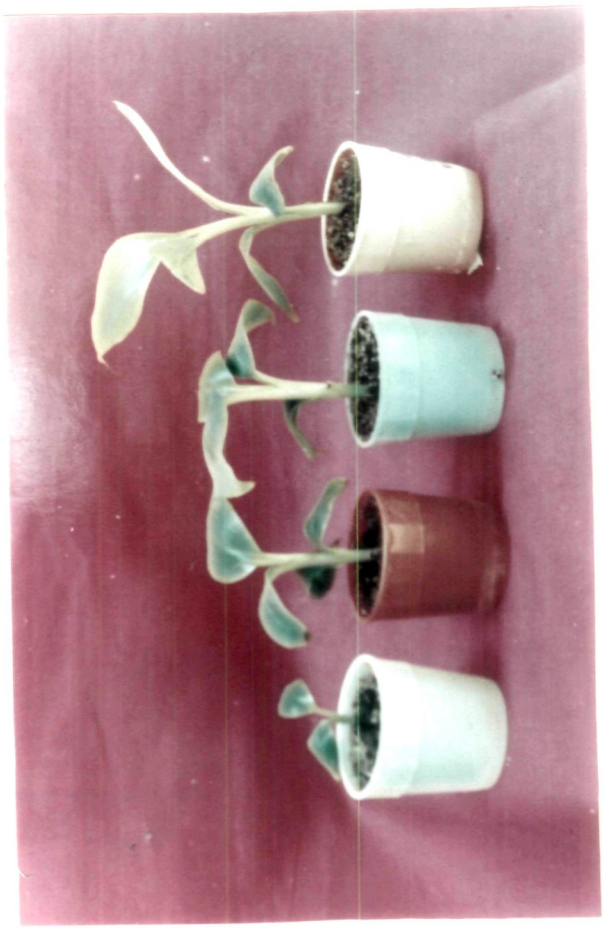


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Table 5 Influence on rooting media on per cent survival of banana plantlets produced *in vitro* (after 45 days)

Media	Survival of plantlets in Greenhouse (%)
Perlite	90.40
Soil	73.80
Sand	70.00
P:S:S (1:1:1)	93.20
P:S:S (2:1:1)	93.00
P:S:S (2:1:2)	96.50
CD @ 5%	7.62

4.2 Nutritional studies on tissue cultured banana

The results of the studies on the nutritional requirement of tissue cultured banana cv. Grande Naine are presented below.

4.2.1 Role of nutrients on growth parameters

4.2.1.1 Plant height

The data on the effect of nutrients on plant height recorded during different periods of crop growth are presented in Table 6, 7 & Fig. 4.

It was observed that the plant height did differ significantly due to increased application of nitrogen and potash during the crop growth from 60 DAP. Application of nitrogen and potash in combination at higher levels recorded higher mean plant height during the different stages of plant growth. At shooting the plant height was maximum in N_3K_3 (218.50 cm) which was on par with other treatments like N_3K_2 (215.50cm), N_3K_1 (214.80cm), N_2K_1 (212.90cm) and N_2K_3(210.40cm). The treatment combinations of nitrogen at 225 g and 300 g with all potash levels recorded better plant height compared to nitrogen at 150 g with any combination of potash levels.

It was observed that, there was an increasing trend in plant height from 60 to 150 DAP in all the treatments and the maximum growth was between 90 to 120 DAP.

4.2.1.2 Pseudostem girth

The pseudostem girth showed significant differences due to different nutrient levels in all the stages of crop growth (Table 8,9 & Fig. 5).

Table 6 **Effect of nitrogen and potash on plant height (cm)**

Treatments	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	At Shooting
N_1K_1	20.17	53.92^{cd}	96.17^{d}	158.90^{cd}	193.90^{def}	196.30^{c}
N_1K_2	20.58	52.92^{d}	95.42^{d}	155.40^{d}	191.20^{ef}	198.70^{bc}
N_1K_3	21.25	54.83^{bcd}	97.83^{cd}	157.10^{d}	189.40^{f}	192.10^{c}
N_2K_1	20.25	58.83^{abc}	104.00^{bc}	163.40^{abcd}	203.90^{bcd}	212.90^{a}
N_2K_2	20.58	54.08^{bcd}	102.00^{bcd}	166.00^{abc}	205.00^{bc}	208.50^{ab}
N_2K_3	19.67	56.17^{abcd}	107.30^{ab}	162.40^{bcd}	201.30^{cde}	210.40^{a}
N_3K_1	20.25	60.63^{a}	108.40^{ab}	168.30^{ab}	213.30^{ab}	214.80^{a}
N_3K_2	20.08	58.67^{abc}	111.00^{a}	170.40^{ab}	210.30^{abc}	215.50^{a}
N_3K_3	20.33	59.11^{ab}	111.60^{a}	171.20^{a}	215.80^{a}	218.50^{a}
CD @ 5%	NS	5.03	6.81	8.72	10.40	11.32

Table 7 **Effect of nitrogen and potash on per cent increase or decrease in plant height**

Treatments	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	At Shooting
N_1K_1	0.000	0.000	0.000	0.000	0.000	0.000
N_1K_2	2.033	-1.855	-0.779	-2.203	-1.392	1.223
N_1K_3	5.354	1.698	1.726	-1.133	-2.321	-2.139
N_2K_1	0.397	9.106	8.142	2.832	5.157	8.456
N_2K_2	2.033	0.297	6.062	4.468	5.725	6.215
N_2K_3	-2.479	4.173	11.573	2.203	3.814	7.183
N_3K_1	0.397	12.444	12.717	5.916	10.005	9.424
N_3K_2	4.462	8.810	15.917	7.237	8.458	9.781
N_3K_3	0.793	9.625	16.045	7.741	11.294	11.309

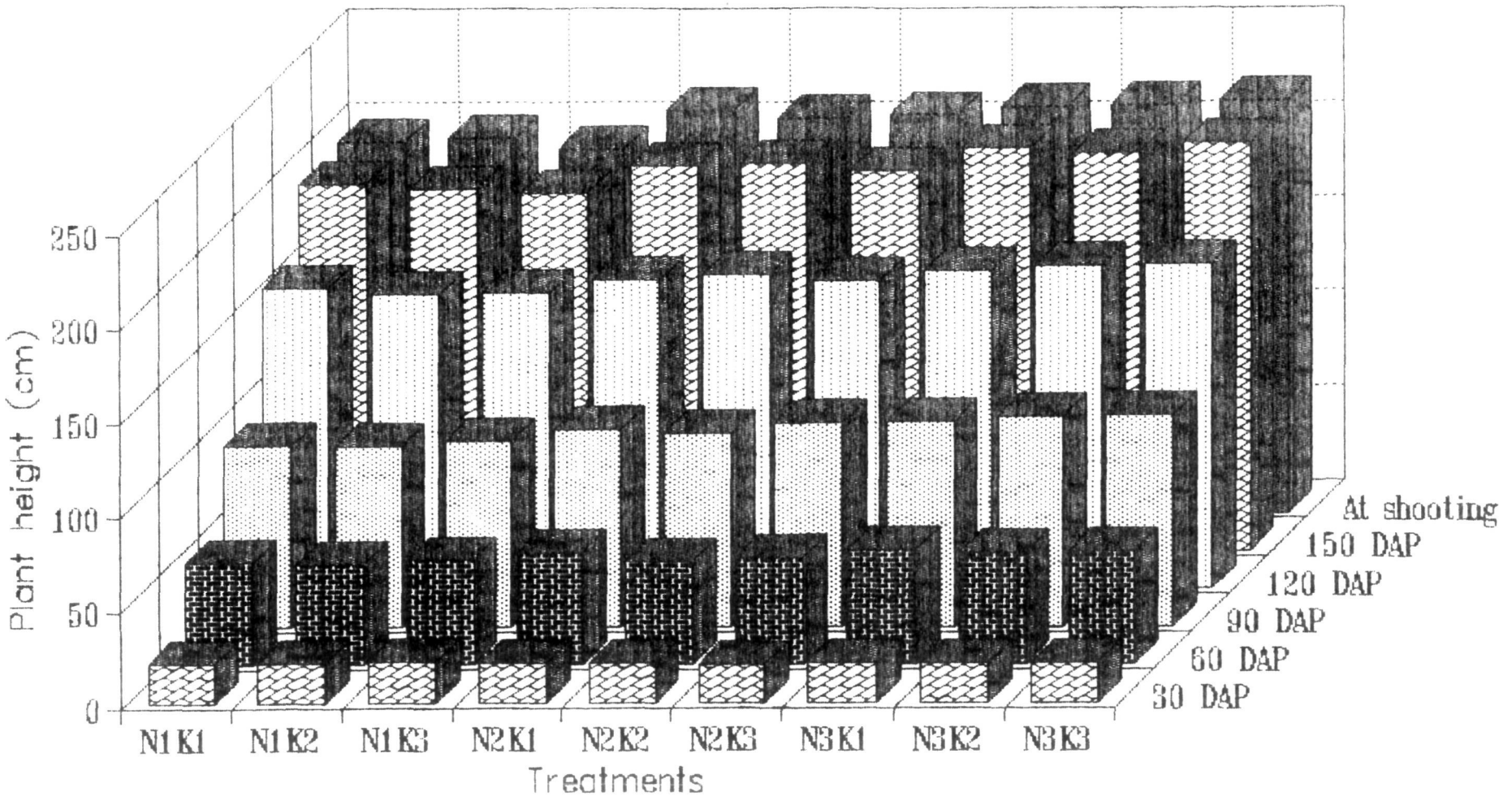

Fig.4 Influence of nitrogen and potash on plant height (cm)

The highest pseudostem girth was recorded in N_3K_2 (23.84cm) which was on par with N_3K_3 (22.83cm) while the lowest girth was in N_1K_1 (20.25cm) at 60 DAP, and a similar trend was observed at 90 and 120 DAP as well. At shooting the maximum pseudostem girth was recorded in the treatment in N_3K_3 (97.92 cm) followed by N_2K_3 (96.08 cm).

It was observed from the data, that the application of nitrogen and potash at lower levels showed least pseudostem girth during all the stages of crop growth. However, with increasing levels of potassium in combination with any nitrogen level, an increasing trend in plant girth was observed. Though there was an increasing trend in pseudostem girth during the crop growth, it was maximum (nearly 80% increase) between 90 and 120 DAP (Table 9) and there was not much significant increase in pseudostem girth after 150 DAP.

4.2.1.3 Number of functional leaves

The number of functional leaves varied significantly due to application of nutrients from 90 DAP till harvest (Table 10, 11 & Fig. 6). It can be observed from the table, that the maximum number of functional leaf production was between 90 and 150 DAP. Application of nitrogen at 225 g and 300 g in all combinations of potash, gave higher leaf production compared to nitrogen at 150 g during all stages of crop growth.

The highest number of functional leaves was recorded at shooting in N_3K_3 (19.25) while it was lowest in N_1K_1 (15.08). A similar trend was also noticed at harvest.

Table 8 **Effect of nitrogen and potash on pseudostem girth [cm]**

Treatments	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	At Shooting
N_1K_1	9.96^{c}	20.25^{b}	34.58^{d}	60.42^{e}	76.67^{c}	83.19^{e}
N_1K_2	11.25^{abc}	21.50^{ab}	36.42^{d}	63.33^{de}	81.92^{b}	87.92^{d}
N_1K_3	10.33^{bc}	21.79^{ab}	37.83^{bcd}	63.75^{de}	84.25^{ab}	89.50^{cd}
N_2K_1	10.54^{abc}	21.25^{ab}	37.00^{cd}	65.42^{cd}	80.83^{b}	89.50^{cd}
N_2K_2	10.51^{abc}	22.71^{ab}	40.33^{ab}	70.90^{ab}	84.17^{ab}	93.50^{bc}
N_2K_3	11.59^{ab}	20.34^{b}	42.75^{a}	69.58^{ab}	86.50^{a}	96.08^{a}
N_3K_1	11.97^{a}	22.07^{ab}	38.75^{bc}	69.17^{bc}	82.47^{b}	90.17^{bc}
N_3K_2	10.76^{abc}	23.84^{a}	40.45^{ab}	72.53^{ab}	86.67^{a}	94.80^{ab}
N_3K_3	9.86^{c}	23.82^{a}	42.88^{a}	73.33^{a}	87.50^{a}	97.92^{a}
CD @ 5%	1.61	2.95	3.29	3.95	3.52	4.05

Table 9 Effect of nitrogen and potash on per cent increase or decrease in girth

Treatments	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	At Shooting
N_1K_1	0.000	0.000	0.000	0.000	0.000	0.000
N_1K_2	12.952	6.173	5.321	4.816	6.848	5.686
N_1K_3	3.715	7.605	9.398	5.511	9.887	7.585
N_2K_1	5.823	4.938	6.998	8.275	5.426	7.585
N_2K_2	5.522	12.148	16.628	17.345	9.782	12.390
N_2K_3	16.365	0.444	23.626	15.161	12.821	15.495
N_3K_1	20.181	8.988	12.059	14.482	7.565	8.399
N_3K_2	8.032	17.728	16.975	20.043	13.043	13.956
N_3K_3	-1.004	17.630	24.002	21.367	14.125	17.706

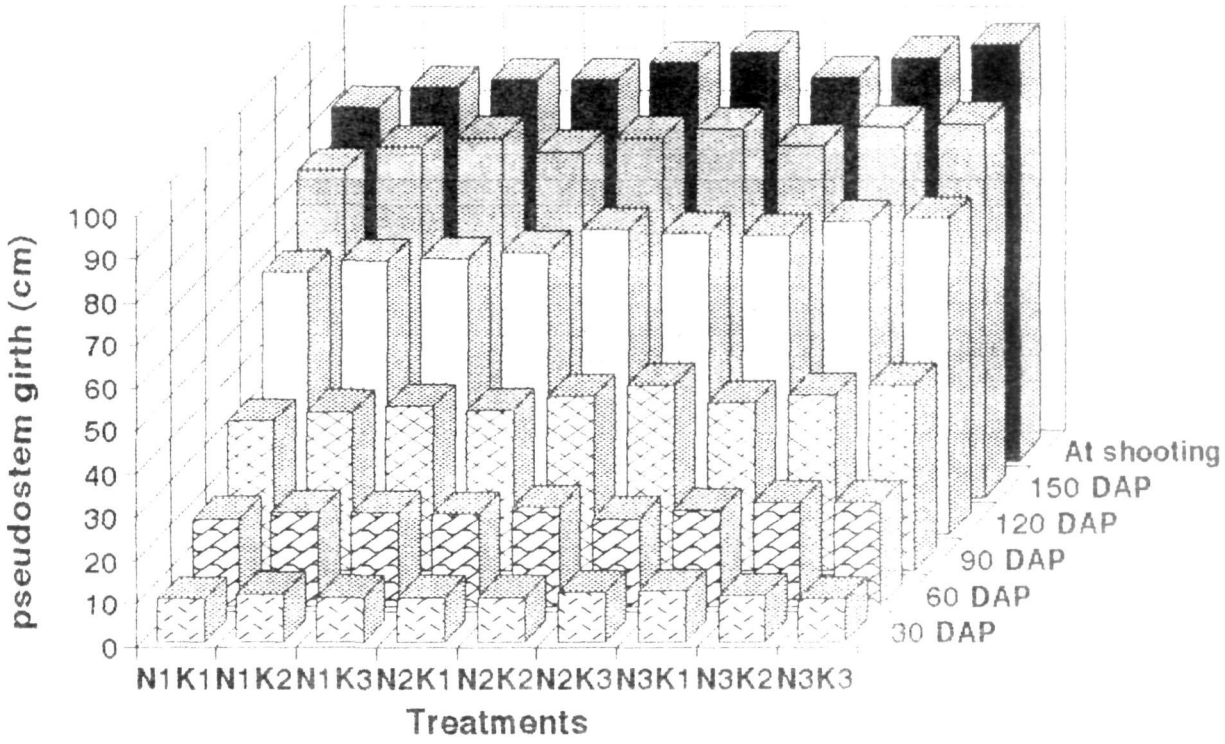

Fig.5 Influence of nitrogen and potash on pseudostem girth (cm)

Table 10 Effect of nitrogen and potash on number of functional leaves

Treatments	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	At Shooting	At Harvest
N_1K_1	7.33	9.25	10.17^{d}	12.00^{c}	14.08^{d}	15.08^{d}	9.58^{d}
N_1K_2	7.92	8.83	10.67^{bcd}	11.67^{c}	14.75^{d}	15.83^{d}	9.92^{d}
N_1K_3	8.00	9.33	10.33^{cd}	12.08^{c}	14.92^{d}	15.92^{d}	9.42^{d}
N_2K_1	7.58	9.33	10.50^{abc}	14.42^{b}	16.33^{c}	17.92^{bc}	11.42^{c}
N_2K_2	7.25	9.92	11.75^{ab}	15.00^{ab}	18.17^{ab}	18.17^{bc}	12.58^{abc}
N_2K_3	7.42	9.00	11.92^{ab}	15.83^{ab}	17.25^{bc}	17.58^{c}	12.83^{ab}
N_3K_1	7.33	9.58	11.92^{ab}	14.92^{ab}	17.00^{bc}	17.67^{c}	11.67^{bc}
N_3K_2	7.17	9.83	12.00^{a}	15.83^{ab}	18.25^{ab}	18.67^{ab}	12.42^{abc}
N_3K_3	7.83	10.00	12.33^{a}	16.25^{a}	18.83^{a}	19.25^{a}	12.92^{a}
CD @ 5%	NS	NS	1.30	1.64	1.35	0.88	1.26

Table 11 Effect of nitrogen and potash on per cent increase or decrease in functional leaves

Treatments	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	At Shooting
N_1K_1	0.000	0.000	0.000	0.000	0.000	0.000
N_1K_2	7.449	-4.756	4.916	-0.028	4.542	4.738
N_1K_3	8.375	0.857	1.549	0.662	5.630	5.276
N_2K_1	3.298	0.857	11.565	16.782	13.778	15.848
N_2K_2	-1.103	6.754	13.447	20.000	22.510	17.006
N_2K_3	1.213	-2.776	14.681	24.195	18.377	14.221
N_3K_1	0.000	3.445	14.681	19.571	17.176	14.658
N_3K_2	5.299	5.900	15.250	24.195	22.849	19.229
N_3K_3	6.386	7.500	17.518	26.154	25.226	21.662

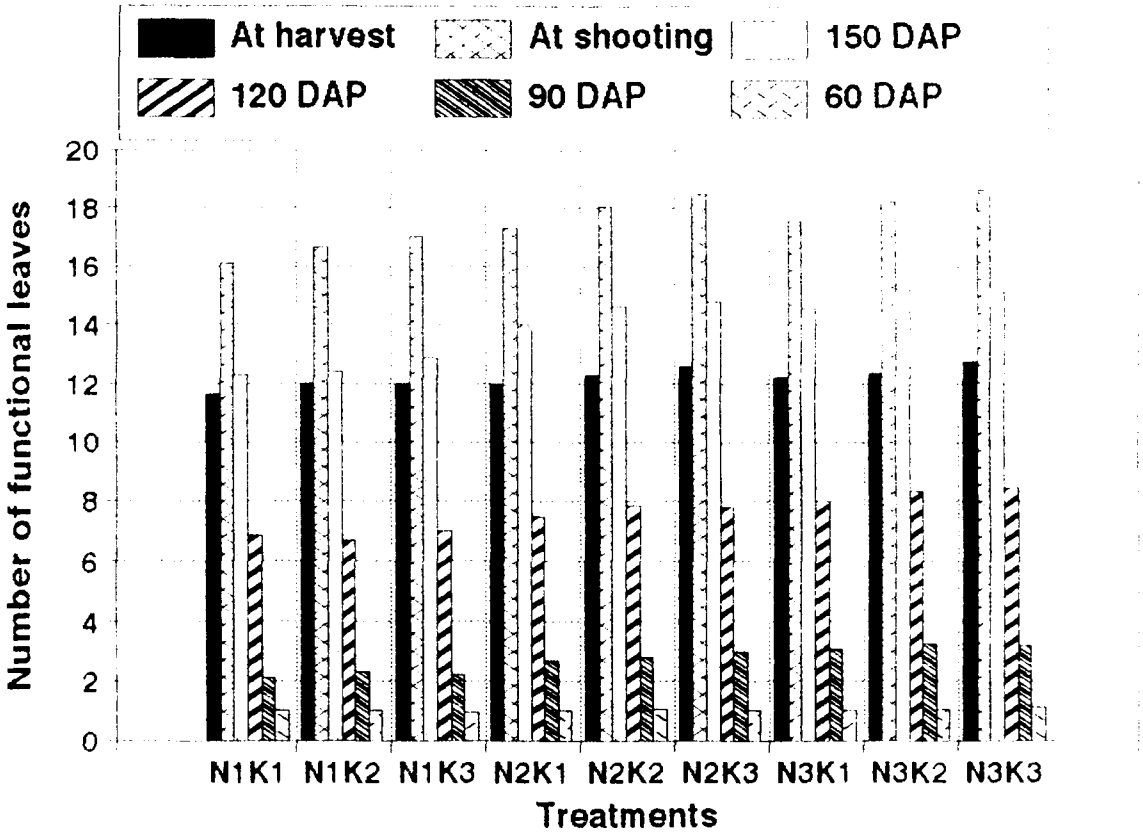

Fig. 6 Influence of nitrogen and potash on number of functional leaves

4.2.1.4. Leaf area (m^2)

A significant difference in leaf area was observed with different levels of nutrients in all the stages of crop growth (Table 12 & Fig. 7)

The maximum leaf area was recorded at shooting in N_3K_3 (18.65 m^2), which was on par with N_2K_2 (18.52 m^2), while N_1K_1 recorded the least leaf area (16.44 m^2). Nitrogen at N_2 and N_3 levels in combination with K_2 and K_3 levels recorded higher leaf area compared to all other treatments.

