

**STUDIES ON SPACING AND PHOSPHORUS
LEVELS ON SEED YIELD AND QUALITY, AND
VARIETAL IDENTIFICATION IN FRENCH BEAN**

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LEVELS ON SEED YIELD AND QUALITY, AND
VARIETAL IDENTIFICATION IN FRENCH BEAN**

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By

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CERTIFICATE

This is to certify that the thesis entitled "STUDIES ON SPACING AND PHOSPHORUS LEVELS ON SEED YIELD AND QUALITY, AND VARIETAL IDENTIFICATION IN FRENCH BEAN" submitted by Mr. PRASHANTH N. D. for the degree of MASTER OF SCIENCE (AGRICULTURE) in SEED SCIENCE AND TECHNOLOGY, to the University of Agricultural Sciences, Dharwad is a record of research work done by him during the period of his study in this university under my guidance and the thesis has not previously formed the basis for the award of any degree, diploma, associateship, fellowship or other similar titles.

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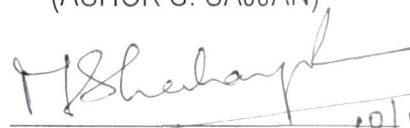
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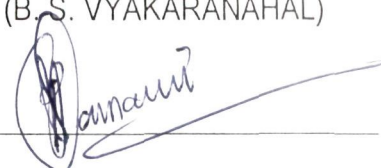
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Introduction

I INTRODUCTION

India is the largest producer of vegetables from temperate to humid tropics and from sea level to snow line regions. India being second largest producer of vegetables in the world next to China, accounting about 2 per cent of the total area of the country, which is very low in view of national requirement. Importance of the vegetables is well known both as food as well as the source of vitamins, proteins, carbohydrates and minerals. The per capita consumption of vegetables in India is around 130 g as against the requirement of 285 g for a balanced diet (Anon., 2000a). Use of green vegetables and fruits in our daily life is as important as cereals and pulses for maintenance of health.

French bean (*Phaseolus vulgaris* L.) is one of the important vegetable pulses grown extensively in the tropical and subtropical areas of world. In India, it is consumed both as green vegetable (french bean) as well as dry seeds (Rajmah). This is the rich source of carbohydrates (61.4%), proteins (17.5 to 28.5%), minerals (3.2 to 5.0 %) and vitamins. (Singh *et al.*, 1996).

French bean is major pulse and commercial vegetable crop grown in Southern and Northern India. It occupies an area of 25.91 million hectares in the world with a total production of 18.84 million tonnes, in which India grown in an area of 9.72 million hectares with a production of 4.34 million tonnes. The productivity in India is 447 kg seed per hectare as against the world productivity of 699 kg seed per hectare (Anon., 2002). In India it is mainly grown in Himachal Pradesh, Punjab, Haryana, Uttar Pradesh, Bihar, Gujarat, Madhya Pradesh, Maharashtra, Karnataka, Andhra Pradesh and Tamil Nadu. In Karnataka it is grown as green

vegetable crop in the districts of Belgaum, Dharwad, Bangalore, Mysore and Hassan *etc.*,

Quality seed is one of the basic and crucial input for any successful vegetable production. The seeds which are produced with intensive care give uniform germination, rapid root development and growth, there by permitting high yields per unit area, the most important aspect in seed programme is the supply of high quality seeds to cultivators for commercial green vegetable production. It is also necessary to produce genetically pure seed and preserve the qualities of seed from harvest to next planting season.

Seed production in vegetables is a specialized field, it includes several factors such as seed source, soil, climate pest and diseases control and cultural practices, which greatly influence the seed production of french bean. For any successful seed production, the knowledge of french bean crop, its growth habit, flowering, mode of pollination, isolation distance, harvesting, threshing, cleaning, grading, packaging and storage are essential. In India very little attention has been paid towards the scientific production of french bean seed, with the growing consciousness about high yielding varieties among the farmers. The demand for quality seed has been increased resulting in more area under seed crop every year. Since the seed yield and quality of french bean either for seed production or for commercial cultivation (for green vegetable purpose) depend on number of factors, among which optimum plant geometry or spacing and proper phosphorus nutrition have a great influence on the growth and yield of seed crop.

Seed yield and quality are the net result of an interplay of diverse metabolic activity taking place in different plant parts, which are largely

influenced by the environmental factors. The maximum seed yield in a crop under a given set of environment can be obtained with a plant density where inter and intra competition among the plants is minimum. This can be achieved with the optimum plant density which not only help to utilize sun radiation, soil moisture and nutrients more effectively, but also avoid excess competition among the plants. This information on this aspects in french bean is lacking hence efforts are made to investigate on this aspect in this study.

Phosphorus is a major plant nutrient known to have a role in synthesis of protein and a source of power in the form of ATP and ADP. Cell division is also affected by phosphorus. It is a component of many enzymes, co-enzymes, nucleic acid and phospholipids. Phosphorus plays an important role in crop growth and seed yield of pulse crop. By manipulating the morphology and physiology of the plant it is possible to bring optimum source sink relationship which results in high seed yield coupled with good seed quality attributes. In pulses, phosphorus is one of the major and crucial limiting factor, the deficiency of it may adversely affect the plant in utilization of full nitrogen and potassium and excess of phosphorus may result in various noticeable disorders. Therefore, there is a need to study the phosphorus nutrition on yield and quality of french bean.

Considering all these points an investigation on the effect of spacing and phosphorus on crop growth, seed yield and quality of french bean was under taken.