There was a steady increase in leaf area from planting till shooting, and the maximum leaf area production was observed between 90 and 150 DAP with nearly 250 per cent increase. This clearly indicates that the major photosynthetic activity was during this period.

4.2.1.5. Leaf Area Index (LAI)

The variation in leaf area index due to application of nutrients was highly significant from 90 DAP till harvest (Table 13). At almost all stages of crop growth, higher levels of nitrogen (N_2 and N_3), in combination with higher levels of potash (K_2 and K_3) recorded better LAI compared to lower levels (N_1 and K_1). At shooting, the LAI was highest in N_3K_3 level (5.63), while N_1K_1 recorded the least LAI (4.96). The maximum increase in LAI was 250 per cent between 90 and 120 DAP.

Table 12 Effect of nitrogen and potash on Leaf area (m^2)

Treatments	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	At Shooting	At Harvest
N_1K_1	0.51	1.00^{ab}	2.06^{d}	6.84^{e}	12.33^{d}	16.44^{e}	11.63^{bc}
N_1K_2	0.47	0.96^{ab}	2.32^{cd}	6.69^{e}	12.45^{d}	16.67^{de}	11.98^{b}
N_1K_3	0.49	0.92^{b}	2.20^{d}	7.00^{de}	12.91^{d}	17.03^{d}	12.01^{b}
N_2K_1	0.49	0.99^{ab}	2.66^{bc}	7.46^{cd}	13.94^{c}	17.35^{cd}	11.98^{b}
N_2K_2	0.53	1.02^{ab}	2.75^{ab}	7.82^{bc}	14.67^{ab}	18.08^{b}	12.32^{ab}
N_2K_3	0.47	1.00^{ab}	2.98^{ab}	7.79^{bc}	14.80^{ab}	18.52^{a}	12.60^{a}
N_3K_1	0.48	1.00^{ab}	3.09^{a}	8.01^{abc}	14.60^{b}	17.61^{c}	12.19^{ab}
N_3K_2	0.48	1.01^{ab}	3.22^{a}	8.35^{ab}	15.22^{a}	18.22^{ab}	12.40^{ab}
N_3K_3	0.49	1.07^{a}	3.17^{a}	8.46^{a}	15.12^{ab}	18.65^{a}	12.81^{a}
CD @ 5%	NS	0.14	0.35	0.61	0.59	0.54	0.83

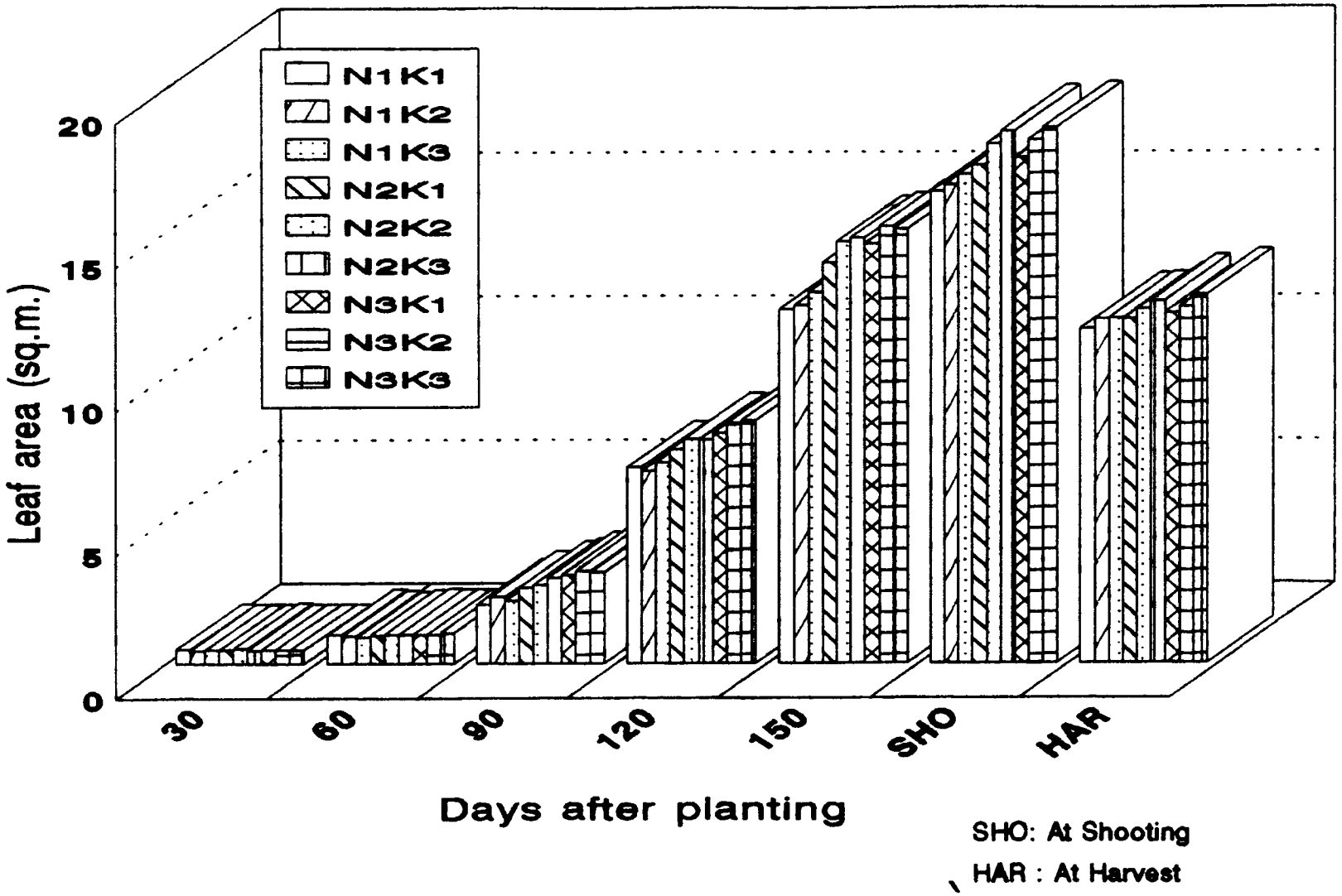

Fig 7 : Effect of nitrogen and potash on leaf area (sq.m.)

Table 13 Effect of nitrogen and potash on Leaf area index

Treatments	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	At Shooting	At Harvest
N_1K_1	0.16	0.30	0.62^{d}	2.07^{c}	3.72^{d}	4.97^{e}	3.51^{c}
N_1K_2	0.14	0.29	0.70^{d}	2.02^{c}	3.76^{d}	5.04^{de}	3.62^{bc}
N_1K_3	0.15	0.28	0.66^{d}	2.11^{c}	3.90^{d}	5.14^{cd}	3.63^{bc}
N_2K_1	0.13	0.29	0.80^{c}	2.25^{bc}	4.21^{c}	5.24^{c}	3.62^{bc}
N_2K_2	0.15	0.21	0.89^{bc}	2.36^{ab}	4.45^{ab}	5.46^{b}	3.72^{abc}
N_2K_3	0.13	0.30	0.89^{ab}	2.35^{ab}	4.47^{ab}	5.60^{b}	3.80^{ab}
N_3K_1	0.15	0.30	0.93^{ab}	2.42^{ab}	4.40^{b}	5.33^{c}	3.68^{abc}
N_3K_2	0.14	0.31	0.99^{a}	2.52^{a}	4.59^{a}	5.50^{ab}	3.74^{ab}
N_3K_3	0.15	0.31	0.96^{d}	2.55^{a}	4.56^{ab}	5.63^{a}	3.87^{a}
CD @ 5%	NS	NS	0.09	0.21	0.18	0.16	0.21

4.2.1.6 Total number of leaves produced

The total number of leaves produced during the crop growth differed significantly between treatments (Table 14). Application of nutrients at N_3K_3 level produced highest total number of leaves (30.17), which was on par with N_3K_2 (29.92) and N_2K_2 (29.50) levels, while it was lowest in N_1K_2 (26.25). All levels of potassium in combination with N_2 and N_3 levels produced higher number of leaves compared to other nutrient levels.

4.2.1.7. Phyllachron

The phyllachron varied significantly between treatments (Table 14). It ranged from 7.5 (N_3K_3) to 9.03 days (N_1K_1). Higher levels of nitrogen (N_2 and N_3) in all combinations of potash recorded a better Phyllachron.

4.2.2 Flowering and Maturity

The data on number of days taken for shooting from planting, number of days taken for maturity after shooting and total number of days from planting to harvest are presented in Table 15 & Fig. 8.

4.2.2.1 Number of days taken for shooting from planting

The differences in number of days taken for shooting from planting was highly significant between nutrient levels. Plants that received highest level of nutrients (N_3K_3) took minimum number of days to shoot (195.00), which was on par with N_3K_2 (195.59), N_2K_3 (196.58) and N_2K_2 (197.50) while, the plants that received lowest level of nutrient (N_1K_1) took maximum number of days (214.17).

Table 14 Effect of nitrogen and potash on phyllachron and total number of leaves produced

Treatments	Phyllachron	Total leaves produced
N_1K_1	9.03^{a}	27.00^{abc}
N_1K_2	8.86^{a}	26.25^{c}
N_1K_3	8.75^{a}	26.50^{bc}
N_2K_1	7.84^{b}	28.58^{abc}
N_2K_2	7.65^{b}	29.50^{ab}
N_2K_3	7.72^{b}	29.08^{abc}
N_3K_1	7.75^{b}	29.00^{abc}
N_3K_2	7.70^{b}	29.92^{a}
N_3K_3	7.50^{b}	30.17^{a}
CD @ 5%	0.36	3.21

4.2.2.2 Days taken for maturity from shooting

Days taken for maturity from shooting differed significantly among the treatments. It ranged between 130.67 days (N_3K_3) to 145.42 days (N_1K_1). It is seen from the table that, N_2K_2 which took 132.25 days for maturity from shooting was on par with the treatment N_3K_3. In general, higher levels of nitrogen (N_2 and N_3)in combination with higher levels of potash (K_2 and K_3) took less number of days for maturity after shooting compared to other nutrient levels.

4.2.2.3 Days taken from planting to harvest

There was a significant difference between treatments with respect to days taken from planting to harvest. Application of higher levels of nutrients (N_3K_3) rendered the plants ready to harvest in 325.67 days, which was on par with the treatments N_3K_2 (328.92) and N_2K_2 (328.83). The lower level of nutrients (N_1K_1) delayed harvesting (359.58 days). Adequate supply of nutrients reduced the duration from planting to harvest by 33.91 days.

4.2.3 Yield

The data on bunch weight, estimated yield and harvest index are presented in the Table 16 & Fig. 9.

4.2.3.1 Bunch weight

The bunch weight differed significantly between treatments due to nutrient effect. Plants receiving higher nutrient level (N_3K_3) recorded the highest bunch weight (49.84

Table 15 Effect of nitrogen and potash on flowering and maturity

Treatments	Days to shooting	Days from shooting to harvest	Days from Planting to harvest
N_1K_1	214.17[a]	145.42[a]	359.58[a]
N_1K_2	210.17[b]	143.50[ab]	353.67[b]
N_1K_3	210.25[b]	141.00[b]	351.25[b]
N_2K_1	202.42[c]	137.67[c]	339.92[c]
N_2K_2	197.50[de]	134.50[de]	332.00[de]
N_2K_3	196.58[e]	132.25[ef]	328.83[ef]
N_3K_1	199.34[d]	135.83[cd]	335.17[d]
N_3K_2	195.59[e]	133.33[def]	328.92[ef]
N_3K_3	195.00[e]	130.67[f]	325.67[f]
CD @ 5%	2.57	2.84	3.96

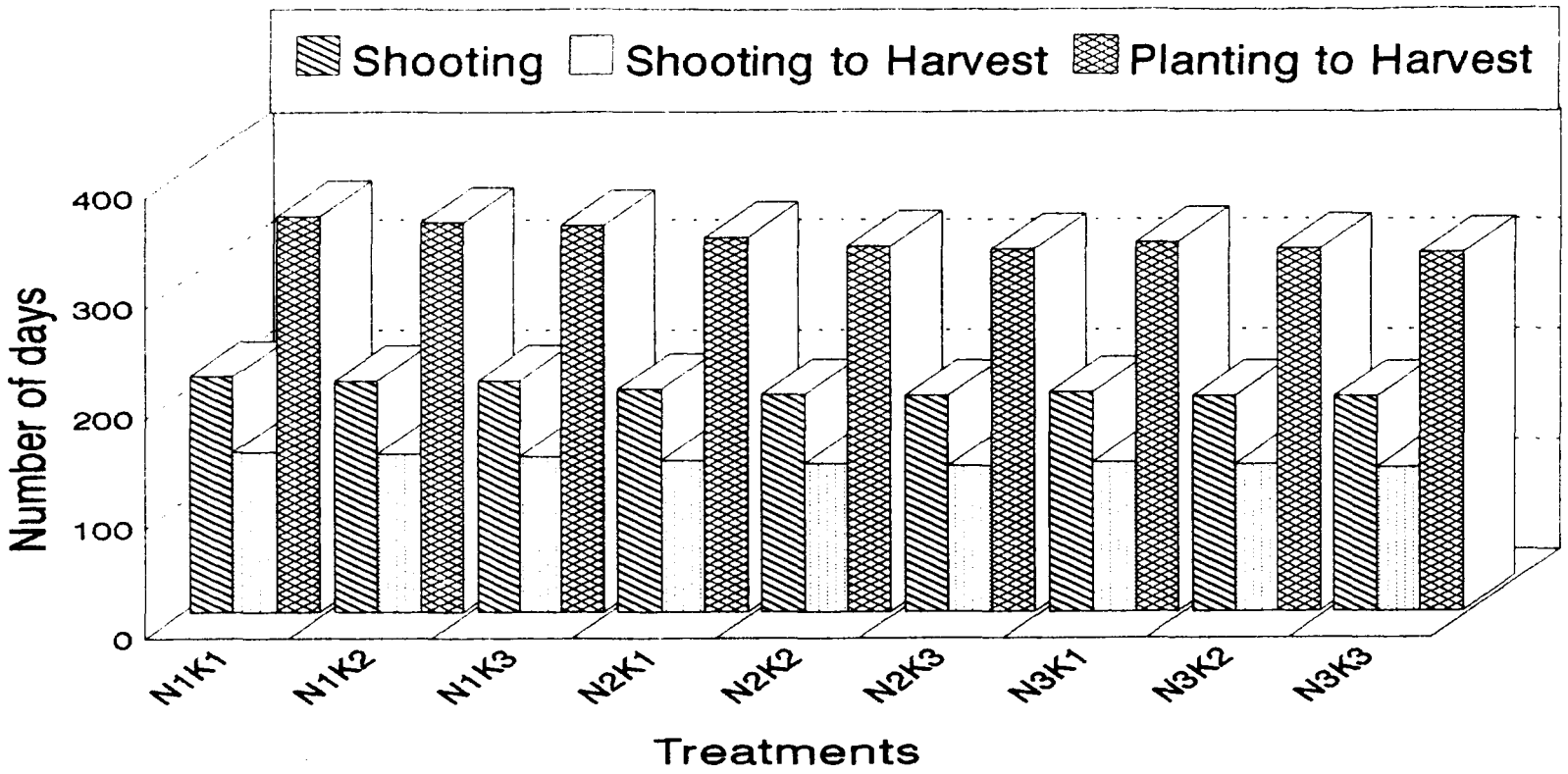

Fig. 8 Effect of nitrogen and potash on flowering and maturity

Banana cv Grande Naine

kg), which was on par with N_2K_2 (47.99 kg). N_1K_1 recorded least bunch weight (35.56 kg). There was an increase in the bunch weight upto 40 per cent due to adequate supply of nutrients.

4.2.3.2. Estimated yield

The application of nutrients had a significant effect on the estimated yield. The highest yield was obtained at N_3K_3 level (150.77 t/ha) which was on par with N_3K_2 (146.62 t/ha) and N_2K_2 (145.17 t/ha). In general, combinations of all levels of potassium with higher levels of nitrogen (N_2 and N_3) resulted in higher yields compared to other treatments.

4.2.3.3. Harvest Index

The harvest index differed significantly between the treatments. Maximum harvest index was recorded in the treatment N_2K_3 (43.307) which was on par with N_3K_2 (43.127), N_2K_3 (42.877) and N_2K_2 (42.313). The treatment N_1K_1 recorded the lowest harvest index (33.553).

4.2.4 Yield components

The data on yield components like, number of hands per bunch, number of fingers per bunch and average weight of fingers are presented in the Table 18.

PLATE II

A. General view of the field experiment

B. Banana bunch size in N at 150 and K at 200 g/pl

C. Banana bunch size in N at 225 and K at 300 g/pl

D. Banana bunch size in N at 300 and K at 400 g/pl

A

B

C

D

Table 16 Effect of nitrogen and potash on Yield of banana

Treatments	Bunch wt [kg/pl]	Yield [t/ha]	Harvest Index
N_1K_1	35.56^{g}	107.57^{g}	33.55^{c}
N_1K_2	37.95^{fg}	114.80^{fg}	36.26^{bc}
N_1K_3	39.09^{ef}	118.25^{ef}	38.85^{b}
N_2K_1	31.69^{de}	126.11^{de}	37.54^{b}
N_2K_2	37.99^{ab}	145.17^{ab}	42.31^{a}
N_2K_3	46.36^{bc}	140.24^{bc}	43.31^{a}
N_3K_1	43.18^{cd}	130.62^{cd}	36.60^{bc}
N_3K_2	48.47^{ab}	146.62^{ab}	43.13^{a}
N_3K_3	49.84^{a}	150.77^{a}	42.88^{a}
CD @ 5%	3.12	8.46	3.18

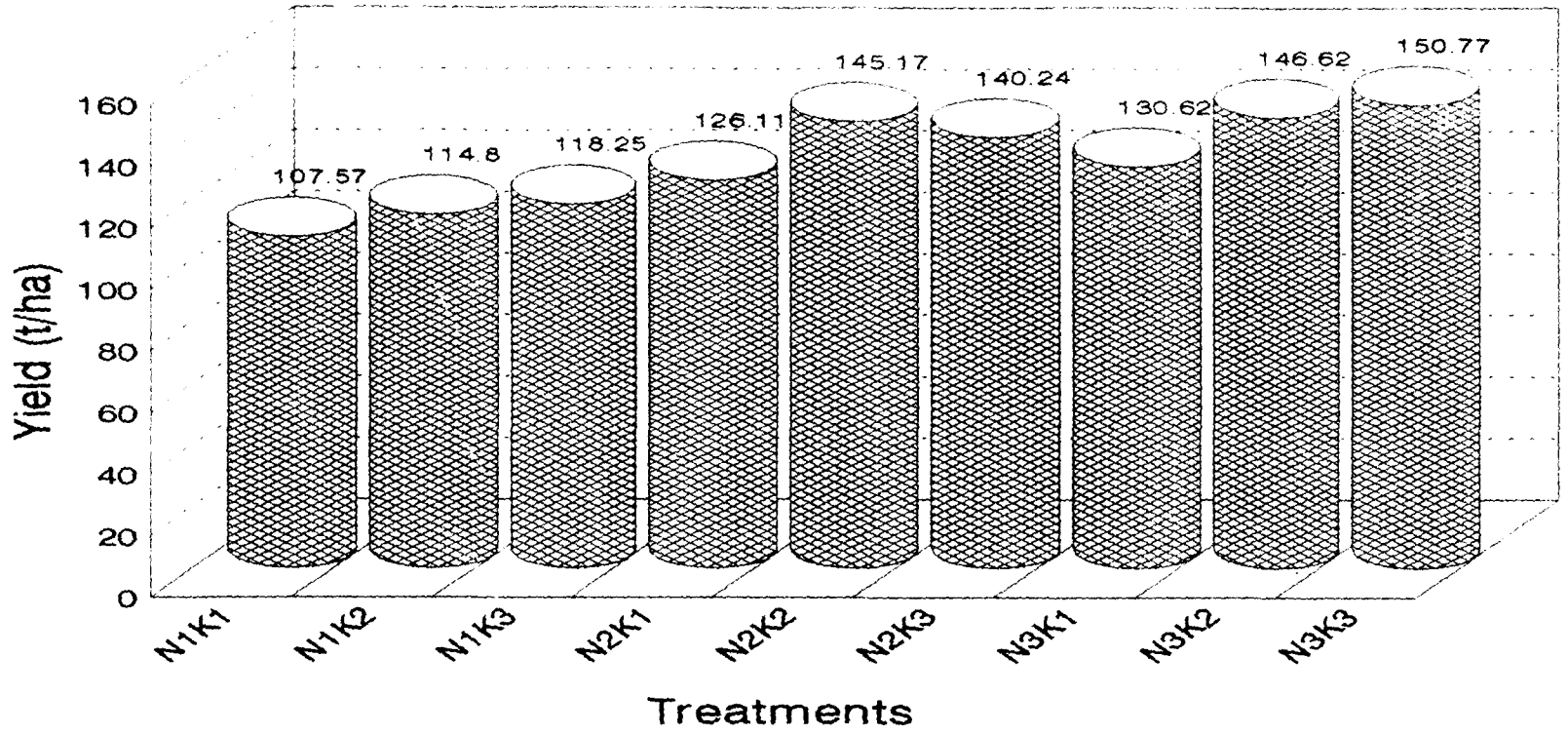

Fig. 9 Effect of nitrogen and potash on yield (t/ha)

Banana cv Grande Naine

4.2.4.1 Number of hands per bunch

The number of hands per bunch varied significantly between the nutrient levels. The maximum number of hands/bunch was recorded in the treatment N_2K_3 (10.42) which was on par with N_2K_2 (10.33), N_3K_2 (10.17) and N_3K_3 (10.00). The treatments N_1K_2 and N_1K_3 recorded the lowest number of hands/bunch (9.17).