In countries having Plant Breeding Right in operation, a new variety is registered only if it is distinct from other varieties, uniform in the characteristics and genetically stable. Therefore, description of characters

for the assessment of varietal purity and identity are essential in any crop for seed production and seed certification programme (Smith *et al.*, 1995). However, Grow-out Test (GOT) is a time consuming process and some times require skilled labour and to be conducted in off-season. Hence, there is a need to develop rapid techniques at molecular level. Electrophoretic technique at protein and DNA level is one of the advanced techniques used to identify the given variety in the seed production and certification programme (Lorenzetti and Falcinelli, 1987 ; Rutz, 1990). Hence, morphological and elecetrophoretic identification of french bean varieties was undertaken.

Considering all these points in view, a study on french bean was initiated with following objectives.

- i) To know the effect of spacing levels on crop growth, seed yield and quality of french bean cv. Arka Suvidha (IIHR-909).
- ii) To study the effect of phosphorus levels on crop growth, seed yield and quality of french bean cv. Arka Suvidha (IIHR-909).
- iii) To study the interaction of spacing and phosphorus levels on crop growth, seed yield and quality of french bean cv. Arka Suvidha (IIHR-909).
- iv) To evaluate french bean varieties for seed yield and quality parameters.
- v) To identify the different french bean cultivars based on morphogenetic characters
- vi) To identify the different french bean cultivars through electrophoretic analysis.

Review of Literature

II REVIEW OF LITERATURE

The available literature relating to growth, seed yield and quality parameters as influenced by the spacing and phosphorus levels and varietal identification have been presented in this chapter.

2.1 EXPERIMENT - I : STUDIES ON SPACING AND PHOSPHORUS LEVELS ON GROWTH, SEED YIELD AND QUALITY OF FRENCH BEAN cv. ARKA SUVIDHA (IIHR-909)

2.1.1 Plant spacing

Crop yield is the result of final plant population, which depends upon the seed germination per cent and survival rates. The higher plant density brings out certain modification in the growth of plants. Plant height increased with increases in plant population due to competition for light. Leaf orientation is also altered due to population pressure.

Decrease in the yield of individual plants at higher plant density is due to reduction in the number of pods. Thus optimum plant population is necessary to obtain maximum seed yield with higher quality.

2.1.1.1 Growth and growth parameters

Pande *et al.* (1974) conducted an experiment to know the effect of spacing on french bean performance in Almora, Uttar Pradesh. They observed that 30 cm spacing resulted in highest plant height over other spacing levels. Whereas, 60 cm spacing recorded the more number of branches per plant (4.0).

In Maharashtra on sandy loam soil, Jadhavo (1993) found that there was no significant differences in plant height, however branches

per plant were numerically higher under low plant density of 2,22,000 plants per ha as compared to 3,33,000 plants per ha.

Shivakumar *et al.* (1996) at IARI, New Delhi observed the highest plant height (20.2 cm) at 30 cm x 15 cm spacing. While more number of branches per plant were seen at the spacing of 45 cm x 15 cm in Frenchbean.

Chatterjee and Som (1991) in West Bengal, on sandy loam soil observed that increasing intra row spacing from 10 to 20 cm increased the plant height from 34.37 to 37.65 cm and the same trend was observed in number of branches per plant, that increased from 6.18 to 7.80, respectively.

Dwivedi *et al.* (1995a) while studying the effect of plant spacing in frenchbean at Jabalpur, Madhya Pradesh. The plant height was the highest (29.20 cm) at spacing of 30 cm x 20 cm, when compared to other spacing levels of 45 cm x 20 cm and 60 cm x 20 cm, which recorded more number of leaves (16.71) and branches per plant (7.65)

At Rahuri on clay soil, Rajmash produced taller plants (27.15 cm) when grown under higher plant density of 4,44,444 plants per ha than to lower plant density of 2,22,222 plants per ha (23.94 cm) (Kenjale *et al.*, 1995).

Ahlawat (1996) at IARI, New Delhi, revealed that under higher plant density (3,33,000) Rajmash grew taller (27.93 cm) than to lower plant density (2,22,000) was (27.33 cm).

Singh *et al.* (1996) recorded maximum plant height (17.5 cm) and branches per plant (6.7) at the spacing of 30 cm x 10 cm over 25 cm x 10 cm in French bean cv. PDR-14.

At Mohanpur (West Bengal), it is observed that a narrow row spacing of 20 cm resulted in taller plants (42.3 cm) than wider row spacing of 30 cm in French bean (Das *et al.*, 1996).

In an experiment carried out by Dhanjal *et al.* (2001) on French bean at Barant, Uttar Pradesh. The highest plant height (26.81 cm) was observed at the plant density of 5,00,000 plants per ha, maximum branches per plant, while maximum dry matter was recorded at the plant density of 2,50,000 plants per ha.

2.1.1.2 Yield and yield parameters

Enyi (1972) reported that seed yield was increased 27, 50 and 48 per cent by adopting 90, 60 and 30 cm spacing respectively and number of pods per ha increased by 24, 57 and 99 per cent, respectively.

Pande *et al.* (1974) found that wider spacing (60 cm) in French bean cv. Black prince recorded maximum pods per plant (15.48), pod length, pod weight and pod yield per hectare over other spacing levels of 45 and 30 cm.

Goulden (1975) found that number of pods per plant and yield per plant were inversely correlated with yield per ha. Yields generally increased with increasing plant density, especially with decreasing in row width, higher yields were obtained by the narrow row spacing.

Prasad *et al.* (1978) concluded that inter and intra row spacing of 25 x 25 cm produced significantly higher seed yield (17.44 q/ha) than other spacings. The minimum yield (13.97 q/ha) was recorded in wider spacing of 30 x 30 cm.

Badillo *et al.* (1978) found that pod and plant weight differed with row spacing. These were highest at 60 cm row spacing than with 30 and 45 cm. The lesser number of pods per plant produced with 30 cm row spacing and they also reported that higher seed yield of 2.27 t per ha was obtained with 30 cm row spacing and lowest seed yield (2.16 t/ha) was obtained in 30 cm row spacing.

Iliev and Ivanov (1988) observed that increased plant densities resulted in increasing yields from 3.72 to 4.02 t per ha at 25 cm spacing in French bean.

The maximum number of pods per plant, seeds per pod 100 seed weight were recorded at plant density level of 2,22,000 plants per ha. Whereas, higher seed yield 1,020 kg per ha was recorded at 3,33,000 plants per ha in French bean at Directorate of Pulse Research, Kanpur, Uttar Pradesh, (Masood Ali and Tripathi, 1988).

Chatterjee and Som (1991) while studying the influence of spacing on French bean cv. Contender seed yield at Kalyani, West Bengal. More number of pods per plant (7.80), number of seeds per pod (5.90) and test weight (385.7 g) was recorded at the spacing level of 40x20 cm where as highest seed yield per ha (20.92 q) was recorded at 40 x 15 cm spacing.

In Maharashtra on sandy loam soil, it was observed that Rajmah produced more number of pods per plant (11.1) and seeds per pod (4.0) at plant density of 1,66,000 plants per ha. while the seed yield was maximum at 2,22,000 plants per ha (Jadhavo, 1993).

Dwivedi *et al.* (1995a) observed that maximum pods per plant, seeds per pod (4.12), filling per cent (85.25) and seed yield per plant (46.35 g) were recorded at spacing level of 60 x 20 cm, whereas seed yield was

highest (21.15 q/ha) at 30 x 20 cm spacing as compared to 45 x 20 cm and 60 x 20 cm spacing in French bean cv. Contender.

Kenjale *et al.* (1995) found that pods per plant were highest at plant density of 2,22,222 plants per ha (7.87). On contrary, seed yield was highest at high plant density level of 3,33,333 plants per ha (1,143 kg /ha).

Shivakumar *et al.* (1996) in an experiment on French bean at IARI, New Delhi, found that maximum pods per plant (5.6) was recorded at spacing of 45 x 15 cm where as maximum pod weight per plant (5.3 g), pod length (8.8 cm) and seed yield (8.3 q/ha) were observed at spacing of 30 x 15 cm.

Spacing level of 30 x 10 cm in French bean recorded the highest number of pods per plant (18.6) pod length (9.85 cm), seeds per pod (3.85) and 100 seed weight (47.2 g) as compared to other spacing level of 25 x 15 cm, 25 x 10 cm and 30 x 20 cm (Singh *et al.*, 1996).

Das *et al.* (1996) noticed that there was no significant difference in pods per plant, seeds per pod and seed weight in the row spacings of 30 cm, where as 20 cm row spacing gave significantly higher seed yield (9.9 q/ha) as compared to 30 cm row spacing (7.5 q/ha).

Dhanjal *et al.* (2001) observed that plant density of 2,50,000 plants per ha yielded highest pods per plant (5.81), seeds per pod (3.88) and 100 seed weight (30.91 g) over other densities of 3,33,000 and 5,00,000 plants per ha.

2.1.1.3 Seed quality parameters

✓ Loato *et al.* (1982) observed that individual seed weight was higher with wider spacing than narrow spacing.

✓ Masood Ali and Tripathi (1988) observed that maximum per cent of seed protein (22.8%), when the population density was 2,22,000 plants per ha as compared to population of 3,33,000 plants per ha (20.5%) in french bean.

2.1.2 Phosphorus levels

Phosphorus forms a source of power in the form of ATP and ADP. Cell division is also affected by phosphorus. It is also a component of many enzymes, co-enzymes, nucleic acid and phospholipids. In determining the yield of pulses phosphorus is one of the major and crucial limiting factor. The deficiency of the phosphorus may adversely affect the plant in obtaining the full supply of nitrogen and potassium and excess of phosphorus may result in various nutritional problems.

2.1.2.1 Growth and growth parameters

✓ Dallyn and Sawyer (1959) observed that significant increase in yield of varieties of lima bean with application of phosphoric fertilizer at 200 pounds per acre recorded significantly increased yield (333 lbs) than lower doses.

Pande *et al.* (1974) recorded the maximum plant height of French bean cv. Black prince (49.28 cm) at 75 kg P₂O₅ per ha, where as number of branches per plant were maximum (4.9) at 125 kg P₂O₅ per ha at Almora, Uttar Pradesh.

Gupta *et al.* (1983) reported significant increase in the plant height and number of branches per plant with increasing levels of phosphorus application in French bean. The plant height was highest (39.6 cm) at 120 kg P₂O₅ per ha but was on par with 40 and 80 kg P₂O₅ per ha.

There was an increase in leaf area index from (0.491 to 1.340 at 80 kg P₂O₅ per ha over 40 kg P₂O₅ per ha which is from 0.364 to 1.289. From 15 to 60 days after sowing in french bean (Srinivas and Naik, 1988).

Application of P₂O₅ at 120 kg per ha was resulted in maximum plant height (36.68 cm) and number of branches cv. Contender (4.87) over the remaining levels like 80 and 40 kg P₂O₅ per ha French bean (Chatterjee and Som, 1991).

The Nodule forming ability (nodule number and dry weight) was increased due to increased supply of P from 0 to 19.05 kg P₂O₅ per ha in french bean (Gobra *et al.*, 1993).

Dwivedi *et al.* (1995b) while studying the influence of phosphorus at Jabalpur Madhya Pradesh it was observed that P₂O₅ at 120 kg per ha recorded maximum plant height (29.6 cm), leaves per plant (16.66) branches per plant (7.62) and leaf area per plant (95.39 cm²) over 40 and 80 kg P₂O₅ per ha in French bean cv. Contender .