4.2.4.2 Number of fingers per bunch

The number of fingers per bunch significantly varied between treatments. The highest number of fingers was recorded at N_3K_2 (190.67) which was on par with N_3K_3 (188.25), N_2K_3 (186.33) and N_2K_2 (183.88), while N_1K_1 recorded the lowest number of fingers (168.75).

4.2.4.3. Average weight of fingers

The nutrient levels had a significant effect on the average weight of fingers. The average weight of fingers varied from 178.26 g (N_1K_1) to 217.83 g (N_3K_3). Nutrient levels of N_2K_2 (215.31 g) and N_3K_2 (211.77 g) were at par with N_3K_3. With supply of adequate fertilizers there was an increase in the average fruit weight by 22 per cent.

4.2.5 Fruit quality

The data on fruit quality parameters like, physiological loss in weight (PLW), pulp to peel ratio, total soluble solids (TSS), reducing sugars, non-reducing sugars, total sugars, acidity and sugar - acid ratio are presented in Table 18.

Table 17 Effect of nitrogen and potash on yield components

Treatments	No. of hands/bunch	No. of fingers/bunch	Av.wt. of fingers [g]
N_1K_1	9.25^{b}	168.75^{c}	178.26^{d}
N_1K_2	9.17^{b}	172.93^{c}	185.43^{c}
N_1K_3	9.17^{b}	174.00^{c}	189.77^{c}
N_2K_1	9.50^{b}	177.58^{bc}	186.56^{c}
N_2K_2	10.33^{a}	183.88^{ab}	209.71^{b}
N_2K_3	10.42^{a}	186.33^{ab}	215.31^{ab}
N_3K_1	9.33^{b}	183.17^{b}	191.00^{c}
N_3K_2	10.17^{a}	190.67^{a}	211.77^{ab}
N_3K_3	10.00^{ab}	188.25^{ab}	217.83^{a}
CD @ 5%	0.50	7.35	6.95

4.2.5.1 Physiological loss in weight

The effect of varied levels of nutrients on PLW of fruits was significant. The per cent loss in weight during ripening was maximum at N_1K_2 (19.03%) compared to a minimum loss in N_3K_3 (12.39%). The other levels of nutrients (N_3K_2, N_2K_2, and N_2K_3) were on par with N_3K_3.

4.2.5.2 Pulp to peel ratio

Pulp to peel ratio differed significantly between the treatments. It varied from 2.66 (N_1K_1) to 3.42 (N_3K_3). Combination of higher nitrogen levels (N_2 and N_3) with potash (K_2 and K_3) had a better pulp to peel ratio compared to other treatments.

4.2.5.3 Total soluble solids

The TSS of the fruit were significantly influenced by different levels of nutrients. The total sugars accumulated at fruit ripening was influenced to a maximum extent at N_3K_3 level (26.23), while N_1K_1 had the lowest TSS (21.5). The TSS content in other treatments N_3K_2, N_3K_1 and N_2K_2 were on par with N_3K_3.

4.2.5.4 Reducing sugars

The per cent reducing sugar content of fruits varied significantly between treatments. The reducing sugar content steadily increased from 17.74 per cent (N_1K_1) to a maximum of 20.23 per cent (N_3K_3). All levels of potash in combination with higher levels of nitrogen (N_2 and N_3) recorded higher per cent of reducing sugars compared to other treatments.

4.2.5.5 Non-reducing sugars

The per cent non-reducing sugars in the fruits did not differ significantly between treatments. However, the per cent non-reducing sugar content varied from to 2.50 to 2.73.

4.2.5.6 Total sugars

There was significant difference in total sugar content of the fruits between treatments. The maximum per cent total sugars was recorded in N_3K_3 (22.96%) while it was lowest in N_1K_1 (20.34%). The nutrient levels N_2K_2, N_2K_3, N_3K_1 and N_3K_2 were on par with N_3K_3 in terms of total sugar content of fruits.

4.2.5.7 Acidity

The per cent acidity in fruits did not differ significantly due to varied levels of nutrients.

4.2.5.8 Sugar - acid ratio

The sugar-acid ratio of the fruits differed significantly between the treatments. The ratio increased from a lower level of 193.8 (N_1K_1) to a higher level of 287.2 (N_3K_3) which was on par with N_2K_2 (260.8).

4.2.6 Dry matter production and distribution

The dry matter production and distribution in different plant parts during various crop growth stages is presented in the Table 19.

Table 18 Effect of nitrogen and potash on post harvest quality of banana fruits

Treatments	PLW[1]	Pulp/peel ratio	TSS[2]	Reducing[1] sugars	Non-reducing[1] sugars	Total[1] sugars	Acidity[1]	Sugar-acid ratio
N_1K_1	18.31^{a}	2.66^{f}	21.50^{d}	17.74^{b}	2.60	20.34^{bc}	0.105	183.80^{g}
N_1K_2	19.03^{a}	2.70^{ef}	22.13^{cd}	18.00^{b}	2.73	20.73^{c}	0.100	207.50^{fg}
N_1K_3	17.94^{a}	2.79^{def}	22.70^{cd}	18.33^{b}	2.73	21.06^{bc}	0.097	217.20^{ef}
N_2K_1	15.30^{b}	2.93^{cdef}	23.40^{c}	19.49^{a}	2.51	22.00^{ab}	0.094	234.50^{de}
N_2K_2	14.50^{b}	3.10^{bc}	24.97^{ab}	19.97^{a}	2.71	22.68^{a}	0.087	260.80^{bc}
N_2K_3	14.60^{b}	2.98^{cde}	24.77^{b}	20.01^{a}	2.50	22.51^{a}	0.083	271.50^{ab}
N_3K_1	13.46^{bc}	3.00^{cd}	25.53^{ab}	19.75^{a}	2.54	22.29^{a}	0.090	247.70^{cd}
N_3K_2	12.44^{c}	3.37^{ab}	26.20^{a}	20.16^{a}	2.59	22.75^{a}	0.084	269.80^{ab}
N_3K_3	12.39^{c}	3.42^{a}	26.26^{a}	20.23^{a}	2.73	22.96^{a}	0.080	287.20^{a}
CD @ 5%	1.97	0.29	1.31	0.91	NS	1.06	NS	19.80

1. Expressed as per cent
2. Expressed as Brix°

From the data it is clear that in all the treatments the maximum dry matter accumulated between 60 to 150 days of planting. Maximum dry matter accumulation was recorded upto shooting and there after a reduction trend was observed in all plant parts till harvest except stalk and fruit.

In corm, in all the treatments there was an increasing trend in dry matter content, especially from 60 DAP till shooting. Thereafter, there was a marginal reduction. Similar trend was observed in case of pseudostem as well. In case of leaves, there was an increased dry matter content upto shooting, but declined at harvest by more than 50 per cent.

The total dry matter production (DMP) at harvest and its distribution in different plant parts differed significantly among the treatments except the dry matter production in stalk, where the treatments did not show any significant variations due to nutritional levels (Table 20 & Figs. 10a, 10b, 10c).

4.2.6.1 Corms

The DMP of corms decreased in general with the increased level of nutrients. The dry matter of corms varied from 3.90 kg (N_3K_2) to 4.48 kg (N_2K_1). The dry matter production of corms at nutrient levels of N_2K_2 and N_3K_3 were on par with each other. The DMP of corms was not affected by application of potash while, nitrogen application had some effect.

Table 19 Effect of N and K on dry matter production and distribution (kg) during various growth stages

Treatments		Corm	Ps. Stem	Leaf	Stalk	Infl	Fruit	Total
	Planting	0.040	0.045	0.085	-	-	-	0.17
N_1	60 DAP	0.778	0.812	0.543	-	-	-	2.13
K_1	150 DAP	4.100	2.315	2.450	-	-	-	8.86
	Shooting	5.004	3.714	3.250		0.648	-	12.61
	Harvest	4.286	3.250	1.374	1.11	-	5.06	15.08
	Planting	0.033	0.057	0.073	-	-	-	0.16
N_1	60 DAP	0.850	0.930	0.570	-	-	-	2.35
K_2	150 DAP	4.000	2.209	2.375	-	-	-	8.58
	Shooting	4.956	3.936	3.175	-	0.749	-	12.81
	Harvest	4.408	3.488	1.502	1.01	-	5.91	16.32
	Planting	0.045	0.051	0.079	-	-	-	0.17
N_1	60 DAP	0.912	0.885	0.514	-	-	-	2.31
K_3	150 DAP	4.206	2.173	2.500	-	-	-	8.87
	Shooting	5.114	3.900	3.210	-	0.675	-	12.89
	Harvest	4.400	3.544	1.644	1.04	-	6.75	17.38
	Planting	0.039	0.051	0.079	-	-	-	0.17
N_2	60 DAP	0.619	0.963	0.647	-	-	-	2.23
K_1	150 DAP	4.206	2.040	2.200	-	-	-	8.45
	Shooting	5.127	3.640	2.900	-	0.637	-	12.30
	Harvest	4.485	3.473	1.705	1.03	-	6.42	17.12
	Planting	0.044	0.053	0.081	-	-	-	0.18
N_2	60 DAP	1.112	1.037	0.750	-	-	-	2.90
K_2	150 DAP	4.420	3.007	2.890	-	-	-	10.32
	Shooting	5.350	4.267	3.628	-	0.797	-	14.04
	Harvest	4.111	3.957	1.900	1.02	-	8.07	19.06
	Planting	0.047	0.054	0.080	-	-	-	0.18
N_2	60 DAP	1.008	0.913	0.675	-	-	-	2.60
K_3	150 DAP	4.159	2.910	2.670	-	-	-	9.74
	Shooting	5.042	4.000	3.339	-	0.817	-	13.20
	Harvest	4.000	3.998	1.916	1.10	-	8.41	19.43
	Planting	0.048	0.050	0.087	-	-	-	0.19
N_3	60 DAP	0.774	0.897	0.703	-	-	-	2.37
K_1	150 DAP	4.315	2.323	2.500	-	-	-	9.14
	Shooting	5.264	3.778	2.850	-	0.727	-	12.62
	Harvest	4.236	3.521	1.798	1.01	-	6.55	17.13
	Planting	0.044	0.049	0.092	-	-	-	0.18
N_3	60 DAP	0.890	0.975	0.778	-	-	-	2.64
K_2	150 DAP	4.239	3.150	2.605	-	-	-	9.99
	Shooting	4.614	4.775	2.960	-	0.734	-	13.08
	Harvest	3.946	4.123	1.950	1.04	-	8.39	19.45
	Planting	0.47	0.52	0.072	-	-	-	0.17
N_3	60 DAP	0.994	0.975	0.808	-	-	-	2.78
K_3	150 DAP	3.980	3.660	2.544	-	-	-	10.18
	Shooting	4.750	4.808	2.980	-	0.895	-	13.43
	Harvest	4.060	4.275	1.992	1.22	-	8.65	20.19

4.2.6.2 Pseudostem

The pseudostem dry matter at harvest increased with increased level of nutrient application. The maximum dry matter of pseudostem was recorded at nutrient levels of N_3K_3 (4.275 kg) and it was on par with N_3K_2 (4.123 kg), N_2K_3 (3.998 kg) and N_2K_2 (3.957 kg). The lowest DMP in pseudostem was observed at N_1K_1 nutrient level (3.250 kg). In general, it was observed that increased level of potash with any level of nitrogen had an increasing trend in DMP.

4.2.6.3 Leaf

The different nutrient levels differed significantly with respect to DMP in leaves at harvest. The maximum dry matter in leaf was observed at a nutrient level of N_3K_3 (1.992 kg) followed by N_3K_2 (1.950 kg), N_2K_3 (1.916 kg) and N_2K_2 (1.900 kg) levels. The lowest dry matter accumulation was obtained with lower level of nitrogen and potash (N_1K_1 - 1.374 kg). In general, as seen in dry matter production of pseudostem, there was an increasing trend in dry matter accumulation in leaves, with increased levels of potash at all levels of nitrogen. However, the DMP of leaves was more influenced by nitrogen levels than that of potash. Both N and K at higher levels (N_2, N_3, K_2 and K_3) recorded significantly higher DMP of leaves compared to N_1K_1 levels.

4.2.6.4 Stalk

The dry matter accumulation in stalk did not differ significantly between nutrient levels. The DMP of stalk varied from 1.006 kg to 1.217 kg.

4.2.6.5. Fruits

The DMP of fruits differed highly significantly between different nutrient levels. The increase in DMP of fruits at higher levels was more than 75 per cent compared to the lowest nutrient level. The maximum fruit DMP of 8.648 kg was recorded in N_3K_3 which was on par with other nutrient levels N_2K_3 (8.412 kg) and N_3K_2 (8.388 kg). The lowest DMP of fruits was recorded at N_1K_1 level (5.060kg). In general, the DMP of fruits recorded was highest at higher levels of N (N_2 and N_3) and K (K_2 and K_3).

The total DMP of the plant at shooting differed significantly due to varying levels of nutrients. The total DMP of the plant varied from 15.08 kg (N_1K_1) to 20.19 kg (N_3K_3). The total DMP was also higher at N_3K_2, N_2K_3 and N_2K_2 levels compared to other nutrient levels. In general, potash at all the three levels with higher levels of nitrogen (N_2 and N_3) has shown higher dry matter production and distribution through out the crop growth.

4.2.7 Nutrient uptake

The nutrient uptake (N,P and K) in different plant parts at various stages of crop growth is presented in the Tables 21, 22 and 23. During vegetative phase, there was a rapid increase in the nitrogen content of leaves in all the treatments upto shooting. The maximum leaf nitrogen uptake at shooting was recorded in the treatment N_2K_2 (59.70 g) followed by N_3K_3 (58.60 g) while the lowest nitrogen content was in N_1K_2 (39.46 g). Leaf nitrogen content declined by more than 60 per cent between shooting and harvest. There was an increasing trend in nitrogen uptake in pseudostem and corm as well till shooting, which declined later at a lower rate.

Table 20 Effect of N and K on dry matter accumulation and distribution in different plant parts at harvest (g/plant)

Treatment	Corm	Ps. Stem	Leaf	Stalk	Fruit	Total
N_1K_1	4.29^{ab}	3.25^{b}	1.37^{d}	1.11	5.06^{e}	15.08^{e}
N_1K_2	4.41^{ab}	3.49^{b}	1.50^{cd}	1.01	5.92^{d}	16.32^{d}
N_1K_3	4.40^{ab}	3.54^{b}	1.64^{bcd}	1.04	6.75^{c}	17.38^{c}
N_2K_1	4.49^{a}	3.47^{b}	1.71^{abc}	1.03	6.43^{c}	17.11^{c}
N_2K_2	4.11^{ab}	3.96^{a}	1.90^{ab}	1.02	8.07^{b}	19.07^{b}
N_2K_3	4.00^{b}	3.99^{a}	1.92^{ab}	1.10	8.41^{ab}	19.43^{b}
N_3K_1	4.24^{ab}	3.52^{b}	1.80^{abc}	1.01	6.55^{c}	17.13^{c}
N_3K_2	3.95^{b}	4.12^{a}	1.95^{ab}	1.04	8.39^{ab}	19.45^{b}
N_3K_3	4.06^{ab}	4.27^{a}	1.99^{a}	1.22	8.65^{a}	20.19^{a}
CD @ 5%	0.46	0.38	0.32	NS	0.47	0.61

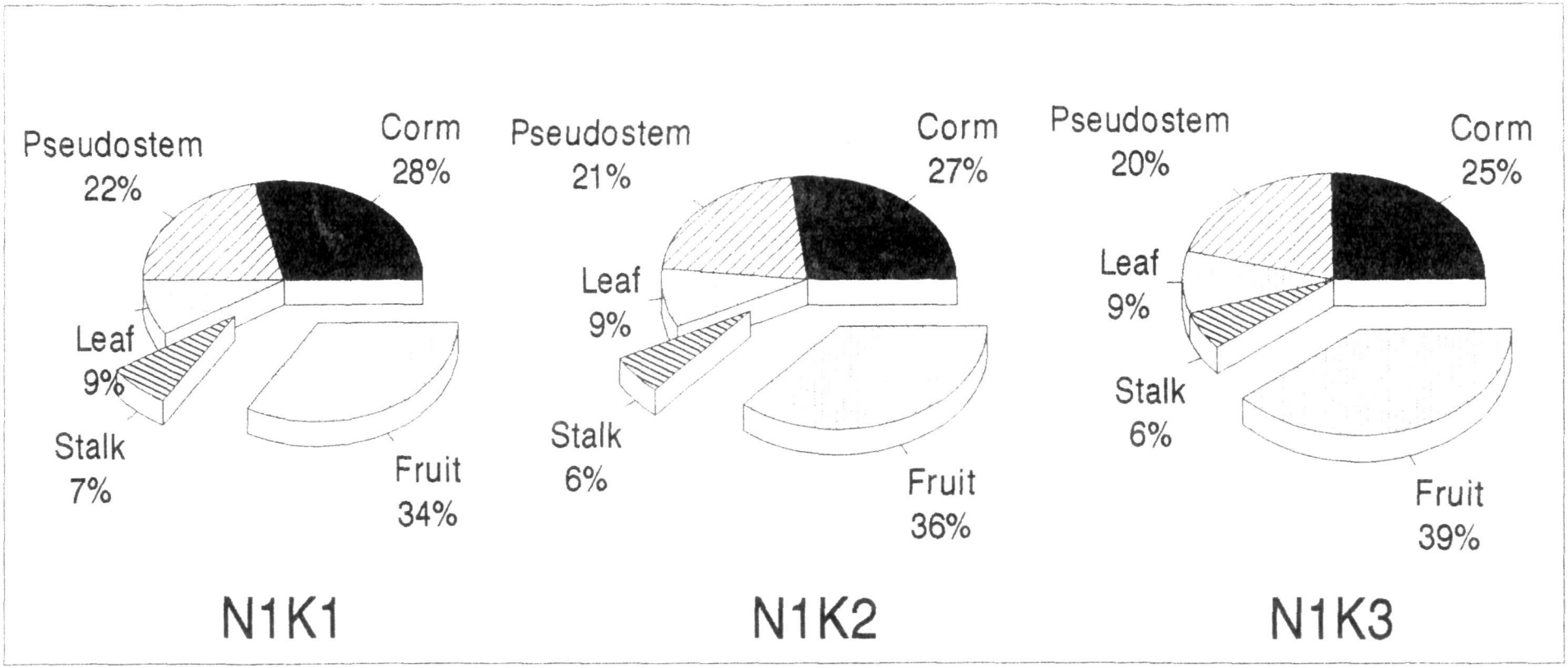

Fig. 10a Distribution of dry matter in different parts at harvest

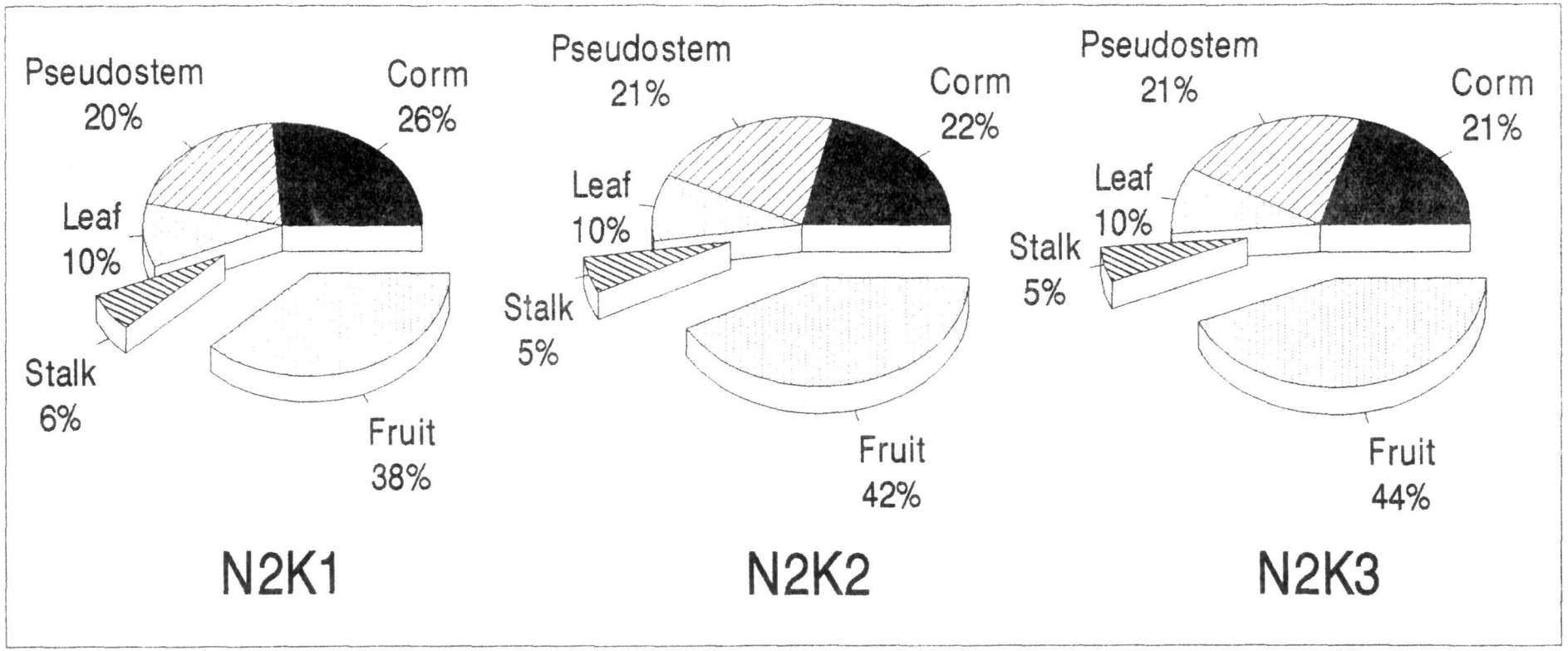

Fig. 10b Distribution of dry matter in different parts at harvest

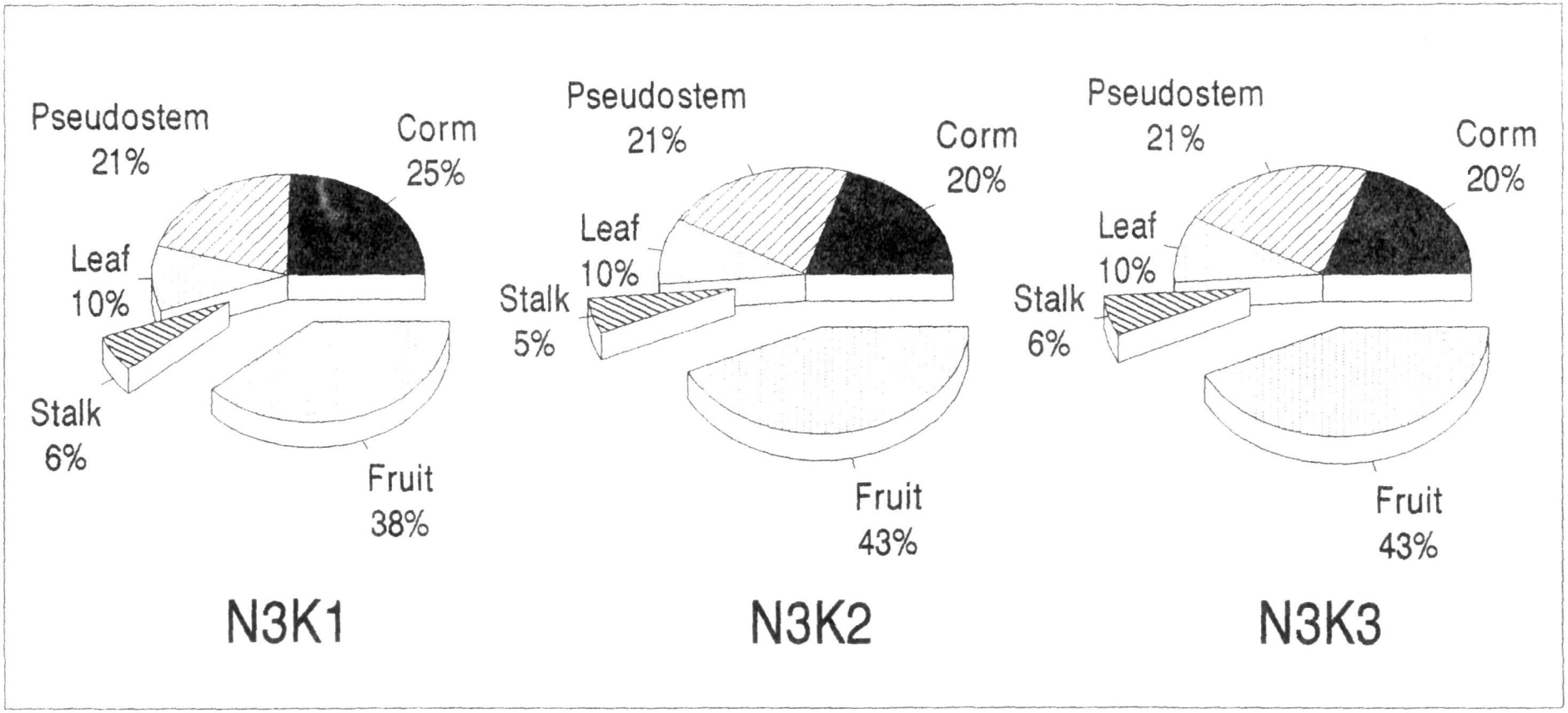

Fig. 10c Distribution of dry matter in different parts at harvest

Table 21 Effect of nitrogen and potash on nitrogen (g) uptake by different plant parts at various growth stages

	Corm	Ps.stem	Leaf	Stalk	Infl.	Fruit	Total
Planting	0.066	0.098	1.176	-	-	-	1.340
N_1 60 DAP	4.116	6.747	10.236	-	-	-	2.109
K_1 150 DAP	29.400	17.462	23.461	-	-	-	7.032
Shooting	46.76	29.714	40.926		5.025	-	122.430
Harvest	37.33	23.160	19.140	12.26	-	49.82	141.710
Planting	0.070	0.087	1.720	-	-	-	1.877
N_1 60 DAP	3.908	5.932	11.158	-	-	-	20.998
K_2 150 DAP	31.746	18.315	25.758	-	-	-	75.819
Shooting	44.605	31.255	39.468	-	4.998	-	120.326
Harvest	37.71	23.56	19.82	12.14	-	49.13	142.360
Planting	0.087	0.103	1.420	-	-	-	1.610
N_1 60 DAP	3.746	5.838	10.750	-	-	-	20.334
K_3 150 DAP	28.887	17.956	24.500	-	-	-	71.343
Shooting	46.090	30.175	42.250	-	5.726	-	124.241
Harvest	39.00	24.18	20.23	11.30	-	50.67	145.380
Planting	0.067	0.088	1.263	-	-	-	1.418
N_2 60 DAP	4.020	6.121	9.885	-	-	-	20.026
K_1 150 DAP	30.856	18.040	23.760	-	-	-	72.656
Shooting	49.123	31.476	40.390		5.375	-	126.364
Harvest	40.16	26.92	25.41	14.82		54.60	161.910
Planting	0.067	0.088	1.263	-	-	-	1.418
N_2 60 DAP	5.564	6.228	12.328	-	-	-	24.120
K_2 150 DAP	47.454	26.970	32.465	-	-	-	106.889
Shooting	51.740	45.750	59.700	-	7.765	-	164.955
Harvest	46.05	31.75	33.37	17.82	-	77.13	206.060
Planting	0.077	0.079	1.493	-	-	-	1.649
N_2 60 DAP	4.074	5.148	11.895	-	-	-	21.117
K_3 150 DAP	42.880	23.270	28.390	-	-	-	94.540
Shooting	53.176	42.674	52.725		6.500	-	155.075
Harvest	40.22	31.22	31.30	16.69	-	75.88	195.310
Planting	0.069	0.132	1.725	-	-	-	1.926
N_3 60 DAP	3.998	5.128	12.000	-	-	-	21.126
K_1 150 DAP	32.177	19.000	28.754	-	-	-	79.931
Shooting	53.080	33.089	40.175		5.860	-	132.204
Harvest	45.390	26.60	26.45	13.55	-	57.64	169.630
Planting	0.117	0.096	1.726	-	-	-	1.939
N_3 60 DAP	4.684	6.025	10.750	-	-	-	21.459
K_2 150 DAP	34.985	17.145	32.600	-	-	-	84.730
Shooting	47.325	37.542	57.440		6.925	-	149.232
Harvest	45.39	31.70	33.42	18.56	-	76.57	205.640
Planting	0.089	0.114	1.420	-	-	-	1.623
N_3 60 DAP	4.789	5.774	11.174	-	-	-	21.737
K_3 150 DAP	33.147	16.996	31.700	-	-	-	81.843
Shooting	49.126	40.146	58.600		7.014	-	154.886
Harvest	44.06	32.26	36.33	18.55		77.41	208.610

Table 22 Effect of nitrogen and potash on phosphorus (g) uptake by different plant parts at various growth stages

Treatments	Corm	Ps.stem	Leaf	Stalk	Infl.	Fruit	Total
N_1 60 DAP	0.713	0.217	0.875	-	-	-	1.805
K_1 150 DAP	3.975	0.804	1.890	-	-	-	6.669
Shooting	5.023	1.524	2.330	-	0.925	-	9.802
Harvest	3.950	1.611	2.060	2.891	-	4.210	14.722
N_1 60 DAP	0.615	0.195	0.950	-	-	-	1.760
K_2 150 DAP	2.850	0.913	1.945	-	-	-	5.708
Shooting	4.690	1.900	2.418	-	0.850	-	9.858
Harvest	4.231	1.810	2.301	2.770	-	4.640	15.752
N_1 60 DAP	0.913	0.200	0.640	-	-	-	1.753
K_3 150 DAP	3.465	0.795	1.850	-	-	-	6.110
Shooting	5.030	1.704	2.500	-	0.900	-	10.134
Harvest	4.482	1.690	2.490	2.611	-	4.740	16.013
N_2 60 DAP	0.712	0.206	0.746	-	-	-	1.664
K_1 150 DAP	2.750	0.880	1.726	-	-	-	5.356
Shooting	4.850	1.800	2.690	-	0.815	-	10.155
Harvest	4.600	1.760	2.610	2.711	-	4.600	16.281
N_2 60 DAP	0.912	0.320	1.025	-	-	-	2.257
K_2 150 DAP	3.900	1.405	2.000	-	-	-	7.305
Shooting	6.000	2.995	3.438	-	1.025	-	13.458
Harvest	5.500	2.760	3.350	3.460	-	7.011	22.081
N_2 60 DAP	0.850	0.265	0.850	-	-	-	1.965
K_3 150 DAP	3.700	1.000	1.650	-	-	-	6.350
Shooting	5.950	2.485	3.365	-	0.960	-	12.760
Harvest	5.430	2.500	3.120	3.280	-	7.500	21.830
N_3 60 DAP	0.650	0.210	0.925	-	-	-	1.785
K_1 150 DAP	2.700	0.875	1.900	-	-	-	5.475
Shooting	5.087	1.825	2.740	-	0.770	-	10.422
Harvest	4.700	1.790	2.600	2.610	-	4.790	16.490
N_3 60 DAP	0.624	0.290	0.900	-	-	-	1.814
K_2 150 DAP	3.005	1.015	2.400	-	-	-	6.420
Shooting	5.900	2.625	3.675	-	0.890	-	13.090
Harvest	5.45	2.451	3.500	3.140	-	6.890	21.431
N_3 60 DAP	0.717	0.300	1.100	-	-	-	2.117
K_3 150 DAP	2.900	1.286	2.540	-	-	-	6.726
Shooting	5.750	2.725	3.600	-	0.925	-	13.000
Harvest	5.440	2.660	3.46	3.750	-	7.091	22.400

Table 23 Effect of nitrogen and potash on potash uptake (g) by different plant parts at various growth stages

Treatments	Corm	Ps.stem	Leaf	Stalk	Infl.	Fruit	Total
Planting	0.104	0.075	0.070	-	-	-	0.249
N_1 60 DAP	12.880	22.850	29.060	-	-	-	65.390
K_1 150 DAP	38.750	51.640	60.725	-	-	-	151.115
Shooting	54.975	72.850	98.325	-	23.56	-	249.710
Harvest	60.570	60.750	21.760	35.980	-	102.56	281.620
Planting	0.145	0.095	0.100	-	-	-	0.340
N_1 60 DAP	10.090	23.125	32.750	-	-	-	65.965
K_2 150 DAP	46.250	50.450	68.250	-	-	-	164.950
Shooting	67.895	73.125	100.500	-	27.40	-	268.920
Harvest	71.220	69.560	25.650	40.670	-	121.53	328.630
Planting	0.125	0.085	0.090	-	-	-	0.300
N_1 60 DAP	13.125	22.250	30.075	-	-	-	65.450
K_3 150 DAP	48.545	51.475	69.800	-	-	-	169.820
Shooting	69.423	75.250	99.750	-	26.750	-	271.173
Harvest	74.330	69.140	30.370	43.760		128.800	346.400
Planting	0.130	0.080	0.110	-	-	-	0.320
N_2 60 DAP	11.550	20.650	30.570	-	-	-	62.770
K_1 150 DAP	43.250	47.250	71.436	-	-	-	161.936
Shooting	60.660	71.285	93.325	-	25.500	-	250.770
Harvest	64.330	67.410	26.030	39.040	-	119.790	316.600
Planting	0.137	0.070	0.105	-	-	-	0.312
N_2 60 DAP	18.265	23.950	32.850	-	-	-	75.065
K_2 150 DAP	69.750	56.100	77.880	-	-	-	203.730
Shooting	81.000	96.700	124.660	-	32.750	-	335.110
Harvest	84.730	86.600	41.960	60.100	-	158.390	431.780
Planting	0.147	0.080	0.090	-	-	-	0.317
N_2 60 DAP	13.450	25.400	35.120	-	-	-	73.970
K_3 150 DAP	56.805	59.200	79.200	-	-	-	195.205
Shooting	72.775	88.500	117.256	-	31.245	-	309.776
Harvest	87.170	85.830	41.030	57.330	-	151.600	422.960
Planting	0.114	0.060	0.080	-	-	-	0.254
N_3 60 DAP	11.365	23.750	29.560	-	-	-	59.675
K_1 150 DAP	37.000	49.075	75.000	-	-	-	161.075
Shooting	50.880	75.250	110.332	-	26.260	-	262.722
Harvest	66.360	69.490	27.770	44.910	-	127.070	335.600
Planting	0.108	0.090	0.100	-	-	-	0.298
N_3 60 DAP	12.100	21.470	34.265	-	-	-	67.835
K_2 150 DAP	39.600	50.000	76.895	-	-	-	166.495
Shooting	69.400	90.120	110.000	-	30.430	-	299.950
Harvest	87.260	85.150	39.590	63.950	-	161.100	437.050
Planting	0.112	0.065	0.085	-	-	-	0.262
N_3 60 DAP	13.200	23.065	33.210	-	-	-	46.423
K_3 150 DAP	40.700	52.456	68.746	-	-	-	161.902
Shooting	88.500	87.850	109.565	-	31.770	-	317.685
Harvest	87.830	88.930	42.190	62.960	-	143.460	425.370

The phosphorus content in different plant parts during various growth stages generally showed an increasing trend till shooting. Thereafter, either the phosphorus content remained the same or decreased negligibly.

During the vegetative phase, leaves had the highest potash content followed by pseudostem and corm. The increase in the rate of potash content was steady throughout the growth period till shooting and thereafter it declined by more than 400 per cent at harvest. The maximum leaf potassium content (124.66 g) at shooting was recorded in the treatment N_2K_2 and the lowest in N_1K_1 (98.32 g)

The data on uptake of nitrogen, phosphorus and potash in different plant parts at harvest are presented in the Tables 24, 25 and 26.

4.2.7.1 Nitrogen

The uptake of nitrogen in different plant parts at harvest varied significantly between the treatments (Table 24 & Fig. 11a). In corm the maximum N uptake was recorded in the treatment N_2K_2 (46.05 g) which was on par with N_3K_2 (45.39 g) and N_3K_3 (44.06 g). In pseudostem the nitrogen uptake varied from 23.16 g (N_1K_1) to 32.26 g (N_3K_3). In leaf the maximum nitrogen uptake was recorded in N_3K_3 (36.33 g) while the minimum was N_1K_1 (19.14 g). In fruits the nitrogen uptake varied from 49.13 g (N_1K_2) to 77.41 g (N_3K_3) which was on par with N_2K_2 (77.13 g).

4.2.7.2 Phosphorus

The accumulation of phosphorus differed significantly in all the plant parts at harvest between the treatments (Table 25). In corm and pseudostem the maximum P

content was observed in N_2K_2 (5.50 g; 2.76 g) while the minimum was in N_1K_1 (3.95 g; 1.61 g). The treatment N_3K_2 recorded a higher uptake of P in the leaf (3.50 g), while N_1K_1 had the lowest (2.06 g). The maximum P uptake in fruits was in the treatment N_2K_2 (7.50 g) and minimum in N_1K_1 (4.21 g).

4.2.7.3 Potash

The potash content in the plant parts differed significantly due to various nutrient levels in all crop growth stages (Table 26 & Fig. 11b). The total potash content was generally higher in the plant when compared to N and P content. The maximum K content in corm, pseudostem and leaf was observed in the treatment N_3K_3 (87.83; 88.93 and 42.19 g respectively), while it was minimum in N_1K_1 (60.57; 60.75 and 21.76 g). The treatment N_3K_2 recorded the maximum potash content in the fruits (161.10 g) which was on par with N_2K_2 (158.39 g). The total K content in the plant varied from 218.62 g (N_1K_1) to 437.05 g (N_3K_2), which was on par with N_3K_3 (425.37 g) and N_2K_2 (422.96 g).

4.2.8 Nutrient status of the soil

The data on availability of major nutrients (NPK) in the soil during different stages of growth are presented in the Tables 27, 28 and 29.

4.2.8.1 Available Nitrogen

The available nitrogen in soil due to various nutritional levels differed significantly during all stages of crop growth. There was an increasing trend in the available soil

Table 24 Effect of N and K on nitrogen uptake by different plant parts at harvest (g/plant)

Treatment	Corm	Ps. stem	Leaf	Stalk	Fruit	Total
N_1K_1	37.33^{d}	23.16^{f}	19.14^{d}	12.26^{de}	49.82^{d}	141.71
N_1K_2	37.71^{d}	23.56^{f}	19.82^{d}	12.14^{de}	49.13^{d}	142.36
N_1K_3	39.00^{d}	24.18^{e}	20.23^{d}	11.30^{e}	50.67^{cd}	145.38
N_2K_1	40.16^{cd}	26.92^{d}	25.41^{c}	14.82^{bc}	54.60^{bc}	161.91
N_2K_2	46.05^{a}	31.75^{b}	33.37^{b}	17.82^{a}	77.13^{a}	206.06
N_2K_3	40.22^{bc}	31.22^{c}	31.30^{b}	16.69^{ab}	75.88^{a}	195.31
N_3K_1	45.39^{cd}	26.60^{d}	26.45^{c}	13.55^{cd}	57.64^{b}	169.63
N_3K_2	45.39^{ab}	31.70^{b}	33.42^{b}	18.56^{a}	76.57^{a}	205.64
N_3K_3	44.06^{ab}	32.26^{a}	36.33^{a}	18.55^{a}	77.41^{a}	208.61
CD @ 0.05	3.25	0.43	2.73	2.15	3.97	-

Table 25 Effect of N and K on phosphorus uptake by different plant parts at harvest (g/plant)

Treatment	Corm	Ps. stem	Leaf	Stalk	Fruit	Total
N_1K_1	3.95^{d}	1.61^{c}	2.06^{c}	2.89^{cd}	4.21^{b}	14.72
N_1K_2	4.23^{cd}	1.81^{c}	2.30^{bc}	2.77^{cd}	4.64^{b}	15.75
N_1K_3	4.48^{bc}	1.69^{c}	2.49^{c}	2.61^{d}	4.74^{b}	16.01
N_2K_1	4.60^{bc}	1.76^{c}	2.61^{b}	2.71^{d}	4.60^{b}	16.28
N_2K_2	5.50^{a}	2.76^{a}	3.35^{a}	3.46^{ab}	7.01^{a}	22.08
N_2K_3	5.43^{a}	2.50^{ab}	3.12^{a}	3.28^{abc}	7.50^{a}	21.83
N_3K_1	4.70^{b}	1.79^{c}	2.60^{b}	2.61^{b}	4.79^{b}	16.49
N_3K_2	5.45^{a}	2.45^{b}	3.50^{a}	3.14^{bcd}	6.89^{a}	21.43
N_3K_3	5.44^{a}	2.66^{ab}	3.46^{a}	3.75^{a}	7.09^{a}	22.40
CD @ 5%	0.44	0.30	0.42	0.57	0.73	-

Table 26 Effect of N and K on potash uptake in different plant parts at harvest (g/plant)

Treatment	Corm	Ps. stem	Leaf	Stalk	Fruit	Total
N_1K_1	60.57^{d}	60.75^{de}	21.76^{d}	35.98^{c}	102.56^{fg}	281.62
N_1K_2	71.22^{bc}	69.56^{bcd}	25.65^{cd}	40.67^{bc}	121.53^{de}	328.63
N_1K_3	74.33^{bc}	69.14^{bcd}	30.37^{bc}	43.76^{bc}	128.80^{d}	346.40
N_2K_1	64.33^{cd}	67.41^{cd}	26.03^{bcd}	39.04^{bc}	119.79^{de}	316.60
N_2K_2	84.73^{ab}	86.60^{a}	41.96^{a}	60.10^{a}	158.39^{a}	431.78
N_2K_3	87.17^{a}	85.83^{ab}	41.03^{a}	57.33^{a}	151.60^{ab}	422.96
N_3K_1	66.36^{cd}	69.49b^{cd}	27.77^{c}	44.91^{bc}	127.07^{d}	335.60
N_3K_2	87.26^{a}	85.15^{ab}	39.59^{ab}	63.95^{a}	161.10^{a}	437.05
N_3K_3	87.83^{a}	88.93^{a}	42.19^{a}	62.96^{a}	143.46^{bc}	425.37
CD @ 5%	9.86	8.90	7.44	14.92	11.03	-