Ahlawat (1996) reported that, French bean produced the highest plant height (27.55 cm) at controlled level of phosphorus when compared to 13.2 and 26.4 kg per ha.

Phosphorus application significantly decreased the days to 50 per cent flowering due to stimulatory effect of P on growth hormones which induced early flowering (Jasrotia and Sharma, 1998).

Linear increase in number of branches per plant was observed due to increased phosphorus fertilization up to 100 kg P₂O₅ per ha (Baboo *et al.*, 1998).

Roy and Parthasarathy (1999) observed that application of phosphorus at 120 kg per ha recorded maximum plant height (58.33 cm) and number of branches per plant (4.66) over control and other P levels of 30, 60 and 90 kg per ha at North-Eastern hills region, Meghalaya.

Singh and Singh (2000) at Allahabad noticed maximum plant height (37.5 cm) at 120 kg phosphorus per ha over 80 kg P per ha (36.8 cm) and control (34.6 cm) in french bean. Similar trend was observed for branches per plant (18.52).

2.1.2.2 Yield and yield parameters

Pande *et al.* (1974) at Almora, Uttar Pradesh found that, *Phaseolus vulgaris* responded up to 125 kg P₂O₅ per ha giving maximum pods per plant, pod length and pod yield per ha as compared to 75 and 100 kg P₂O₅ per ha, which yielded 129.99 and 147.45 kg green pod per ha respectively.

Srinivas and Prabhakar (1985) studied the response of French bean cv. Burpee's stringless to phosphorus application at 4 levels (0, 50, 100 and 150 kg P₂O₅/ha) and recorded the increased yields with 150 kg P₂O₅ per ha up to 44 per cent.

Srinivas and Naik (1988) observed that French bean cv. Arka Komal produced higher pod yield from 1136 to 8813 kg per ha by increasing phosphorus level from zero to 40 kg P₂O₅ per ha.

Chatterjee and Som (1991) obtained significantly higher yield at 80 kg P₂O₅ per ha (23.50 q/ha) over 40 kg P₂O₅ per ha (22.76 q/ha), at West Bengal in french bean.

The experiment was conducted at Jabalpur, Madhya Pradesh to know the effect of phosphorus on French bean seed yield. Significantly more number of seeds per pod, filling per cent (85.25), shelling (73.28) and seed yield per ha (19.11 q) at 120 kg P₂O₅ compared to 80 and 40 kg P₂O₅ per ha (Dwivedi *et al.*, 1995b)

Deshpande *et al.* (1995) observed that application of phosphorus at 75 kg per ha recorded maximum seed yield (2353 kg/ha) when compared to 50 kg P₂O₅ (2053 kg/ha) and 25 kg P₂O₅ per ha (1923 kg/ha) in french bean at Pune.

Trivedi (1996) recorded that application of phosphorus @ 60 kg per ha increased the pods per plant, seeds per plant, 1000 seed weight and seed yield (7.64 q/ha) over control in Black gram.

Phosphorus application at 26.40 kg per ha recorded the highest pods per plant (10.36), seeds per pod (4.90), 100 seed weight (41.80 g) and seed yield of 1,444 kg per ha over other lower level of zero and 13.2 kg P₂O₅ per ha in french bean cv. HUR 15 (Ahlawat, 1996).

Roy and Parthasarathy (1999) observed that the maximum branch number (4.66), pod length (12.20), pod breadth (1.05 cm), pod thickness (0.78 cm) and yield (76.86 kg/ha) with phosphorus level of 120 kg P₂O₅ per ha per ha as compared to other levels of 0, 30, 60 and 90 kg P₂O₅ per ha in french bean.

Singh and Singh (2000) noticed that application of 80 kg P₂O₅ per ha recorded significantly higher 100 seed weight (54.38 g), seed yield

(28.50 q/ha) and seed protein (24.7%) over 0 and 60 kg P₂O₅ per ha and it was on par with 120 kg P₂O₅ per ha in french bean.

Tomar (2001) reported in French bean significantly higher seed yield (21.83 q/ha), with the application of 60 kg P₂O₅ per ha, but on par with 80 kg P₂O₅ per ha.

2.1.2.3 Seed quality parameters

Viera (1986) reported that seed vigour was higher when French bean plants were grown in soil with 36 ppm (100 kg P₂O₅) than in 2 or 15 ppm phosphorus.

The higher seed quality parameters like germination (96.31%), field emergence (90.83%), root length (16.59 cm), shoot length (9.04 cm), dry weight of seedlings (86.92 mg) and vigour index were observed with application of 125 kg phosphorus per ha as compared to lower level in French bean (Khyad, 1996).

Meena *et al.* (2001) reported increased test weight (151.5 g) and seed protein content (23.8%) with the application of phosphorus at 60 kg per ha to chickpea.

2.2 SCREENING OF FRENCH BEAN VARIETIES FOR SEED YIELD AND QUALITY PARAMETERS

Twenty seven collections of broad bean (*Vicia faba* L.) were studied for their relative performance for seed yield per plant, protein content of dry seeds and some agronomic traits. The best performance in terms of maximum seed yield per plant was exhibited by EC 284374 (53.81 g), EC 284375 (42.74 g) and Palampur Local 3 (42.64 g). in dry seeds high

protein content was obtained in lines JV1 (26.69 %), JV5 (26.25 %) and EC 284347 (26.23 %) (Singh *et al.*, 1997).

Dhanjal *et al.* (2001) evaluated three french bean varieties namely HUR-87, VL-63 and PDR-14 for yield and quality parameters, maximum number of branches per plant were observed with VL-63 (7.78) whereas maximum number of pods per plant (4.91) and 100 seed weight (32.60 g) was observed with PDR-14.