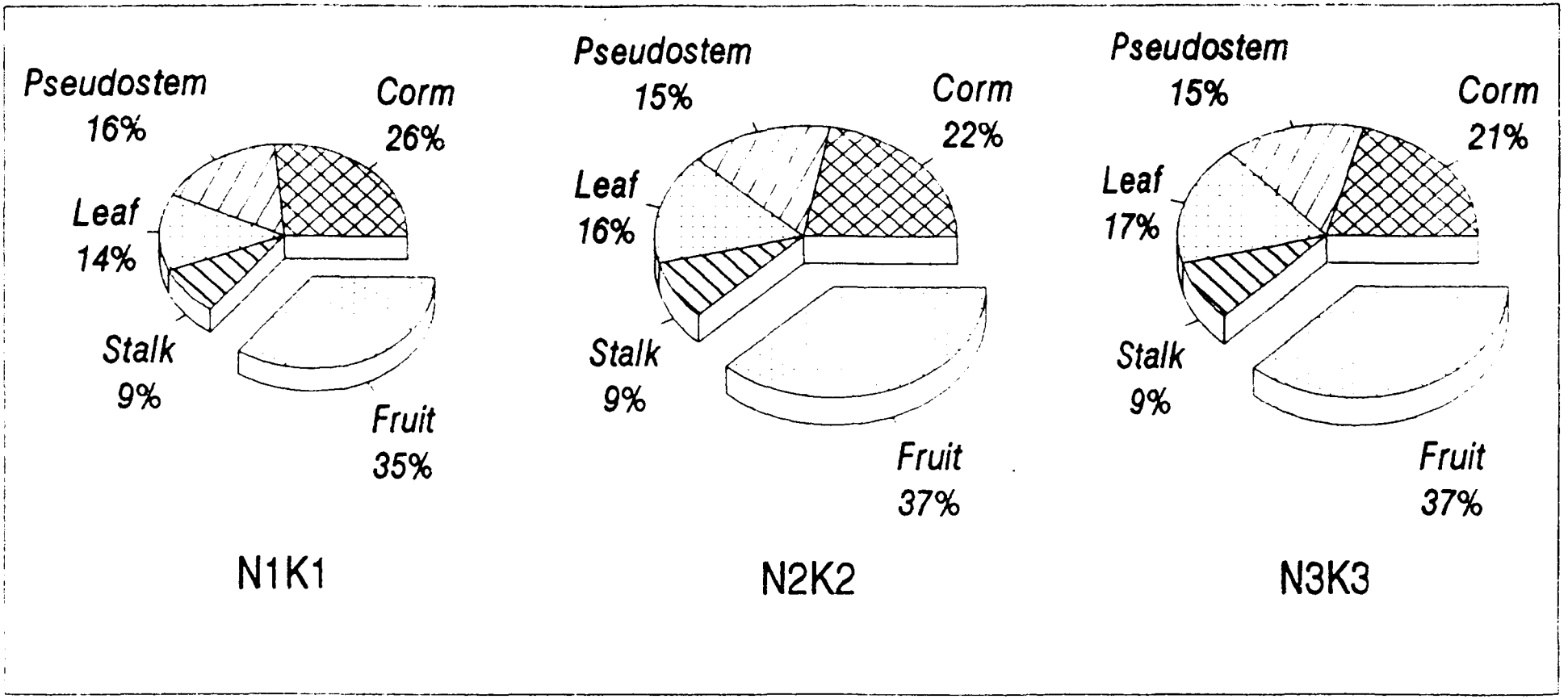

Fig 11a : Uptake of nitrogen in different plant parts at harvest

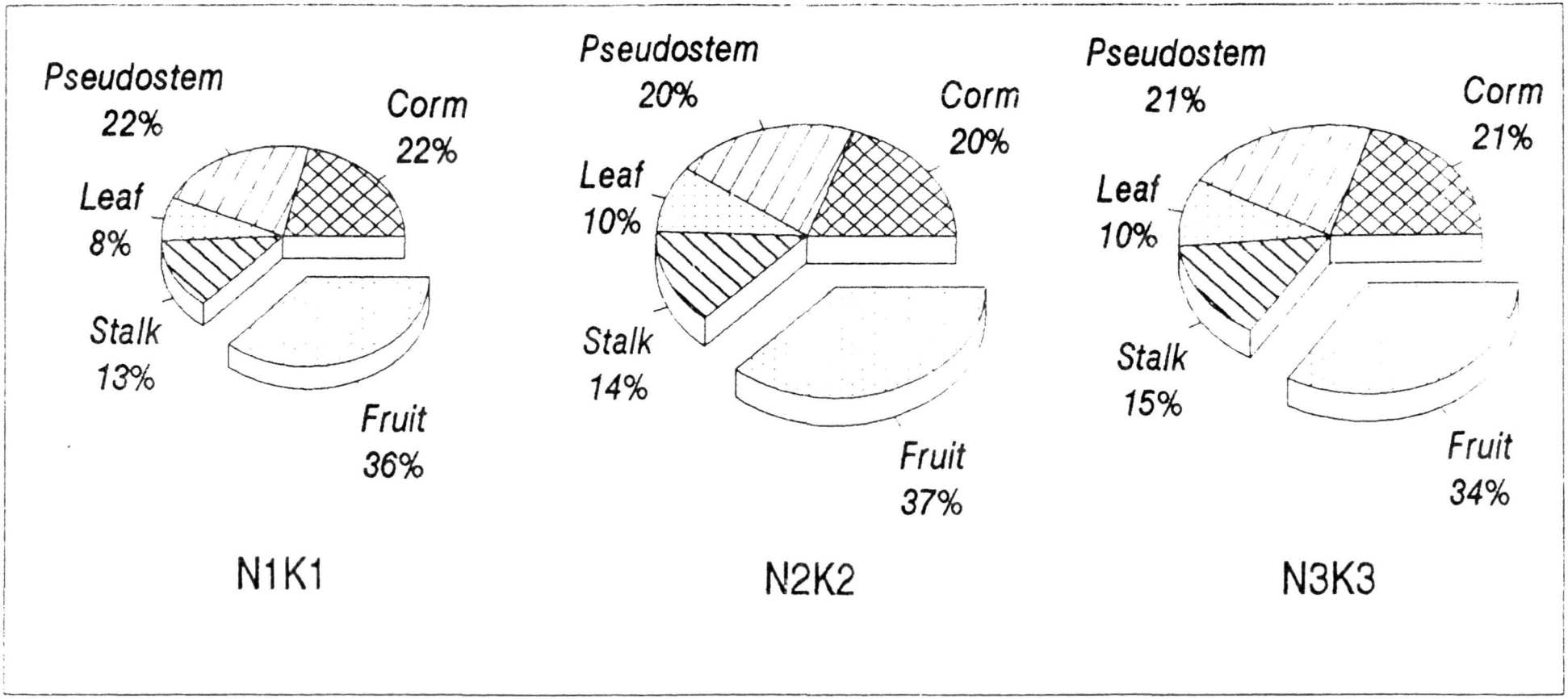

Fig 11b : Uptake of potash in different plant parts at harvest

nitrogen from 60 DAP till shooting and after shooting it declined in all the treatments. The available soil N at shooting ranged between 217.40 kg/ha (N_1K_2) to 237.77 kg/ha (N_3K_3) while at harvest the same was 171.07 kg/ha (N_1K_3) to 194.23 kg/ha (N_3K_3).

4.2.8.2 Available phosphorous

The available phosphorous in the soil did not differ due to various nutrient levels. From the data it can be observed that there was a little increase in available phosphorous in the soil from 60 DAP to 150 DAP and after 150 DAP till harvest a steady decline in small quantities.

4.2.8.3 Available potash

The available potash in the soil differed significantly between the treatment on all stages of crop growth except at 60 DAP. The available potash in the soil increased steadily from 60 DAP till 150 DAP and after that there was a decline during shooting at harvest. The maximum available potash in the soil was observed at 150 DAP in the treatment N_1K_3 (178.02 Kg/ha). While the minimum was observed in N_2K_2 at harvest (129.64).

4.2.9 Correlation studies

Correlation coefficients were computed to study the relationship between the yield (dependent variable) and the different independent variables, including growth parameters, flowering, maturity and uptake of nutrients (Table 30).

Table 27 Effect of nitrogen and potash on available nitrogen in soil (Kg/ha).

Treatment	60DAP	150DAP	Shooting	Harvest
N_1K_1	189.00^{ab}	213.46^{d}	219.56^{e}	172.28^{f}
N_1K_2	187.61^{b}	215.66^{cd}	217.40^{e}	174.56^{ef}
N_1K_3	190.72^{ab}	215.43^{cd}	218.75^{e}	171.07^{f}
N_2K_1	190.65^{ab}	219.46^{b}	223.72^{d}	180.50^{cd}
N_2K_2	190.72^{ab}	218.22^{bc}	220.47d^{e}	176.75^{de}
N_2K_3	191.64^{ab}	220.27^{b}	235.70^{ab}	183.35^{c}
N_3K_1	192.43^{ab}	224.46^{a}	231.05^{c}	169.50^{b}
N_3K_2	193.42^{ab}	225.61^{a}	233.67^{bc}	192.75^{ab}
N_3K_3	195.43^{a}	225.90^{a}	237.77^{a}	194.23^{a}
CD @ 5%	7.16	2.79	4.09	4.07

Table 28 Effect of nitrogen and potash on available phosphorus in soil (Kg/ha).

Treatment	60DAP	150DAP	Shooting	Harvest
N_1K_1	20.12	24.82	21.81	16.65
N_1K_2	21.62	23.75	20.23	15.47
N_1K_3	20.33	25.84	20.06	17.32
N_2K_1	20.75	27.66	22.98	17.65
N_2K_2	19.88	26.93	20.65	16.40
N_2K_3	20.92	25.14	21.75	18.00
N_3K_1	21.00	26.36	20.98	18.54
N_3K_2	20.50	29.45	21.70	18.04
N_3K_3	21.25	28.45	21.50	19.00
CD @ 5%	NS	NS	NS	NS

Table 29 Effect of nitrogen and potash on available potash in soil (Kg/ha)

Treatment	60DAP	150DAP	Shooting	Harvest
N_1K_1	140.75	173.32^{ab}	160.50^{c}	139.00^{ab}
N_1K_2	142.85	176.24^{a}	162.35^{c}	137.95^{ab}
N_1K_3	142.00	178.02^{a}	163.70^{bc}	138.06^{ab}
N_2K_1	141.25	174.50^{ab}	162.80^{c}	137.55^{ab}
N_2K_2	143.06	170.65^{b}	164.05^{bc}	129.64^{d}
N_2K_3	143.79	176.80^{a}	168.71^{ab}	135.11^{abc}
N_3K_1	140.72	174.35^{ab}	165.07^{bc}	139.53^{a}
N_3K_2	141.36	175.45^{a}	170.55^{a}	134.75^{bc}
N_3K_3	143.44	174.69^{ab}	171.69^{a}	131.22^{cd}
CD @ 5%	NS	4.75	5.24	4.75

Plant height, plant girth, total number of leaves produced, number of functional leaves and leaf area showed high positive correlation with yield. There was a high negative correlation between number of days taken to shooting, number of days from shooting to harvest and number of days from planting to harvest with yield.

The total uptake of nitrogen, phosphorus and potash by the plant also showed a significantly high positive correlation with yield.

4.2.9 Benefit-Cost ratio

The benefit- cost ratio differed significantly due to nutrient application (Table 31 & Fig.12). The treatment where received a nutrient level of N_2K_2 had a maximum benefit-cost ratio (6.10) which was on par N_3K_2 (6.04) and N_3K_3 (6.03). The minimum benefit-cost ratio was in the treatment N_1K_1 (4.76).

Table 30 Relation between growth parameters, flowering, maturity and nutrient uptake on yield of banana

Sl No.	Characters	r value
1.	Plant height	0.815
2.	Plant girth	0.945
3.	Functional leaves	0.942
4.	Leaf area	0.969
5.	Total number of leaves	0.930
6.	Days to shooting	- 0.972
7.	Shooting to harvest	- 0.967
8.	Planting to harvest	- 0.971
9.	Total nitrogen uptake	0.976
10.	Total phosphorus uptake	0.887
11.	Total potash uptake	0.925

Table 31 Effect of nitrogen and potash on benefit - cost ratio

Treatment	Benefit - cost ratio
N_1K_1	4.76
N_1K_2	4.93
N_1K_3	4.93
N_2K_1	5.46
N_2K_2	6.10
N_2K_3	5.72
N_3K_1	5.53
N_3K_2	6.04
N_3K_3	6.03

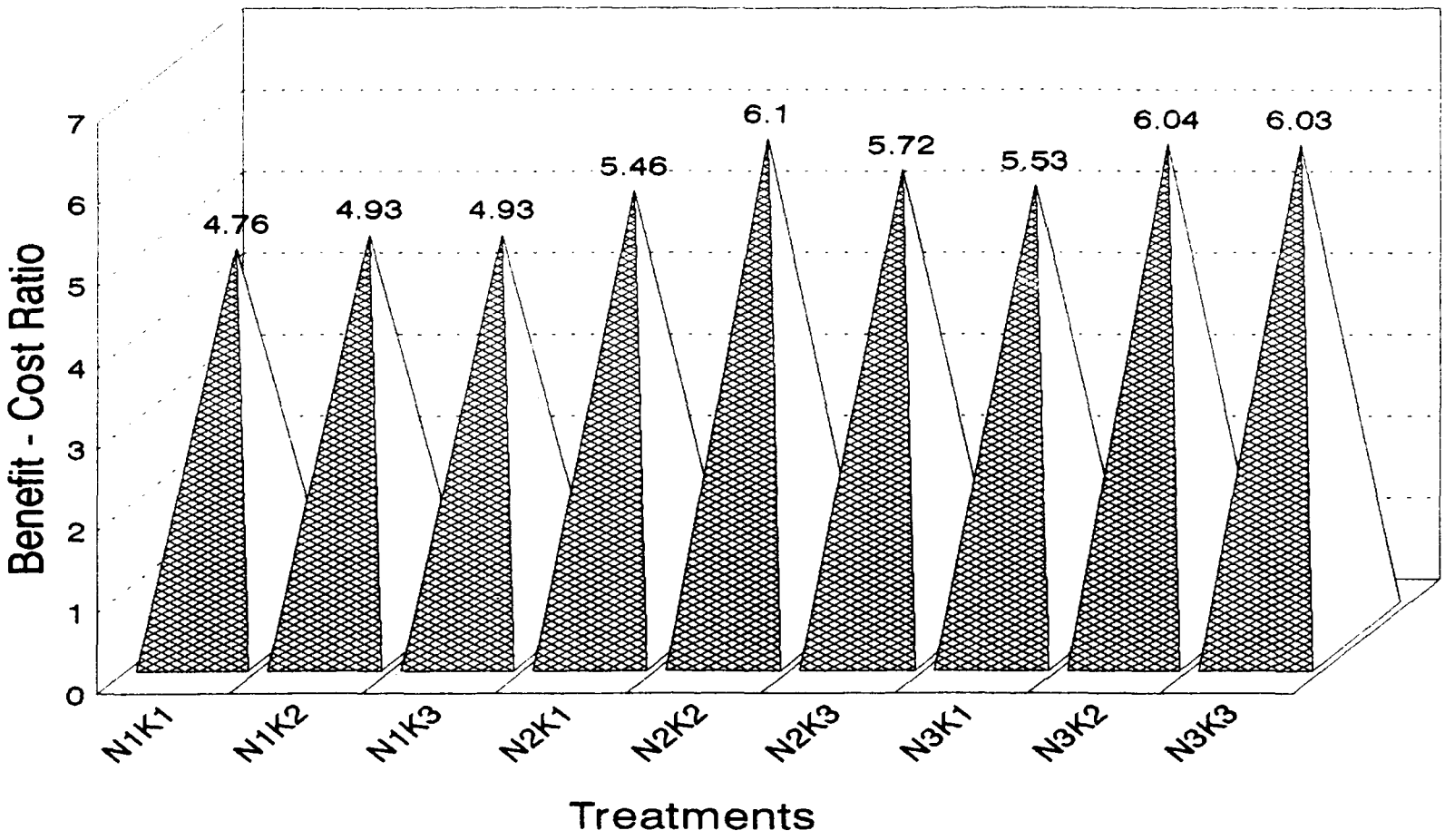

Fig. 12 Effect of nitrogen and potash on benefit-cost ratio

DISCUSSION

V DISCUSSION

Banana is one of man's oldest and most valued tropical fruit crops. The importance of both cooking and dessert banana in socio-economics of the countries in tropical and subtropical regions cannot be overstated. They often play a vital role in human and animal nutrition. Additionally banana has an importance for tannin, latex, alcohol and fibre production. Banana plants possess anti-ulcer properties as well (Cronauer and Krikorian, 1986).

Tissue culture studies

Edible banana does not form or produce seeds and is conventionally propagated vegetatively through suckers. Each mother plant produces five to ten sword suckers at the time of harvest (14 to 18 months). Thus, planting material is in great demand, as most of the growers replant their orchards every season to adjust the harvesting time for better profitability and for export. It is risky to use suckers obtained from the field because they act as a source of inoculum for many important diseases, and it becomes increasingly difficult to obtain large number of disease free suckers. A meristem culture technique first reported by Ma and Shii (1972) was employed to induce the formation of adventitious buds from the decapitated shoot apex of a sucker. This was consequently developed for mass propagation of disease free plantlets on a commercial scale (Hwang *et al.*, 1984). Micropropagated plantlets are convenient to handle, with higher survival rate, more uniformity in growth, and produce higher yields than

suckers. Moreover, the harvesting period is shortened by four to five months (Wu and Su, 1990). Currently, 180 to 200 million plants are produced world wide every year by tissue and cell culture techniques. Conventional plant production has been replaced by large scale micropropagation, mainly aiming for higher production, elimination of pathogens, rapid multiplication of new genotypes, selection of somaclonal and induced variants, preservation of germplasm and for long term storage (Reuther, 1990). The importance of tissue culture in banana is so much that in Taiwan alone, in the last five years, more than 10 million plantlets have been produced.

In the present study, influence of explant size indicated that both the small sized (2-3 mm) and medium sized (4-5 mm) explants performed better in terms of *in vitro* establishment in liquid MS media supplemented with BAP and adenine sulphate compared to big sized explants (6-7 mm). The small sized explants recorded an establishment of 78.50 per cent while, medium and big sized explants recorded 81.26 and 69.25 per cent respectively. Further, it was observed that the small sized explants had lesser microbial contamination compared to medium and big sized explants. They also responded better to plant growth substances (cytokinins) which resulted in better percentage establishment. These results are in conformation with the earlier studies by Murashige (1977), Doreswamy *et al.* (1983) and Vuylsteke and De Langhe (1985) who also reported that the probability of recovering virus free plants even from infected material is greater with smaller explants and that they responded better to minerals and cytokinins.

Murashige and Skoog's basal media is the most widely used of all plant media for shoot and single node culture, cells and suspension culture, direct and indirect shoot formation and embryogenesis. In this study, a combination of various concentrations of BAP and adenine sulphate were supplemented to MS media to assess their influence on establishment. The MS basal media supplemented with BAP at 40 µM and adenine sulphate at 200 µM were highly effective in establishment of explants both in liquid (78.50 %) and in semi-solid (89.50 %) media followed by BAP at 40 µM and adenine sulphate at 300 µM (71.25 and 85.25 % respectively). It may be attributed to the effect of cytokinins on plant cell division inhibiting apical dominance and there by promoting direct or indirect rapid proliferation and shoot initiation. The earlier reports of Banerjee and De Langhe (1985), Jarret *et al.* (1985b) and Gupta (1986) have also indicated that cytokinins with MS media stimulated proliferation resulting in better establishment.

Established shoot-tip cultures were utilized as a source of explants for further development of multiple shoots. The small sized shoot tip explants incubated for a further eight weeks resulted in a higher development of multiple shoots (18.25) when the MS media was supplemented with BAP at 40 µM and adenine sulphate at 200 µM followed by BAP at 40 µM and adenine sulphate at 300 µM (16.10). Further, MS media supplemented with BAP at 40 µM and adenine sulphate at 200 µM resulted in the maximum height of pseudostem (5.72 cm), while BAP at 40 µM and adenine sulphate at 300 µM resulted in more number of leaves (4.20). This may be due to stimulation of axillary branching/adventitious bud formation and the high rate of proliferation by cytokinins. Further, wounding the plants at the tip may have

reduced/eliminated the apical dominance thereby resulting in a high number of multiple shoots. These findings are in conformity with earlier works by Vessey and Rivera (1981), Damasco and Barba (1985) and Jarret *et al.* (1985a) who reported multiple shoot development ranging from 3 to 31 in different genomes of banana due to the effect of cytokinins.

The elongated shoots from the previous stage were cultured on an MS media supplemented with different sources of auxins like IAA, IBA and NAA to study their influence on *in vitro* root production. Among the different auxins tried, MS basal media supplemented with NAA produced higher number of roots compared to IBA or IAA. The maximum number of roots (7.66) was recorded in the media supplemented with NAA at 10 µM followed by IBA at 5 µM (7.00). IAA as a source of auxin did not induce *in vitro* root production effectively. The *in vitro* root production due to auxins may be because of promotion of polyamine synthesis. Moreover, banana being monocotyledonous in nature, produces adventitious roots more easily. The results of the present investigation are in conformity with the earlier reports by Doreswamy *et al.* (1983), Vuylsteke and De Langhe (1985) and Banerjee and De Langhe (1985).

The *in vitro* rooted banana plantlets were hardened inside a green house to acclimatize the plantlets to outside environmental conditions for better establishment in the field later. The plantlets were exposed to a temperature of 25 - 27° C and a relative humidity of 60 to 70 per cent inside the green house. Three different media (Perlite, soil and sand) and their combinations were tried to determine the best media for establishment of banana plantlets. The highest survival percentage of banana plantlets (96.50) was achieved when a media of perlite, soil and sand was used in the ratio of

2:1:1. Similar results were obtained when other combinations of perlite, sand and soil and perlite alone were used. This may be due to better aeration in the root zone as compared to soil, which may also be one of the reasons for comparatively lower establishment of plantlets in soil media. These results are in conformity with the earlier reports by Cronauer and Krikorian (1984a), Damasco and Barba (1985) and Rodriguez *et al.* (1987).

Nutritional studies

Banana requires high amounts of nutrients for good growth and production. It exhausts large quantities of nutrients from the soil during its growth, and these have to be replenished, to maintain the soil fertility for sustained production of high yields, by applying organic, and more efficiently, by application of mineral fertilizers which supply nutrients in concentrated and readily available form. This requirement of nutrients in banana is further enhanced by intense leaching of nutrients and of mobile nutrients mineralized from plant residues.

Among the major nutrients, nitrogen largely controls the growth and fruiting of most of the crop species. High mobility of nitrogen in the soil necessitates its timely replenishment to correspond with critical phases of growth. The yield increase owing to the contribution of nitrogen in fertilizers has been very well documented (Simmonds, 1980; Rajeevan and Mohan Kumaran, 1990; Reddy, 1992).

Banana consumes the highest amount of potash when compared to other major nutrients, as it plays a vital role in almost all the metabolic processes. The time of application as well as the stage of plant growth determines the uptake and translocation

of potash (Cuddus and Bunemann, 1974). Potassium supply in early vegetative stage has greater influence on yield of banana (Summerville, 1944, Alexandrowicze, 1955, Twyford, 1965). Hence both nitrogen and potash, the two major nutrients, play a vital role in growth and productivity of banana.

Further, the tissue cultured banana plants with well developed root system at the time of planting compared to bananas raised from suckers have the ability to utilize nutrients from the very early stages of crop growth. As fertilizers constitutes a considerable part of the input costs, efficient use of the nutrients play a greater role in reducing the cost of banana production. Thus, the present study was undertaken to standardize the optimum level of two major nutrients (nitrogen and potash) in tissue cultured banana Cv, Grande Naine. The results of present investigation are discussed here under.

Growth

Plant height and girth

In banana, height and girth of pseudostem are important parameters to judge the vigor. In the present study there was a significant influence on plant height and girth due to application of high levels of nitrogen (225 to 300 g) and potash (300 to 400 g). The uptake of the nutrients, more specifically of nitrogen and potash, ultimately leads to the formation of complex nitrogenous substances such as proteins, aminoacids, etc. to build up new tissue. Nitrogen is the chief constituent of chlorophyll, where synthesis of proteins and amino acids is accelerated through increased supply of nitrogen (Pafli, 1965).

Potassium ions act as general metabolic activators, increasing the respiration rate and photosynthetic potential of leaf (Martin-Prevel, 1982). The requirement of nutrients is especially high between 90 and 120 days of crop growth as evidenced from the fact that the response to fertilizers during this period was significant. It may be due to triggering of initiation of vegetative meristem to generative tissue formation, which subsequently differentiates into floral primordium that determines the further size of the bunch. Hence, more nutrients are required during this period . This is in conformation with earlier reports that nitrogen and potash, increase the pseudostem height and plant growth (Arunachalam, 1972; Kohli *et al.*, 1985; Bellie 1987; Parida *et al.*, 1994; Natesh *et al.*, 1993). The study also revealed a significant positive correlation between pseudostem height and girth with yield which obviously indicated the necessity for short pre-shooting vegetative growth. Similar relationships between plant height, girth and yield in banana have been reported by several workers (Crouchuer and Mitchell, 1940, Warner *et al.*, 1974, Krishnan and Shanmugavelu, 1983).

Leaves

The number of functional leaves is a good index for the nutritional status in banana and the rate of production of leaves is influenced by mineral nutrition (Murray, 1960), and stage of plant growth (Champion, 1970). Nitrogen and potash at high levels resulted in a higher number of functional leaves, increased leaf area, higher leaf area index, total number of leaves and lower rate of phyllachron.

Increased production of leaves might help to elaborate more photosynthates and flowering stimulus thus influencing early flowering, as there was maximum leaf number

and leaf area at shooting stage. The effective leaf area available for photosynthetic activity might have influenced the development of fruits, and in turn, the gross yield (Venkatesam *et al.*, 1965). Kothavade *et al.*, 1985) observed that as the number of leaves per plant increased, the weight of bunch progressively increased. Low amount of nitrogen and potash decreased the longevity of leaves. Reduced longevity may be due to the high mobility of K + ions from older to younger leaves (Lahav, 1972; Mustaffa 1987; Baruah and Mohan, 1991,). The leaf area at flowering was found to be positively correlated with bunch weight corroborating the findings of Croucher and Mitchell (1940), Turner (1970), and Kothavade *et al.* (1985). The lowest phyllachron at higher levels of nitrogen and potash indicates that higher rates of nitrogen and potash promote faster rate of leaf production which lends support to the findings of Baruah and Mohan (1992) and Roy *et al.* (1993). The number of functional leaves, leaf area index and faster rate of leaf production between 90 to 120 DAP of the crop might have influenced increased uptake and distribution of nutrients for better growth and development. The lower level of nitrogen, reduces the rate of leaf production by more than 50 per cent, resulting in lower yield (Martin-Prevel and Montagut, 1966).

Crop duration

The time taken for shooting and subsequently for maturation and harvest varied with the application of nutrients. Plants supplied with nitrogen (225 to 300 g) and potash (300 to 400 g) reduced the time taken for shooting, shooting to harvest and planting to harvest by 14-20 , 11-15 and 25-35 days, respectively. Owing to earlier production of leaves with larger leaf area per plant and better disposition of

photosynthetic activity, the required net assimilation presumably was reached early in plants receiving higher dose of nitrogen, hastening the process of initiation and emergence of inflorescence. (Parida *et al.*, 1994). A higher potassium content in the plant tissue could exert an effect on inhibition factors eventually suppressing leaf primordial growth and activating flower bud initiation. During the transition stage, there is higher production and utilization of proteins and ascorbic acid, resulting in a higher metabolic state in the cell and producing DNA and RNA at a faster rate, thus resulting in differentiation (Evans, 1971 and Vadivel, 1976). Potassium is a general metabolic activator increasing the respiration and photosynthetic rate; a higher level of potash application thus enhanced flowering and faster development of bunches. Other earlier studies (Martin-Prevel, 1962; Israeli and Lahav, 1986; Singh *et al.*, 1990) have also reported that an optimum supply of nutrients stimulated early shooting and shortened the number of days to fruit maturity. Further, they observed that when nutrients were applied in lower quantity, maturity was delayed. In the present study, it was observed that there was an increase in all the growth parameters, when plants were supplied with an optimum dose of fertilizer during the cropping period between 90 to 150 days after planting, which resulted in early flowering and maturity. This is in confirmation with earlier reports (Kohli *et al.* 1985, Chattopadhyay and Bose 1986, Hegde and Srinivas, 1991).

Yield and yield parameter

Application of fertilizers, especially the two major mineral nutrients nitrogen and potash, exerted a positive influence on yield and yield attributes such as number of

hands/bunch, number of fingers/bunch and average weight of finger. All the parameters were significantly affected by the application of different levels of nitrogen and potash.

The highest yield (150.77 t/ha) was obtained when the plants were supplied with 300 g and 400 g of nitrogen and potash, respectively. The yield was on par (145.17 t/ha) with the treatment, in which nitrogen and potash were applied at the rate of 225 and 300 g/plant, respectively. This increase in yield was also associated with a corresponding increase in the number of hands/bunch, number fingers/bunch and average weight of finger. The increase in yield could be due to the observed increase in plant height, girth, number of functional leaves, leaf area index and faster rate of leaf production and also higher nutrient uptake by the plants. This is in confirmation with the findings of Bowman and Eastwood (1940), Ho (1969), Jumbulingam (1971), Rodriguez (1980), Pathak *et al.* (1992) and Roy *et al.*(1993). The low yields at lower level of nitrogen and potash was probably due to the low uptake of nutrients and consequent low dry matter production, since potassium deficiency is known to restrict fruit growth by reducing the translocation of carbon compounds from leaves to the fruits and their conversion to starch (Martin-Prevel 1973).

Martin-Prevel and Montagut (1966) also reported that lower nitrogen uptake retards the total dry matter production. Similarly, in the present study, lower nutrient levels reduced leaf size, delayed flower initiation, reduced fruit number/bunch, number of hands/bunch and fruit size. The findings are also supported by the reports of Lahav (1972), Martin-Prevel and Charpentier (1963a) and Murray (1959). The increased trend in dry matter at harvest due to application fertilizers might have contributed for

higher number of fingers/bunch, higher average weight of fingers, more number of hands/bunch resulting in increased yield which may be due to timely availability of required amount of nutrients during the flower bud initiation. These are in conformity with the findings of Garita and Jaramillo, (1984), Pathak *et al.* (1992), Pandit *et al.* (1992) and Natesh *et al.* (1993).

Fruit quality

A marked effect on fruit quality was observed with the application of adequate amounts of nutrients. A high TSS, high per cent of reducing and total sugars, high sugar-acid ratio but a lower acidity were obtained by application of nitrogen and potash at 300 and 400 g/plant, respectively. Similar results were obtained when plants received nitrogen at 225 g and potash at 300 g. Ram and Prasad (1989) reported that high levels of TSS (21.21°) with application of 200 g of nitrogen. The reduction in acidity when 200 g of nitrogen was given, could be due to neutralization of organic acids with increased potassium levels in the tissues and the increase in sugars could be due to respirational demand and adequate supply of nutrients, synthesis of invertase and starch splitting enzymes as suggested by Singh *et. al.* (1973), Vadivel and Shanmugavelu (1978) and Mustaffa, (1988). These effects of potassium could also be due to its involvement in synthesis of carbohydrates, break down and translocation of starch, synthesis of proteins and neutralization of physiologically important organic acids (Greenberg and Preiss, 1965; Tisdale and Nelson , 1966; Akatesula and Nelson, 1966).

With low nutrient levels, the translocation of photosynthates to the bunch was probably reduced by lower rates of photosynthesis. Potash application also improved the storage life by reducing the incidence of peel split which lends support to the results of Baruah and Mohan (1986). The pulp to peel ratio increased with increasing level of nutrients and improved the keeping quality of fruits. The results are in confirmation with findings of Venkatarayappa *et al.* (1978).

Dry matter production

The biomass accumulation and its distribution in various plant parts plays a vital role in determining production in banana. Biomass accumulation and distribution was maximum with higher levels of nitrogen and potash (N_2, N_3, K_2 and K_3). The higher accumulation and distribution of dry matter in these treatments indicate that there was a better uptake and translocation of nutrients, which is reflected in higher bunch weights. The higher dry matter production in these treatments may be due to better plant growth, more number of functional leaves, more leaf area and better uptake of nutrients from the soil. These findings confirm the reports of Kohli *et al.* (1985) and Turner and Barkus (1981). Murray (1961) observed that a low level of potassium restricted the growth of leaves resulting in smaller leaves thus affecting the leaf area and photosynthetic efficiency, thereby reducing the total dry matter production of the plant. As indicated by Kohli *et al.* (1985), application of higher levels of nutrients resulted in a diversion of biomass distribution towards leaf, rachis, flower buds and fingers indicating a high yield potential compared to plants receiving low level of nutrients, where the biomass accumulation was more in rhizome and pseudostem.

Soil Nutrient status

Available Nitrogen

Available soil nitrogen exhibited an increasing trend during crop growth till shooting, and the maximum available soil nitrogen was found between 60 to 150 DAP in treatments where higher quantity of fertilizers were applied. This may be due to better availability of added fertilizers. Further, banana being a heavy feeder, has utilized nitrogen and potash to a higher degree as indicated from the leaf nitrogen and potassium contents (Hewitt and Osborne, 1962; Lahav, 1972; Baruah and Mohan, 1986). The low amount of available nitrogen in the soil in the treatment combination of N_1K_1 may be due to poor nitrogen availability possibly because a portion of nitrogen applied might not have been utilized by the plant due to soil factors. Nitrogen content and availability are chelated by a number of soil factors. Addition of nitrogen has always been found to have a positive influence on crop growth as seen from the experiment that higher nitrogen application had additive influence on growth parameters (Minhas and Borah, 1982).

Available Phosphorus

The availability of phosphorous in the soil is influenced by pH. In the soils of the experimental plots studied, it was observed that there was a steady decrease in the available soil phosphorous throughout the crop growth period except at 150 DAP. This may be due to continuous uptake of phosphorous in smaller quantities by the plants. The available soil phosphorous in the experimental plots increased with higher levels of nitrogen and potash corroborating the findings of Srikhande and Yadav (1954), who

reported that for uptake of phosphorus, nitrogen and potash were necessary. Another factor could be due to solubilization of phosphorus, making it available to the plant.

Available potash

In all the treatments there was an increase in the available potash upto shooting, but declined later. The decreasing trend during the later stage of the crop may be due to higher requirement and uptake of potash by banana crop as suggested by Simmonds (1987). The high amount of potash in the soil may also be due to initial higher potassium content of the soil. The decreasing trend in the available soil potash after shooting confirms that potash is required in higher amounts during floral differentiation and fruit development stage resulting in higher yields and high quality fruits. The decrease in soil potash at harvest may be due to higher amounts of potash being absorbed and utilized for development and improvement of size and quality of bunch, and also for development of corms, because mobility of potash towards sink can be significant (Montagut and Martin-Prevel, 1965; Balakrishnan, 1980; Obiefuna, 1984).

Nutrient uptake

The use of nutrient concentrations in diagnosis of nutrient deficiencies is usually based on an assumed relationship between concentration and crop performance. Plant analysis is not only helpful in diagnosing of toxicity or deficiency, but it gives useful information on absorption of nutrients (Turner, 1979). In the present study an increase in uptake of all nutrients in different plant parts was observed till shooting, at higher levels of nitrogen and potash. The uptake of potash was maximum and all the nutrients

decreased after shooting. The higher uptake of nutrients may be due to their higher availability. The uptake of potash and nitrogen were observed to be positively correlated to the bunch weight. The results are in conformation with the findings of Buragohain and Shanmugavelu (1980). Twyford and Walmsky (1974) who conducted exhaustive studies on nutrient uptake of Robusta revealed that a high yield was very much associated with high uptake of potash than the uptake of other nutrients. Turner (1985) reported that a fully grown banana contained more potash than any other mineral element; indeed the amount of potash was slightly greater than the amount of all minerals combined, which was also observed in the present study. The robust and vigorous growth of tissue cultured plants may also have contributed significantly for the high level of nutrient uptake.

Benefit cost ratio

Efficacy of the different nutrient treatments was worked out by computing the benefit - cost ratio. The benefit - cost ratio of 6.10 was obtained in the treatment which received nitrogen at 225 g and potash at 300 g/plant. The cost of fertilizer and its application was very meager compared to the profit obtained.

SUMMARY

VI SUMMARY

The present study was conducted to develop a suitable protocol for micropropagation of banana cv Grande Naine and to standardize the optimum fertilizer requirements, especially the two major elements, nitrogen and potash, for better growth and yield of banana raised through tissue culture. The present micropropagation studies were carried out at the Plant Tissue Culture Laboratory of the Division of Horticulture, University of Agricultural Sciences, Bangalore, while field studies on nutritional requirement were conducted at Rashmi farm near Channasandra village in Bangalore South taluk.

The size of explant had a positive influence on the initial *in vitro* establishment in liquid media. Both small sized (2-3mm) and medium sized (4-5mm) explants were found to be the best material for initial establishment. Nevertheless, smaller sized explants were preferred on account of higher probability of recovering disease free plants.

The MS media supplemented with BAP at 40 µM and adenine sulphate at 200 µM was found to be the best culture media for both initial establishment (78.5%) in liquid media and for further establishment (89.5%) on semi-solid media. The addition of cytokinins to MS basal media helped in better establishment of explants.

The MS media supplemented with BAP at 40 µM and adenine sulphate at 200 µM was found to be more ideal for further subculturing, as it resulted in higher number of

multiple shoots (18.25), maximum height of pseudostem (5.72) and higher number of leaves (3.96) under *in vitro* conditions.

3-Naphthalene Acetic Acid produced higher number of roots (7.6)in *in vitro* developed plantlets at a concentration of 5 µM compared to IBA and IAA. The response of *in vitro* developed plantlets to IBA with respect to rooting (7.0) was similar to that of NAA.

The initial hardening of *in vitro* plantlets in the green house with perlite (90.40 %) and combinations of perlite, soil and sand in the ratio of 2:1:2, 1:1:1 and 2:1:1 resulted in better per cent survival (96.50, 93.20 and 93.00, respectively).

Banana cv. Grande Naine can be effectively propagated rapidly on a large scale by tissue culture using the above procedures to obtain disease free and vigorous plantlets.

There was a positive response to plant growth in terms of height and girth to nutrient application. nitrogen at 225 g and 300 g with potash at 300 and 400g resulted in better plant growth.

Higher number of functional leaves with higher LA and LAI was recorded with the application of nitrogen at 225 g and 300 g of potash. This was on par with nitrogen applied at the rate of 300 g with 400 g potash.

The interval between production of successive leaves was shorter and the total number of leaves produced during the crop growth was higher when nitrogen was supplied at 225 g or 300 g in combination with higher levels of potash (300 or 400 g/pl).

The initiation of flower was earlier by 20 days when plants received higher levels of nitrogen (225 g and 300 g) and potassium (300 g and 400 g). Further, the fruits matured 15 days earlier after shooting thus reducing the total crop duration from planting to harvesting by 35 days.

Uniform flowering was observed in tissue cultured plants which is a rare phenomenon in banana crop raised through suckers. A maximum bunch weight of 49.84 kg accounted for an yield of 157.77 t/ha at the highest levels of nitrogen and potash application. An economic yield of 144.17 t/ha with better harvest index was obtained when plants were supplied with 225 g and 300 g of nitrogen and potash respectively. The higher yield was attributed to higher number of hands/bunch, number of fingers/bunch and higher average weight of finger.

The dry matter accumulation and distribution gave positive response at higher levels of nutrient application giving vigorous and better growth, which was reflected in maximum bunch weight. An increased trend in the uptake of N,P & K was observed at higher levels throughout the crop growth in all parts of the plant. The nutrients applied were utilized efficiently which was reflected in better dry matter production and distribution.

The available soil NPK was higher with higher levels of nutrients upto flowering and decreased later on; this indicated that a larger portion of the nutrients were utilized during the later part of the growth, i.e from flower initiation to bunch production.

The finer aspect of the study revealed that better quality fruits having higher TSS, reducing sugars, total sugars with low acidity and better sugar/acid blend were obtained at higher level of nutrients (225 g or 300 g N with 300 g or 400 g of K per plant).

The other interesting finding from this study is the uniform flowering in tissue cultured banana which facilitates synchronous harvesting for easy transportation and marketing. From the storage point of view fruits could be stored for a longer period without much damage to the fruit an account of better peel-pulp ratio under ordinary conditions compared to fruits obtained from lower levels of nutrients.

An economical yield of 144.17 t/ha was recorded by providing 680 kg of nitrogen with 900 kg of potash/ha (225 g of N with 300 g of K/plant) which works out to a benefit-cost ratio of 6.10:1.0.

Future line of work

1. Standardization of series of subculturing to maintain true to type characters.
2. To study the effect of repeated sub-culturing on somaclonal variations to obtain disease resistant varieties.
3. More experimentation needs to be carried out with reference to crop management and crop production aspects with special reference to planting age, scheduling and water management of tissue cultured plants.

REFERENCES

VII REFERENCES

*Akatesula,T. and O.E.Nelson, 1966. Granule-bound adenosine diphosphate glucose starch glycosyl transferase of maize seeds. J. Biol Chem., 41: 228-2286.

*Alexandrowicze, L., 1955. Etude du development de l'inflorescence du bananier main. IFAC Annales, 9:32. Cited from Ram,M., Ra, Ram Manasi and F.C.Steward. 1962. Ann. Bot., 26: 657-672.

Anonymous, 1992. FAO Production Year book. Vol.46, pp 181-182.

Anjorin, H.O and G.O. Obligbesan, 1984. Influence of nitrogen fertilization on the early growth and development of plantain (***Musa paradisiaca***) Fruits, **38(4)**: 300-302

A.O.A.C. , 1960. **Official methods of analysis.** Published by A.O.A.C., Washington, D.C.

*Arunachalam,A., 1972. Studies on the effect of nitrogen on Cavendish clones (*M. cavendishii* Lambert). M.Sc.(Ag) Thesis submitted to Annamalai University, Annamalainagar.

Arunachalam,A., N. Ramaswamy, and C.R. Muthukrishnan, 1976. Studies on the nutrient concentration in leaf tissue and fruit yield with nitrogen level for cavendish clones. Prog. Hort., 8(2):13-22.

Bailon,A.F., E.Holmes and A.H.Lewis., 1933. The composition of and nutrient uptake by the banana plant with special reference to the Canaries Trop. Agric., 10: 139-144.

*Bakry F., 1985. Choix du material a utilizer pour l'isolement protoplastes de bananier(*Musa* sp.) *Musaceaes*. Fruits: 39: 449-449.