Roy and Parthasarathy (1999) screened six varieties of french bean namely Tender crop, Stringless cluster, Canadian wonder, Meghalaya dwarf (bush types), Meghalaya pole and Manipuri (pole types) for yield parameters. Maximum number of branches per plant were observed with Stringless cluster (5.44), maximum number of pods per plant with Canadian wonder (15.64) whereas variety Tender crop recorded maximum seed yield (79.57 q/ha).

2.3 EXPERIMENT- III : VARIETAL IDENTIFICATION OF FRENCH BEAN CULTIVARS

2.3.1 Morphological characters

The variety identification and varietal purity assessment are very important for maintenance of variety, hybrids and further multiplication and seed certification.

The term variety is defined as an assemblage of cultivated plants, which are distinguishable by morphological, physiological, chemical and cytological characters, provided their characters are heritable, stable and distinct. According to guidelines of International Union for the protection of Varieties (Anon., 1979), new crop variety has to be distinct from other varieties and uniform in its characteristics which are genetically stable.

Therefore, such characters have to be precisely described in order to identify the varieties.

The literature on identification of crop cultivars through seed, Seedling morphology and plant morphological characteristics has been reviewed hereunder.

2.3.1.1 Seed morphology

The useful morphological traits for varietal identification of a number of crops based on seed coat colour, glume colour, shape, size and test weight.

Elsaheed (1967) reported that varietal differences in Belati and Rebag 34 of broad beans, based on seed weight, which were 192.7 g and 317.1 g respectively per 500 seeds.

Srikantaradya (1980) classified sixty variety of soybean based on the seed coat colour. Among them, thirteen varieties were found black, three varieties were found brown to brick red and rest of the forty four varieties were cream to dull white.

Chakrabarty and Agarwal (1989a) developed seed keys for the identification of 16 black gram varieties on the basis of seed size and colour of the seeds. The seed size varied from small to medium and colour from shiny black to dull black.

The 44 chickpea cultivars were collected from the different sites of Uttar Pradesh (Anon., 1992) and these cultivars were grouped based on their variation in seed size (small, medium to large), seed shape (round and wrinkled) and seed colour (light to dark brown yellow, pink and white).

Mohanrao (1993) reported that 100 seed weight was highest (20.50 g) in PK-472 followed by Brag, KB-60 and Hardy soybean genotypes where as the lowest (14.24 g) was seen in KHSB-2.

Copeland and Mc Donald (1997) identified soybean cultivars of chippewa-64 can be easily distinguished from other varieties as they do not have black hila. Hence, there is need to use sophisticated techniques for distinguishing the varieties with black hila.

2.3.1.2 Seedling morphology

Seedling characters like pigmentation colour on hypocotyl, radicle length and seedling pubescence *etc.* were used in varietal identification.

Wanger and Mc Donald (1982) classified 36 genotypes of soybean into two groups, based on hypocotyl colour. Among them 27 genotypes were purple hypocotyl and nine genotypes having green hypocotyl.

Kozykowski and Burgoon (1983) tabulated 64 soybean cultivar based on distinguishable characters of hypocotyl pigmentation colour, pubescence colour and pubescence angle.

Larinde (1986) classified rice genotypes based on seedling length, sheath colour (green and colourless) and coleoptide, mesocotyl length. The japonica and bulu types cultivars showed short mesocotyls compared to indica types.

Chakrabarthy and Agarwal (1989b) developed seed keys for identification of 16 blackgram genotypes using seedling characters like pigmentation (strong, moderate, weak), stem hairiness (Glabrous, pubescent), leaflet shape (Lanceolate obovate), hypocotyl and radicle length.

Agarwal and Pawar (1990) classified 13 soybean genotypes into three groups on the basis of hypocotyl length (long, medium, short), two groups based on seedling pigmentation (dark purple and green) and two groups based on seedling pubescence (intense and sparse).

2.3.1.3 Plant morphological characteristics

The soybean varieties classified based on spreading type, presence of pubescence on stem, leaf shape and size, flower colour, pod colour at maturity and days taken for maturity (Agarwal, 1984).

Bahrenfus and Fehr (1984) compared soybean cultivars of Cumberland with Harper. Harper has purple flower, tawny pubescence, brown pods at maturity and higher seed yield than Cumberland, but both cultivars were similar in days to maturity and plant height.

Forty four chickpea cultivars were collected from different sites of Uttar Pradesh. The cultivars exhibited variation with regard to seeds per pod (1-30), foliage colour (green, purple and half green), plant height (20-55 cm) and branches per plant (4-10) (Anon., 1992).

Characterisation was carried out for 32 french bean cultivars. The parameters observed are ranged as leaf length (5-12 cm), branches per plant (2-6) pod length (6.50-15.00 cm), seeds per pod (3.4-8.2) and 100 seed weight (18.4-50.6 g) (Anon., 2000b).

2.3.2 Electrophoretic analysis

The basic principle of electrophoresis is the migration of charged particle under the influence of electric current. Macro molecules like amino acids, peptides, proteins, nucleotides and nucleic acids, which are involved in various biological processes, possess ionisable groups and

therefore be made to exist in solution as electrically charged species either cation or anions. Hence, when an electric current is applied across a support media, which may be polyacrylamide gel, starch gel, agarose gel or cellulose acetate, these charged proteins migrate. This migration depends on charge and size of the protein molecule which is characteristic to such protein (Wilson, 1986).

2.3.2.1 Protein and isozyme polymorphism

The composition of seed protein is highly constant and is unlikely to be affected by environmental conditions or seasonal fluctuations. Since most of the released genotypes have almost similar grain appearance, the feasibility of this technique over morphological tests is seen greater.