Balakrishnan, R., 1980. Studies on growth, development, sucker production and nutrient uptake at different ploidy levels in banana (*Musa* sp.) Ph.D. thesis submitted to TNAU., Coimbatore.

Banerjee N. and E.A.L. De Langhe, 1985. A tissue techniques for rapid clonal propagation and storage under minimal growth conditions of *Musa* (Banana and plantain). Plant Cell Report 4:351-354.

Banerjee N. Schoofs; S.Hollevoet and E.A.L. De Langhe, 1986a. Aspects and prospects of somatic embryogenesis in *Musa* ABB (Bluggoe). In Boxus p. (Ed.) *In vitro* problems related to mass propagation in horticultural plants. **Acta Horticulturae**, 212: 727-730

Banerjee N., D. Vuylsteke and E.A.L. De Langhe, 1986b. Meristem culture of *Musa*: Histomorphlogical studies in shoot bud proliferation. In Whithers, L.A. and P.G.Alderson. **Plant tissue culture and its applications Butterworth,** London pp. 139-147

Baruah,P.J. and N.K.Mohan, 1985. Effect of potassium on vegetative growth of banana (*Musa* AAA Cavendish Subgroup Jahaji). **Banana News Letter, 8** : 23-24.

Baruah,P.J. and N.K. Mohan, 1986. Effect of potassium on fruit sugar, acidity, TSS and sugar : acid ratio of `Jahaji' banana (*Musa*, AAA, Cavendish subgroup). **Banana News Letter, 9** : 14-15.

Baruah,P.J. and N.K.Mohan, 1991. Effect of potassium on leaf area index, phyllochron and number of leaves of banana (*Musa* AAA group, Cavendish subgroup`Jahaji'). **Banana News Letter, 14**: 21-22.

Baruah,P.J. and N.K.Mohan, 1992. Effect of potassium on yield and yield attributing characters of Dwarf Cavendish (*Musa* AAA group, Cavendish Subgroup) in Assam. **Banana News Letter, 15**: 24-25.

*Battikhah,G. and R.Khalidy, 1962. Effect of three levels of nitrogen on the inorganic composition and growth of the banana. **Publ. Pac. Agr. Sci. Amer. Univ. Beirut, 17**: 1-15.

Bellie, T., 1987. Studies on the effect of increased dose of phosphorus and split application of fertilizers to Nendran banana (AAB). M.Sc.(Hort.) Thesis submitted to TNAU, Coimbatore.

*Berg L.A. and M. Bustamante, 1974. Heat treatment and meristem culture for the production of virus free banana. **Phytopathology, 64**: 320-322.

*Bhan, K.C. and P.K. Majumdar, 1956. Manurial investigations with banana (Martaman variety) in West Bengal. **Indian J. Agric. Sci., 26**(4) : 337-350.

*Bhangoo,M.S., F.G.Altman and M.L.Karon, 1962. Investigations on the Giant Cavendish banana-I. **Trop. Agric. Trin., 39**(3): 189-201.

Bhavanisanker,K., 1980. Nutritional investigation in Nendran banana (French plantain - AAB) M.Sc.(Agri) Thesis submitted to TNAU, Coimbatore.

*Bowman,F.T. and H.W.Eastwood, 1940. Banana fertilizer experiments. Agric. **Gaz. N.S.W., 15** : 572-573.

Brzesowsky,W.J. and J.Van Biesen, 1962. Foliar analysis in experimentally grown Lacatan bananas in relation to leaf production and bunch weight. **Neth. J. Agric. Sci. 10**: 118-126.

Buragohain,R. and K.G.Shanmugavelu, 1986. Studies on the nutrient content and uptake of the `Vayal Vazhai' banana (ABB). **Banana News Letters.** 9 : 19-23.

*Burries, R.H., 1957. Nitrogen nutrition **Pl. Physiol. 10**:301-308

*Butler,A.F., 1960. Fertilizer experiments with the Gros Michel banana. **Trop. Agric. Trin., 37** : 31-50.

Chadha,K.L., 1995. **The ninth biennial group discussion of the All India Coordinated Research Project on tropical Fruits,** held at PAU, Ludhiana from Nov. 27-30th, 1995.

Chakravarthy,S.P., 1979. Effect of crop sequence on the physical properties and mineralizable nitrogen of soil **J. Indian Soc. Soil Sci. 27**: 184-185.

*Champion,J., 1970. Plantation growing in puerto Rico. **Fruits, 25**: 369.

*Champion,J., F.Duganna, R.Maignien and Y.Dommergues., 1958. Lessols de bananeraies et lew amelnoration en Guinee. **Fruits, 13**: 415-462.

*Charpentier,J.M. and P.Martin-Prevel., 1965. Major and minornutrient deficiencies in bananas. **Fruits,** 20: 521-557.

Chattopadhyay,P.K. and T.K.Bose., 1986. Effect of NPK nutrition on growth, yield and quality of dwarf cavendish banana (*Musa cavendishii* Lamb). **Indian Agriculturist, 30** : 213-222.

Chattopadhyay,R.K., Halder,N.C., Maiti,S.C. and Bose,T.K., 1980. Effect of nitrogen nutrition on growth, yield and quality of "Giant Governor" banana. **National Seminar on Banana Production Technology.** TNAU, **Coimbatore.** pp. 109-112.

Chen W. and Ku Z., 1985. Isolation of mesophyll cells and protoplasts and protoplast fusion and culture in Banana. **J. agric. Assoc. of China. 129**: 55-66.

Chu,C.C., 1960. The effect of potash fertilizer on the production of banana. **Soil and Fert., 24**: 1704.

*Chu,C.C. 1961. The effect of potash fertilizer on the production of banana. **Soil and Fert., 25**: 39-41.

Chundawat, B.S., S.K.Dev and N.L.Patel., 1983. Effect of sources of nitrogen on yield and economics of Basrai banana. **Haryana J. Hort. Sci. 12(3)**: 156-159.

*Claypool,L.L., 1936. Irrigation of orchards. **Proc. Wash. Hort. Sci. 30** : 78-104.

*Cox E.A., G.Stotzky and R.D.Goos, 1960. *In vitro* culture of *Musa balbisiana* Cola embryos. **Nature 185**: 403-404.

Cronauer S.S. and A.D.Krikorian, 1983. Somatic embryos from cultured tissues of triploid plantains (*Musa* ABB). **Plant cell Reports 2**: 289-291.

Cronauer,S.S. and A.D.Krikorian, 1984a. Rapid multiplication of bananas and plantains by in vitro shoot tip culture. **Hort. Sci. 19**(2) : 234-235.

Cronauer,S.S. and A.D.Krikorian., 1984b. Multiplication of *Musa* from excised stem tips. **Ann. Bot.,53**: 321-328.

Cronauer,S.S. and A.D.Krikorian., 1985. Aseptic multiplication of banana from excised floral apices. **Hort. Sci. 20**(4): 770-771.

Cronauer,S.S. and A.D.Krikorian, 1986. Banana (*Musa* Spp.)**In Biotechnology in Agriculture and Forestry,** Vol.1. Trees I Ed. by Y.P.S. Bajaj. Springer / Verlag Berlin, Heidelberg 1986.

Cronauer, S.S. and A.D. Krikorian, 1987. Adventitious shoot production from calloid cultures of banana. **Plant Cell Reports 6**: 443-445.

Cronauer S.S. and A.D.Krikorian, 1988. Plant regeneration via somatic embryogenesis in the seeded diploid banana *Musa ornata* Roxb. **Plant Cell Reports 7**: 23-25.

*Croucher,H.H. and W.K.Mitchell. 1940. 'Fertilizer investigations with Gros Michel Banana'. **Dept. Agric. Jamaica, Bull., 19**, pp. 30.

*Cuddus,P. and G.Bunemann, 1974. The effect of seasonally varied potassium available on apple tree III. The influence on vegetative growth. **Gartenbanwissen,** 39: 69-84.

Damasco D.R. and R.C.Barba, 1985. *In vitro* culture of saba banana *M. balbisiana* cv. Saba (BBB) In **Biotechnology in International Agricultural Research,** Philippines, Manila. International Rice Research Institute, pp.41-44.

Dave, S.K., J.S. Katrodia and M.L. Patel, 1991. Studies on tissue analysis and nutrient requirement in banana. **Indian J. Hort.,** 48(4): 305-308.

De Guzman, E.V., A.C.Decena and E.M. Ublade, 1980. Plantlet regeneration from unirradiated and irradiated banana shoot tips cultured *in vitro.* **The Philippine Agriculturist, 63**(2):140-146.

De Langhe E., 1984. The role of *in vitro* techniques in germplasm conservation. in J.H.W. Holden and J.T. williams (eds). **Crop Genetic Resources conservation and Evaluation.** George Allen and Uniwin, London pp. 131-137.

Desjardins Y, A. Goselin and S.Yellow, 1987. Acclimatization of *in vitro* strawberry plantlets in CO_2 enriched environment and supplementary lighting. **J. Amer.Soc. Hort. Sci. 112**(5): 846-852.

Dhed's,D., F.Dumortier, B.Panis, D.Vuylsteke and E.De Langhe, 1991. Plant regeneration in cell suspension cultures of the cooking banana cv. Bluggoe. **Fruits 46:** 125-135.

Doreswamy,R. and L. Sahijram, 1989. Micropropagation of banana from male floral apices cultured *in vitro.* **Scientia Horticulturae 40** : 181-188.

Doreswamy, R., N.K. Srinivasa Rao and E.K. Chacko, 1983. Tissue culture propagation of banana. **Scientia Horticulturae, 18**: 247-252.

*El-Khoreiby,A.M.K. and A.T.Salem, 1991. Effect of potassium on the vegetative growth and the nutritional status of Dwarf Cavendish banana. **Bulletin of faculty of Agriculture, University of Cairo, 42**(1) : 247-258. Hort. Abstr.

Escalant, J.V, and C.Teisson, 1988. Somatic embryogenesis in *Musa* spp.) Embryo genise somatic chez *Musa* spp. **Comptes Rendus de I Academic des science III (sciences de la vie), 306**(8): 277-281.

Evans, L.T., 1971. Flower induction and the floral concept.A. Rev. Pl. Physiol., **22** : 365-94.

Fitchet,M., 1987. Somatic embryogenesis in callus of D.C. banana. **Information bulletin No. 176.** Citrus subtropical Institute, South Africa, pp.1-2.

*Fourcroy and Vanquelin, 1807. Analyse du sue de bananier. **Anns. Mus.d' Hist. Nat. 9**, 301-302.

Funaioli,A., 1962. La concimazione de banano in Somalia. **Riv. Agr. Subtrop. Trop. 56**: 381-394.

Garcia, Martinezal,J.L. and M.A.Garciapapi, 1979. Influence of gibberellic acid on early fruit development, diffusible growth substances and content of micro nutrients in seedless clementine Mandarin. **Scientia Horticulturae., 11** : 337-349.

Garita,C.R. and C.R.Jaramillo, 1984. Response of cultivar Giant Cavendish (*Musa* AAA) to increasing rates of potassium in soils on the Caribbean coast of Costa Rica. **Informe Mensual UFEB, 8** : 50-55.

George, E.F. and P.D. Sherrington, 1984. Plant propagation by tissue culture: **Hand book and directory of commercial laboratories** , Exegetices Ltd., Evesely, Baningstoke, UK.

Gopimony, R., K.C. Marykutty and K. Kannan, 1980. Effects of top dressing with urea at flower initiation time in banana cv. Zanzibar. **Agric. Res. J. Kerala, 17(2)**:293-295

*Gottreich,M., D. Bradu and Y.Halovy, 1964. A simple method for determining average banana fruit weight. Ktavim 14 : 161-162. Cited from Lahav, E. 1972. **Trop. Agric. Trin., 49** : 321-325.

Greenberg,E. and J.Preiss, 1965. Biosynthesis of bacterial glycogen. II. Purification and properties of the adenosine diphospho glucoseglycogen transflucosylase of *Avthrobacter* species. **J. Biol. Chem., 240**: 2341-48.

Gupta,P.P., 1986. Eradication of mosaic disease and rapid clonal multiplication of banana and plantain through meristem tip culture. **Plant Cell Tissue and Organ Culture 6**: 33-39.

Hazarika,D.N. and N.K.Mohan, 1991. Effect of nitrogen on growth and yield of banana cv. Jahaji (AAA group). **Horticultural J. 4**(1): 5-10.

Hazarika,D.N. and N.K.Mohan. 1992. Effect of nitrogen on leaf N, P and K contents and yield of "Jahaji" (*Musa* AAA group) Cavendish sub-group. **South Indian Hort., 40**(3): 172-174.

Hegde, D.M. and K. Srinivas, 1991. Growth, yield, nutrient uptake and water use of banana crops under drip and basin irrigation with N and K fertilization. **Trop. Agric., 68**:4.

Hewitt,C.W., 1955. Leaf analysis as a guide to the nutrition of bananas. **Emp. J. Exp. Agric., 23** : 11- 16.

*Hewitt,C.W. and R.E.Osborne, 1962. Field studies on leaf analysis of Lacatan bananas as a guide to the nutrition of the plant. **Emp. J. Exp. Agric., 30** : 249-56.

Ho,C.T., 1968. The influence of K, split applications of fruit yield and some characters of banana. **Soils and Fertilizer, 33** : 16-27.

*Ho,C.T., 1969. Study on correlation of banana fruit yield with leaf potassium content. **Fertilite, 33**: 19-29.

Huber, S.C., 1985. Role of potassium in photosynthesis and respiration. In **Potassium in Agriculture** (ed. Munson,R.D.). American Society of Agronomy, Wisconsin, USA : 369-396.

*Humphries,E.C., 1956. **Modern method of plant analysis, 1** : 468-502.

Hwang,L.C. and D.L.Chi, 1988. Pivotal role of picloram and gelrite in banana callus culture. **Environmental and experimental Botany, 28**(3): 249-258.

Hwang, S.C., C.L.Chen, J.C.Lin and H.L.Lin, 1984. Cultivation of banana using plantlets from meristem culture. **Hort. Sci. 19**(2) : 231-233.

Irizarry, H., F. Abruna, J. Rodriguez-Garcia and N. Diaz, 1981. Nutrient uptake by intensively managed plantations as related to stage of growth at two locations. **J. Agric. Univ. Peurto Rico, 65(1)**: 331-345.

Israeli,Y. and Lahav,E., 1986. **Banana in CRC Hand Book of Fruit set and Development** (Ed. Monselise, S.P.) CRC Press, Florida : 45-73.

Jackson,J.E., 1958. **Soil Chemical Analysis.** Prentice Hall Inc.,Englewood Califfs, N.J.

*Jackson,M.L., 1973. **Soil chemical analysis.** Constable and Co.Ltd., Lond. pp. 123.

*Jacob, H. and vonVexkull, 1960. **Fertilizer use, nutrition and manuring of tropical crops.** Second edition, Hannovar

*Jagirdar, S.A.P., M.A. Bhutto and A.M. Sheik, 1963. Effect of spacing, interval of irrigation and fertilizer application on Basrai banana (*Musa cavendishii* L.). **W. Pakistan J. Agric. Res., 1**(2):5-20.

*Jambulingam,A.R., 1971. Studies on the effect of potash on Robusta banana (*Musa cavendishii* Lambert.). Thesis submitted to the Annamalai University for M.Sc.(Ag.) degree.

*Jambulingam,A.R., N.Ramaswamy and C.R.Muthukrishnan, 1975. Studies on the effect of potassium on `Robusta' banana. **Potash Rev., 27** : 4-6.

Jaramillo,C.R. and R.Bazan., 1976. The effect of urea and sulphur - coated urea on banana production in the cultivar Giant Cavendish in Guapiles, **Costa Rica, Turrialoa, 26**(1): 90-5.

Jarret, R.L., 1986. *In vitro* propagation and genetic conservation of banana and plantains. In international board for plant genetic resources, **Report of third meeting held in Valencia, Spain 4 to 6, June, 1986.**

Jarret,R.L., J.B.Fisher and R.E.Litz, 1985a. Organ formation in *Musa* tissue cultures. **Plant Physiol. 12**(2) : 123-130.

Jarret,R.L., W.Rodriguez and R.Fernandez, 1985b. Evaluation tissue culture propagation and dissemination of `Saba' and `Pelipita' plantains in Costa Rica. **Scientia Horticulturae (Amst) 25**: 137-147.

*Joseph,A.R., 1969. Studies on progressive changes in forms, fixation and availability of K in soils of Tamil Nadu as influenced by nitrogenous fertilizer application. M.Sc.(Ag.) Thesis submitted to TNAU, Coimbatore.

*Joseph,K.T., 1971. Nutrient content and nutrient removal in bananas as an initial guide for assessing fertilizer needs. **Planter, 47** : 7-10.

*Katyal,S.L. and K.L.Chadha., 1961. Banana. **Fertilizer News, 6**: 16-19.

*Kohli, R.R., E.K. Chacko and G.S. Randhawa, 1976. Effect of spacing and nutrition on growth and fruit yield of Robusta banana. **Indian J. Agric. Sci., 36(8)**:382-386.

Kohli, R.R., B.R.V. Iyengar and Y.T.N. Reddy, 1984. Growth, dry matter production and yield in banana as influenced by different levels of nitrogen. **Indian J. Hort., 41 (3/4):**194-198.

Kohli, R.R., G.S. Randhawa and E.K. Chacko, 1981. Growth and yield response of Robusta banana to different spacing and nutrition.**National seminar on Banana Production Technology 1980.** TNAU, **Coimbatore,** 104-106.

Kohli,R.R., Reddy,Y.T.N. and Iyenger,B.R.V. 1985. Effect of nitrogen on biomass distribution in Robasta banana. **Gartenbau Wissen Sehaft, 50(3):** 117-20.

Kothavade,D.V., P.R.Mahajan, K.U.Sanghavi and D.R.Patil, 1985. Effect of leaf area on the growth and yield of Basrai banana. **South Indian Hort., 33**(2): 122-123.

Kotur, S.C. and M.M.Mustaffa, 1984. Effect of spacing and nitrogen on the leaf nutrient status of Robusta banana. **South Indian Hort.,32(6):** 324

Krikorian A.D., 1989. *In vitro* culture of bananas and plantains: Background, update and call for information. **Trop. Agric.,** (Trinidad), **66**(3): 194-200.

Krikorian A.D. and S.S.Cronauer, 1984. Aseptic culture techniques for banana and plantain improvement. **Econ.Bot., 38:** 322-331.

Krikorian A.D. and S.S.Cronauer, 1985. Meristem and cell culture for multiplication and production of disease tolerant banana clones of plantain and dessert bananas pp. 146-147. In **Memoria del Primer,** Congress International Sobre Patano. Manizales Colombia Junio 6 al 10 de 1983.

Krishnan,B.M. and K.G.Shanmugavelu, 1983. Correlation studies in banana cv. `Robusta'. **South Indian Hort., 31:** 110-111.

*Lacoeuilhe,J.J., 1973. Rhythm of K absorption in relation with growth (pineapple and banana). In : **Potassium in tropical crops and soils. Proc. 10th Collog. IPI Switzerland** ,pp. 177.

Lahav,E., 1972. Effect of different amount of potassium on growth of the banana. **Trop. Agric., 49** : 321-335.

Lahav,E. 1973. Effects and interaction of manure and fertilizers in a banana plantation. **Israel J. Agric. Res. 23**: 45-57.

Lahav,E. and D.W.Turner, 1983. **Banana nutrition.** IPI - Bulletin No.7. International Potash Institute, Bern, Switzerland.

Langenegger,A. and B.L.Smith, 1990. N requirement of bananas in South Africa **Fruits, 43**(1): 639-643.

Laxmikanth D.M. and K. Nataraja, 1988. Micropropagation of banana through shoot-tip culture. **Curr. Sci. 58**(3): 140-141.

*Leigh,D.S., 1969. Fertilizers for banana. **Agric. Gas.**, N.S.W., 80

Lin,K.C., R.T.Li and C.T.Lin, 1962. The optimum N-P-K ratiofor banana Agric. **Res. Taiwan. 11**: 38-44.

Litz,R.E., 1984. *In vitro* somatic embryogenesis from nucellar cultures mono embryonic *Mangifera indica* L. **Hort. Sci. 19**: 715-717.

Litz,R.E., 1985. Somatic embryogenesis in tropical fruit trees. **Tissue culture in Forestry and Agriculture** I Edn. pp. 179-194. Plenum Press, New York.: 369-72.

*Ma S.S. and C.T.Shii, 1972. *In vitro* formation of adventitious buds in banana shoot apex following decapitation. **J.Hortic. Sci. China 18** : 135-142.

*Ma S.S. and C.T.Shii, 1974. Growing banana plantlets from adventitious buds (in Chinese). **China Horticulturae 20**: 6-12.

Ma S.S., C.T.Shii and S.C.Wang, 1978. Regeneration of banana plants from shoot meristem tips and inflorescence selections *in vitro*. **XXth International Horticultural Congress No.1639,** Sydney, Australia 15 to 23, August, 1978.

*Marques,M.A. and P.Monteiro, 1971. Notes on fertilizing bananas. **Rev. Agric., 136** : 29-32.

Martin-Prevel,P., 1962. Mineral elements in banana plants and fruit bunches. **Fruits, 17**: 123-128

*Martin-Prevel,P., 1964. Nutrient elements in the banana plant and fruit. **Fertilite, 22**: 3-14.

*Martin-Prevel,P., 1966. Influence of very large application of fertilizer on the mineral composition of banana. **Fruits, d'Outre Mer. 21**: 175-185.

Martin-Prevel,P., 1969. A systematic variants' trial on the banana. **Fruits, 24** : 193-215.