The most commonly observed differences among genotypes were total number of bands, intensity of bands and relative mobility values making it an effective tool for species varietal identification.

Electrophoretic isozyme technique was applied on primary leaf, stem and root tissues from seedlings of 34 common bean cultivars. Among the isozyme system studied peroxidase (PER) and esterase (EST) were found to be suitable for cultivar identification (Bassiri and Adams, 1978).

Hussain *et al.* (1986) developed electrophoretic procedures for seed proteins which can distinguish cultivars of field bean (*Phaseolus vulgaris*). Proteins were extracted from seven varieties and the extracts were analysed using acid and SDS PAGE Electrophoresis. Using a combination of band number and band location both methods had demonstrated satisfactory discrimination of the seven cultivars.

Wynne *et al.* (1991) evaluated 25 isozymes and found only three, *viz.*, GOT, isocitrate dehydrogenase (ICD) and phospho hexose isomerase

(PHI) were consistently polymorphic. Among them, GOT and PHI could be distinguished as botanical types.

The polymorphic protein bands obtained after two-dimensional PAGE allowed identification of rice sub-species and was also used to differentiate between wild perennial *Oryza rusipogan* and annual *Oryza nivara* (Saruyama and Shinbashi, 1993).

Galagalo and lopes (1994) studied leucino amino peptidase (LAP), peroxidase (POX) and glutamate oxaloacetate transaminase (GOT) in five cultivars of groundnut. LAP and POX were not discriminative and only GOT was polymorphic.

Kumar *et al.* (1995) conducted PAGE of pearl millet hybrids and their parental lines were able to characterise each line based on number, position and intensity of bands.

2.3.2.2 Random amplified polymorphic DNA (RAPD, the DNA marker)

The RAPD technology has quickly accepted and importance mainly gained because it has provided a relatively simple tool for genetic analysis in biological systems. RAPD detects nucleotide polymorphism using only one primer of an arbitrary nucleotide sequence. It has been successfully used for cultivar analysis in a number of plant species (Demeke *et al.*, 1992).

Scott *et al.* (1994) evaluated the degree of RAPD marker variability between Andean and Middle American gene pools of common bean (*Phaseolus vulgaris* L.). The overall level of polymorphism between Andian and Middle American gene pool was 83.4 per cent. The overall level of polymorphism between the races within the same gene pool was similar for Andean races (60.4%) and middle American races (61.7%).

Thirty-two varieties of *Vigna radiata*, procured from IARI, New Delhi and released in six different locations were analysed using Random Amplified polymorphic DNA (RAPD) technique. A total of 48 random dicamer primers were surveyed and 21 primers were found to be useful. Various levels of polymorphism were obtained with different primers (Anon., 1998).

The genetic composition of 69 chilean landraces, 15 commercial cultivars of common bean grown in chile were examined using RAPD technique. The 25 primers used and they created 106 polymorphic bands (Mitrick *et al.*, 1997).

Nkongolo (2003) conducted an experiment to study the pattern and extent of RAPD marker variation within and among the cowpea populations from different agro ecological zones to determine the degree of genetic relationships and gene flow among the different land races. Overall 80 per cent of the scored loci were polymorphic. The genetic distance values among accessions varied between 0.09 to 0.57.

Material and Methods

III MATERIAL AND METHODS

A field experiment was carried out to investigate the effect of spacing, levels of phosphorus in french bean cv. Arka Suvidha during *kharif* season 2002 at Main Agricultural Research Station, and seed quality was studied in Laboratory of Seed Science and Technology Department and varietal identification studies were conducted in the Laboratory of NSP/BSP Unit, College of Agriculture, Dharwad. The details of material and methods employed are presented in this chapter.

3.1 EXPERIMENT - I : EFFECT OF SPACING AND PHOSPHORUS LEVELS ON GROWTH, SEED YIELD AND QUALITY OF FRENCH BEAN cv. ARKA SUVIDHA (IIHR-909)

3.1.1 General description

3.1.1.1 Location of the experimental site

Dharwad is situated in Agroclimatic zone-8 (Northern transitional tract) of Karnataka State. Dharwad is located between 15° 26' North latitude and 76° 27' East longitude and at an altitude of 678 m above mean sea level (Plate 1).

3.1.1.2 Soil properties of experimental site

The experimental site consisted of red sandy soil which is porous and well drained in nature. The various physical and chemical properties of experimental site are given in Table 1.

The total rainfall during 2002-03 was 428.40 mm as against average rainfall of 779.4 mm for a period of 51 years. The maximum and minimum



Plate 1: General view of the field experiment at flowering and maturity stage

Table 1. Physical and chemical properties of soil from experimental site

Particulars	Values obtained	Methods adopted
A. PHYSICAL PROPERTIES		
Clay (%)	32.70	Hydrometer method (Piper, 1966)
Silt (%)	9.50	Hydrometer method (Piper, 1966)
Fine sand (%)	31.24	Hydrometer method (Piper, 1966)
Coarse sand (%)	26.56	Hydrometer method (Piper, 1966)
B. CHEMICAL PROPERTIES		
Total N (kg/ha)	265.00	Modified Kjeldal's method (Jackson, 1967)
Available P ₂ O ₅ (kg/ha)	10.80	Olsen's method (Muhr <i>et al.</i> , 1965)
Available K ₂ O (kg/ha)	245.00	Flame photometer (Muhr <i>et al.</i> , 1965)
pH	6.70	pH meter (Jackson., 1967)

temperature during the crop season from June to October were 30.7 and 20.0°C, respectively (Table 2).

3.1.2 Experimental details

The experiment consisted of twelve treatments involving two factors *viz.*, four spacing levels, as one factor and three phosphorus levels as another factor and it was laid out in split plot design with three replications. The variety used for study was Arka Suvidha (Fig. 1).

3.1.2.1 Treatment details

Main factor I : Spacing (S)

S₁- 30 x 10 cm

S₂- 45 x 10 cm

S₃- 30 x 20 cm

S₄- 45 x 20 cm

Sub factor II : Phosphorus levels (P)

P₁- 75 kg P₂O₅ /ha

P₂- 100 kg P₂O₅/ha

P₃- 125 kg P₂O₅/ha

Combination of treatments

T₁- S₁P₁

T₇-S₃P₁

T₂- S₁P₂

T₈-S₃P₂

T₃-S₁P₃

T₉-S₃P₃

T₄-S₂P₁

T₁₀-S₄P₁

T₅-S₂P₂

T₁₁-S₄P₂

T₆-S₂P₃

T₁₂-S₄P₃

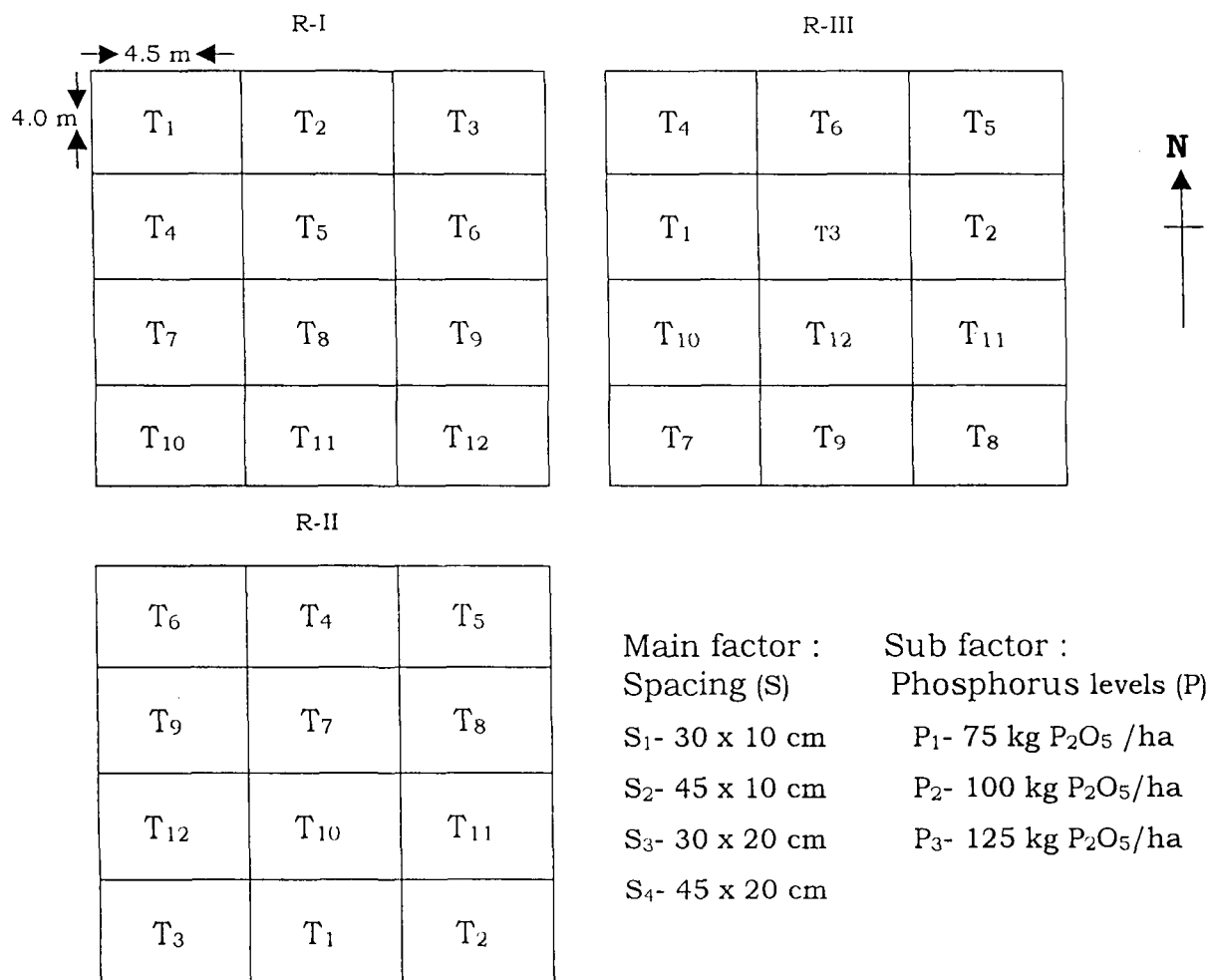
Design : Split plot design

Replication : Three

Plot size : 4.5 x 4 m

Table 2. Monthly meteorological data for the year 2002 and average of 52 years (1950-2001) of Main Agricultural Research Station, UAS, Dharwad