*Martin Prevel,P., 1973. Influence of the level of potassium nutrition on the physiological functions of tropical plants and the quality of their products. In: potassium in Tropical Crops and Soil. **Proc. 10th Colloq. I.P.T., Switzerland,** P. 223-48.

Martin-Prevel,P. 1982. New data about the physiological functions of K in the banana plant fixation, translocation and metabolism of C. **International Banana Nutrition Newsletter, 5** : 18.

Martin-Prevel,P. and Charpentier,J.M., 1963a. Culture Sur milien artificial symptomes de Carences en Six elements minerank Chez le bananier **Fruits 18**: 221-247.

*Martin-Prevel,P. and Charpentier,J.M., 1963b. **Fruit d' Outne. Mer. 18**: 221-247.

*Martin-Prevel,P. and G.Montagut, 1966. Soil plant experiments on bananas. The dynamics of N in the growth and development of the plant. **Fruits, d'Outre Mer. 21**: 283-294.

Mathew, V. and M.Aravindakshan, 1981. Nutrient uptake in rainfed banana var. Palayankodan. **Agric. Res.J. Kerala. 19(2)**: 54-61.

Matsumoto K., 1986. Isolation, culture and fusion of banana bract protoplasts. **Japanese J. Breeding 33**(2) : 302-303.

*Melin, P., 1970. Effects of heavy fertilizer application on banana. **Fruits, d'Outre Mer. 25**: 763-766.

Mengel,K., 1985. Potassium movement within plants and its importance in assimilate transport. In: **Potassium in Agriculture** (ed. Munson,R.D.) American Society of Agronomy, Wisconsin U.S.A., pp. 397-412.

Minhas,R.S. and N.C.Borah, 1982. Distribution of organic carbon and forms of nitrogen in a topographic sequence of soils. **J. Indian. Soc. Soil Sci., 30** : 135-139.

Mohan Ram H.Y. and F.C.Steward ,1964. The induction of growth in explanted tissue of banana fruits. **Can. J. Bot., 42**: 1559-1579.

*Monica, I., B.V.Defelipo, R.Conde., J. Lino and L.C.C.Passos, 1978. The response of banana cultivar Nanicao to fertilization with three levels of nitrogen, phosphorus and potassium. **Revistaceres , 25(142)**: 549-553.

Montagut,G. and P.Martin-Prevel, 1965. Essais Sol-plante Surbananiers. 1. Besoins en engrais des bananeraies antiliaises. 2. (et Lacoewilhe, J.J.) Nutrition minerale comparee dans six essais. **Fruits, 20** : 265-273, 398-410.

Montserin,B.G., 1956. Banana growing in Trinidad and Tobago J. Agric. Soc. **Trin. and Tobago 56**: 527-575.

*Morel,G.M., 1960. Producing virus free cymbidiums. **Ame. Orchid Soc. Bull. 29**: 495-497.

*Morel G.M. and C.Martin, 1952. Guerison de dahlias atteints d'une maladie a virus. **C.H. Acad.Sci. Paris 253** : 1324-1325.

Murashige T., 1974. Plant propagation through tissue cultures. **Ann. Rev. Plant Physiol. 25**: 135-166.

Murashige T., 1977. Clonal crops through tissue culture. **In plant tissue culture and biotechnological applications.** pp 392-403.

Murray,D.B., 1959. Deficiency symptoms of major elements in the banana. **Trop. Agric. Trinidad, 36**: 100-107.

Murray,D.B., 1960. The effect of deficiencies of the major nutrients on growth and leaf analysis of the banana. **Trop. Agric. Trinidad, 37**: 97-106.

Murray,D.B., 1961. Shade and fertilizer relations in the banana. Trop. Agric. **Trin., 38** : 123-132.

Mustaffa, M.M., 1983. Effect of spacing and nitrogen on growth, yield and quality of hill banana. **South Indian Hort.,31**(6) : 270-273.

Mustaffa,M.M., 1987. Growth yield of Robusta banana in relation to potassium nutrition. **J. Potassium research, 3**(3): 129-132.

Mustaffa,M.M., 1988. Effect of spacing and nitrogen on growth, fruit and yield of Robusta banana grown under rainfed conditions. **South Indian Hort., 36**(5): 228-231.

Nair,G.V., S.R.Nair and G.R.Sulekha, 1990. Effect of nitrogen, potassium and their split applications on the yield and yield components of Nendran banana grown in rice fields. **South Indian Hort.**, **38**(2): 58-62.

Nambiar, I.P.S., K.C.Marykutty, S.Balakrishnan, M.R.C.Pillai and M.C.N.Nair, 1979. Effect of split application of N and K on banana var. Nendran. **Agric. Res.J. Kerala, 17**(2): 275-277.

Nanjan, K., K.M.P.Nambisan, B.M.Krishnan, Veeraraghavatham, Paulas, N.Ramaswamy and J.B.M.Md A.Khader, 1980. The nitrogen fertilization of banana under Periyar River Command Area in Tamil Nadu. National Seminar on Banana Production Technology. TNAU. Coimbatore.113-117.

Natesh,B.B., M.Aravindakshan and P.K.Vatsalakumari, 1993. Effect of split applications of fertilizers in banana, Musa AAB 'Nendran'. **South Indian Hort., 41**(2) : 67-73.

Norris,R.V. and C.V.R.Ayyar, 1942. 'The nitrogen and mineral requirements of the plantain'. **Agric. J. India, 20** : 463-467.

Novak F.J., R.Afza, A.Micke and T.Hermelin, 1987. *In vitro* mutation induction in meristem tip culture of banana and plantain. In **Proc. Second Annual Conference of the International plant Biotechnology Network (IPB Net)** Bangkok, Thailand Jan. 11-16, 1987.

Obiefuna,E., 1984. Effect of potassium application during floral initiation stage of plantains. (Musa AAB). **Fertilizer Research, 5** : 315-319.

*Obiefuna,J.C. and P.O.Onycle, 1987. The effect of nitrogen and potassium imbalance on the growth and yield of plantains. **Hort. Abstr, 57(12)**: 10005.

Ochse, J.J., Soule, M.J., Dijkman, M.J. and Wehlburg, C., 1961. **Tropical and subtropical Agriculture** Vol.1. The Macmillan Company New york pp. 371-521.

Osborne,R.E. and C.W.Hewitt, 1963. The effect of frequency of application of nitrogen, phosphate and potash fertilizers on locatan bananas in Jamaica. **Trop. Agric. Trin., 40**(1) : 1-8.

*Oubahou,A.A. and A.Dafiri, 1987. Banana N and K nutrition. **Hort. Abstr. No.**: 6050: 57(7) : 630.

Pafli, G., 1965. Relations between abundant N supply and amino acid concentration on leaves of rice plants. **Pl. Soil, 23**: 275-284

Pandit, S.N., C.Singh., R.N.Ray., K.K.Jha and B.P.Jain, 1992. Fertilizer scheduling for dwarf cavendish banana in Bihar. **J. Res., Birsa Agricultural University. 4(1)**: 25-29.

Panis,B.A., Van Wanwe and Swewen, 1993. Plant regeneration through direct somatic embryogenesis from protoplast of banana (*Musa* sp.). **Plant Cell Report 12**: 403-407.

Parida, G.N., D.P.Ray, N. Nath and D.K.Dora, 1994. Effect of graded levels of NPK on growth of Robusta banana. **Indian Agric.38(1)**: 43-50

Pathak,R.A., B.V.Singh and C.M.Ojha, 1992. Studies on the combined effect of nitrogen and potash on the growth and yield of banana cv. Harichal. **National Symposium on Banana, Abstracts 38.**

Pillai,O.A.A. and J.B.M. Md.A.Khader, 1980. Studies on thefertilizer requirements of Robusta banana. **National seminar on banana production technology, TNAU, Coimbatore 118-121.**

Pillai,O.A.A. and K.G.Shanmugavelu, 1977. Studies on the effect of number of functional leaves on flower bud initiation in banana cultivar Poovan. **Indian J. Hort., 34** : 358-361.

Pillai,G.R., S.Balakrishnana, P.G.Veeraraghavan, G.Santhakumari and R.Gopalakrishnan, 1977. Response of Nendran banana to different levels of N, P and K. **Agri. Res. J. Kerala, 15(1)**: 37-40.

Prabhuram, R., 1992. Effect of organic manures and urea on the growth and development of banana cv. Rasthali and changes in the soil ecosystem. M.Sc.(Hort). Thesis. TNAU, Coimbatore.

Rajeevan, P.K. and N. Mohankumaran, 1992. Split application of nutrients in banana cv.Mysore. **National Symposium on banana Abstracts. P.36.**

Ram,R.A. and J.Prasad, 1988. Effect of nitrogen, phosphorus and potassium on fruit quality of bananacv. Campiergang local (*Musa* ABB). **South Indian. Hort., 36(6)** : 290-292.

Ram,R.A. and J.Prasad, 1989. Studies on nutritional requirement of banana cv. Campeirgang Local (*Musa* ABB). **Narendradeva J. Agric. Res., 4(2)** : 196-200.

Ramanathan,G., Vadivelu,S., Krishnamoorthy,K.K., Helkiah,J. and Shetty,K.S., 1973. Effects of ammonium chloride and ammonium sulphates on banana. **Madras Agric. J. 60(8)**: 1066-67.

Ramaswamy,N., 1971. Studies on the effect of nitrogen on growth and development of Robusta banana. M.Sc.(Ag.) thesis submitted to the Annamalai University

Ramaswamy,N. and C.R.Muthukrishnan, 1971. Effect of application of different levels of nitrogen on Robusta banana. **Prog. Hort.2**: 31-36.

Ramaswamy,N. and C.R.Muthukrishnan, 1974. Correlation studies in the nutrition of Robusta banana. **Indian J. Hort. 31(2)**: 145-147.

Randhawa,G.S. and C.P.A.Iyer, 1978. Three decades of research on fruits-Tropical fruits, **Indian Hort. 23(1)** : 5, 43-44.

Randhawa,G.S., C.B.Sharma,R.R. Kohli and E.K.Chacko, 1973. Studies on nutrient concentration of leaf tissue and fruit yield with varying planting distances and nutritional levels in Robusta banana. **Indian J. Hort., 30** : 461-465.

Ranganna,S., 1977. **Manual of analysis of Fruits and Vegetables products**. Tata Mcgrow Hill Pub. Co., Ltd., New Delhi.

Rao,S.N.K., E.K.Chacko., R.Doreswamy and S.Narayanaswamy, 1982. Induction of growth in explanted banana inflorescence axis of banana. **Curr. Sci., 51(13)**: 666-667.

Ray,P.K. and J.P.Yadav, 1994. Response of `Basrai' banana to different doses and time of nitrogen application in calcareous soil of North Bihar. **Horticultural J., 7(2)**: 89-96.

Ray,P.K., S.B.Sharma and J.P.Yadav, 1993. Potassium fertilization of basrai banana in alkaline calcareous soils. **J. Potassium Res., 9(1)** : 31-37.

Reddy,B.M.C., 1992. Influence of graded doses of N and its time of application on growth, yield and quality of Robusta banana. **National Symposium on Banana Abstracts.** p.38.

Reuther,G., 1990. Current status and future prospects of large scale micro-propagation in commercial plant production. **Food Biochem. 4(1)**: 445-459.

Rodriguez, G. M., 1980. Preliminary studies of potassium nutrition in bananas in Central America. **Fruits, 35** : 283-294.

Rodriguez,G.A., J.R.L.Martin and M.J.R.Enriquez, 1987. *In vitro* propagation of canary island banana (*M. acuminata* Colla AAA Var. Cavendish). Studies of factors affecting culture obtention, preservation and conformity of plants. **Acta Horticulturae 212**: 577-583.

Rowe,P. and D.L.Richardson., 1975. Breeding bananas for disease resistance, fruit quality and yield. **Tropical Agriculture Research Services**. No.2, pp 1-44.

*Samuels,G. Beale,A. and Torres,S., 1976. **24th Ann. Cong. Amer. Soc. Hort. Sci. Trop. Reg.** Mayaguez. Pueto Rico. pp. 215-224.

*Sandoval,J.A., 1985. Micropropagation de Musaceas **Asbana. 9**: 21-23.

Sannasgala,K., F.Dumortier and E.De Langhe, 1987. Plants from "proembryo like"structures in *Musa* cv. Bluggoe. **Symposium on plant micropropagation in Horticultural plants**

Selvaraj,M. and R.S.Azhakiamanavalan, 1992. Nutrient response of banana cultivars under wet land system of cultivation. **National Symposium on Banana Abstracts 39.**

Shanmugam,K.S. and Velayutham,K.S., 1972. Better manure your bananas. **Agri. Digest., 3** : 17-23.

Shanmugavelu,K.G., M.Selvarajan and Thamburaj 1987. Review of research in fruit crop in Tamil Nadu. **South Indian Hort., 35**(1/2): 24-33.

Sharma,A.K. and A.R.Roy, 1973. Fertilizer cum spacing trial on banana (*Musa paradisiaca* L.). **Indian J. Agric. Sci. 41(6)**: 493-498.

Sharma,S.B., 1985. Effect of soil and foliar application of urea on the growth and yield of banana cv. Basrai. **Punjab Hort. J., 24**(1/3): 89-91.

Shawkey,I., Z.Zidan and M.Riad., 1974. The distribution of N P and K in differnt parts of the banana leaf during life. **Egypt. J. Hort. 1**: 73-78.

*Shawkey,I., A.Bondon, S.EL-Shazly and A.EL-Gazzar, 1993. Effect of different levels of nitrogen on vegetative growth and mineral content of `Hindy' banana plant grown in sand culture. **Annals of Agricultural Science, (Cairo), 38**(2) : 699-710.

Simmonds,N.W., 1959. **Bananas**. Long mans, London. pp. 466.

Simmonds,N.W., 1966. **Bananas**. Second Edition, Longmans, London.

Simmonds,N.W., 1980. **Bananas**. Third Edition, Longmans. London.

Simmonds,N.W., 1987. **Bananas**. 4th Edn. Longmans, London.

Singh,H.P., Yadav,I.S. and Singh,K.D., 1990. Response of a plant crop of dwarf cavendish banana (*Musa* AAA, Cavendish) to N and K growing in sandy loam soils of sub-tropics. **J. Potassium Res. 47**: 133-39.

Singh,J. and P.Kashyap, 1992. Effect of spacing and levels of N on growth and yield of banana cv. Robusta. **Advances in Plant Sci. 5(1)** : 203-207.

Singh,R., 1969. **Fruits**. National Book Trust, India. New Delhi.

Smith,R.H. and T.Murashige, 1970. *In vitro* development of isolated shoot apical meristems of Angiosperms. **Am. J. of Bot. 57**: 562-568.

*Singh,S.S., 1972. Studies on the effect of nitrogen and potash on Robusta banana (*Musa cavendishii* Lambert) M.Sc.(Ag.) Thesis, Annamalai Univ. Annamalai Nagar.

*Singh,S.S., P. Kalyanasundaram and C.R.Muthukrishnan, 1973. Studies on the effect of N and K on the physio-chemical characters of Robusta banana. **Annamalai University Agricultural Research Annual, 4 & 5** : 23-26.

Singh,U.R., H.Khan and G.Singh, 1979. Effect of NPK fertilization on growth, yield and quality of banana cv. Basrai dwarf. **Punjab Hort. J. 17(1/2)** : 64-69.

Srikande,S.J.C. and J.S.P.Yadav, 1954. Organic matter utilization. **J. Indian. Soc. Soil. Sci., 18**: 51.

*Steinhausen,V., 1957. Die banana Anbau and Dungung Ruhrsticket off A.G.Bochum. 1957. Schriffe Nreine Ukhertrypische and subtropische Kultrup - flanzen cited from Ramaswamy, N., 1971.

*Summerville,W.A.T., 1944. Studies on nutrition as qualified by development in *Musa Cavendishii* L. **Qld. J. Agric. Sci., 1**: 1-127.

Sundar Raj,N., Nagaraj,S., Venkataram,M.N. and Jagannath,M.K.,1972. **Design and analysis of experiments**. Directorate of Research, UAS, Bangalore. pp. 189-198.

Tanaka, 1937. Studies on the effect of three fertilizing elements upon the banana plants. **Hort. Tropic.** 7: 421-431.

Tisdale and Nelson, 1966. **Soil Fertility and fertilizers.** Macmillan Co., London. pp. 81.

*Turner,D.W. 1969. Research into fertilizers for bananas. **Agric. Gaz. N.S.W., 80** : 511-513.

*Turner,D.W., 1979. Growth and mineral nutrition of the banana - an integrated approach. Ph.D. thesis, Maequarie. **Univ. North. Ryde.**

Turner,D.W., 1985. Fertilizing banana nitrogen, phosphorus and potassium **Ag. Facts., H. 6.3.7**: 1-4.

Turner,D.W. and B.Barkus, 1981. Nutrient concentrations in a range of banana varieties grown in the sub-tropics. **Fruits 36**: 217-222.

Turner,D.W. and B.Barkus, 1982. The uptake and distribution of mineral nutrients in the banana in response to supply of K, Mg and Mn. **International Banana Nutrition News letter, 5** : 25-26.

*Turner,D.W. and J.H.Bull, 1970. Some fertilizer problems with banana. **Agric. Gang., 81** : 365-367.

*Twyford,I.T., 1967. Banana nutrition : A review of principles and practices. J. **Sci. Fd. Agric., 18** : 177-183.

*Twyford,I.T. and J.K.Coulter, 1964. Foliar diagnosis in banana fertilizer trials. Plant analysis and fertilizer problems. **Proc. Fourth Collq.**, 357-370.

Upadhyay,N.P., 1989. Effect of N, P and K fertilizer on growth yield and quality of banana cv. Harichal. **Prog. Hort. 20(3-4)**: 257-258.

*Vadivel,E., 1976. Studies on growth and development of *Musa* (AAA group Cavendish Subgroup) `Robusta' in relation to potassium nutrition. M.Sc.(Ag.) Thesis submitted to TNAU, Coimbatore.

Vadivel,E. and K.G.Shanmugavelu, 1978. Effect of increasing rates of potash on the quality of banana cultivar robasta. **Potash Review, 24(8)**: 1-4.

Veeraraghavan,P.G., 1972. Manuring cum liming experiment on Nendran banana. **Agri. Res. J. Kerala. 10(2)**: 116-118.

Venkatarayappa, T., B. Narasimham and C. Venkatesam, 1978. Effect of post-shooting application of urea on the development and composition of banana fruits. **South Indian Hort., 26**:109-117.

Venkatesam,C.K., V.Reddy and V.S.Rangacharlu, 1965. Studies on the effect of nitrogen, phosphoric acid and potash fertilization on the growth and yield of banana. **Indian J. Hort. 22(2)**: 175-184.

Vessey, J.C. and J.A.Rivera., 1981. Meristem culture of bananas. **Turrialba** 31(2): 162-163

Vuylsteke, D. and E.A.L. De Langhe, 1985. Feasibility of *in vitro* propagation of banana and plantains. **Trop. Agric., 62**: 323-328.

Wardle,K., K.B.Dobbs and K.C.Short, 1983. *In vitro* acclimatization of aseptically cultured plantlets to humidity. **J. Amer. Soc. Hort. Sci. 108**: 386-389.

*Warner,R.M., R.L.Fox and S.Prasomsook, 1974. Nutritional guidelines for the Williams Hybrid banana. **Hawaii Farm Sci., 2** : 4-6.

Williams, R.F., 1946. The physiology of plant growth with special reference to the concept of net assimilation rate. **Ann. Bot., 10**: 41-72.

Wong, W.C., 1986. *In vitro* propagation of banana (*Musa* Spp.):Initiation, proliferation and development of shoot tip cultures on defined media. **Plant cell tissue and organ culture, 6**: 159-166.

Wood.R.C., 1939. A note book of Tropical Agriculture. Imperial College, **Tropical Agriculture**, Second Edition pp: 143-145.

Wu,R.Y. and H.J., Su, 1990. Production of manoclonal antibodies against banana bunchy top virus and their use in enzyme-linked immunosorbent assay. J. **Phytopath. 128**: 203-208.

Yadav,I.S., H.P.Singh and K.D.Singh, 1988. Response of banana to different levels and frequency of potassium application. **South Indian Hort. 36(3)** : 167-171.

Yang, P.S. and P.T. Rao, 1962. Studies on the effect of potash on banana. **Potash Review Sub., 27(4)**: 37.

Zamora A.B., R.C.Barba and O.P. Damasco, 1986. Status and prospectus of tissue culture Research on Banana. **Proc. Int. Workshop,** Davao, Philippines 25-27 FEb. 1085. PCARRD Book Series No.41 pp.78-88.

APPENDICES

Appendix I

Composition of Murashige and Skoog's basal media

Sl. No.	Stock solution	Actual conc. (mg/l)
Macronutrients		
1.	KNO3	1900.000
2.	NH4NO3	1650.000
3.	CaCl2 2H2O	440.000
4.	MgSO4 7H2O	370.000
5.	KH2PO4	170.000
Micronutrients		
6.	MnSO4 4H2O	22.300
7.	ZnSO4 7H2O	8.300
8.	H3BO3	6.200
9.	KI	0.830
10.	CuSO4 5H2O	0.025
11.	Na2MoO4 2H2O	0.025
12.	COCl2 2H2O	0.025
13.	FeSO4 7H2O	27.800
14.	Na2 EDTA	37.300
Vitamins		
15.	Myo-inositol	100.000
16.	Thiamine HCl	0.100
17.	Nicotinic acid	0.500
18.	Pyridoxal HCl	0.500