Months	Rain fall (mm)		Relative humidity(%)			Temperature (°C)			
						2002		1950-2001	
	2002-2003	1950-2001	2002-2003	2002-2003	1950-2001	Max	Min	Max	Min
Jan	00.00	0.098	55	63.72	30.1	14.1	29.22	14.12	
Feb	62.70	00.00	50	51.22	31.9	17.9	34.59	15.96	
Mar	00.00	7.22	49	56.69	36.0	20.0	35.75	18.77	
April	67.00	47.98	53	77.82	37.2	21.7	37.07	21.33	
May	57.90	83.42	62	67.00	34.9	23.0	36.55	21.45	
June	60.50	111.55	80	81.86	29.6	21.9	29.49	21.20	
July	15.00	150.80	79	87.74	28.4	21.3	27.03	20.95	
Aug	48.10	5.76	82	64.39	26.6	20.5	29.15	13.41	
Sept	06.60	97.88	73	86.71	29.8	20.0	27.02	20.63	
October	103.60	103.91	67	82.65	30.7	20.4	28.76	20.17	
Nov	07.00	133.11	62	76.80	30.5	17.0	30.10	19.28	
Dec	00.00	33.75	47	68.14	30.2	14.4	29.42	15.45	
Total	428.40	779.47							



Legend

T ₁ - S ₁ P ₁	T ₇ -S ₃ P ₁
T ₂ - S ₁ P ₂	T ₈ -S ₃ P ₂
T ₃ -S ₁ P ₃	T ₉ -S ₃ P ₃
T ₄ -S ₂ P ₁	T ₁₀ -S ₄ P ₁
T ₅ -S ₂ P ₂	T ₁₁ -S ₄ P ₂
T ₆ -S ₂ P ₃	T ₁₂ -S ₄ P ₃

Fig. 1 : Plan and layout of the experiment I

3.1.3 Salient feature of variety Arka Suvidha (IIHR-909)

French bean cultivar Arka Suvidha is an erect, bush type and developed at Indian Institute of Horticulture Research, Bangalore through selection generally cultivated in *kharif*. It has got green, fleshy, long, straight and stringless pods producing 200-250 quintals per ha of green pods or 15-20 quintals of seed per ha. The duration of the crop is 85-90 days. Seeds are cream in colour with smooth seed coat.

3.1.4 Cultural operation

3.1.4.1 Preparation of land

The land was ploughed once and harrowed. The clods were crushed and soil was brought to good tilth. Then the layout was carried out.

3.1.4.2 Fertilizer application

The recommended quantity of Nitrogen (62.5 kg/ha) and Potash (75 kg/ha) and measured quantity of P_2O_5 were applied as per treatments, in the form of urea, murate of potash and single super phosphate respectively.

Fifty per cent of nitrogen and full dose of phosphorus and potassium were applied as basal dose at the time of sowing as per the treatment combination on the furrows to a depth of 5 cm manually at different row spacings as fixed in the treatment. The measured quantity of fertilizers was thoroughly mixed and applied uniformly in the furrow before sowing.

The remaining fifty per cent of nitrogen was applied as top dress 30 days after sowing.

3.1.4.3 Seed sowing

The seeds were hand dibbled in the rows in accordance with treatment, to a depth of 5.0 cm. Two seed per hill were dibbled and after sowing, the seedlings were counted uniformly with moist soil. Fifteen days after sowing, thinning was carried out by retaining one healthy seedling per hill.

3.1.5 After care

Irrigation was given at critical stages of crop growth and earthing up was done 30 days after sowing. The plots were kept weed free by interculturing and hand weeding. The Endosulfan @ 2 ml per litre and Carbendazim @ 0.5 g per litre foliar sprays were given for the control of pod borer and leaf rust.

3.1.6 Harvesting, drying and threshing

The pods were harvested as and when they attain physiological maturity (pod showing yellow colour). The harvested pods were sun dried for one day and the seeds separated manually by beating with stick. Then the seeds were cleaned and further dried till the moisture content reduced to nine per cent.

3.1.7 Details of the data collection

3.1.7.1 Growth parameters

3.1.7.1.1 Plant height

The height of five randomly selected and tagged plants were measured in cm from the base to the tip of the plant at 30, 60 days and at harvest in each treatment combination

3.1.7.1.2 Number of branches per plant

The number of branches per plant at randomly selected and tagged plants were counted manually at 30, 60 days after sowing and at harvest.

3.1.7.1.3 Number of leaves per plant

Number of leaves per tagged plants in each treatment were counted manually at 30, 60 days after sowing and at harvest.

3.1.7.1.4 Plant dry weight at harvest

The five tagged plants were uprooted from each treatment at harvest. These plants were oven dried at 80°C for 24 hours and this dry weight was recorded in grams per plant.

3.1.7.2 Yield and yield parameters

3.1.7.2.1 Pod length

The length of ten pods was measured in cm from the base to the tip of the pod and average was calculated.

3.1.7.2.2 Pod weight

Weight of pod from each plot of tagged plants was recorded and average weight was computed and expressed in grams.

3.1.7.2.3 Pods per plant

The total number of pods per plant in each plot from tagged plants were counted and average was worked out and expressed as number of pods per plant.

3.1.7.2.4 Seeds per pod

The total number of seeds from the ten selected pods from tagged plants were counted and mean seeds per pod was calculated.

3.1.7.2.5 Seed weight per pod (g)

Seed weight from the selected pods of tagged plants was recorded. The mean value was worked out as seed weight in gram per pod.

3.1.7.2.6 Seed weight per plant (g)

Seeds from the pods of all the five tagged plants from each treatment was weighed separately and the average seed weight per plant was calculated and expressed in grams.

3.1.7.2.7 Seed yield per plot

The seeds separated from each net plot were weighed and seed yield per plot expressed in kilograms using electronic balance.

3.1.7.2.8 Seed yield per hectare

The unprocessed seed yield from each plot area was cleaned and graded using recommended screen size of 11.0 mm (R) x 4.75 mm oblong (Anon., 1998) and weighed. Further, using this data the processed seed yield per hectare was computed and expressed in kilograms.

3.1.7.2.9 Seed recovery percentage

Seed recovery percentage was calculated by using following formula,

$$\text{SR (\%)} = \frac{\text{Weight of processed seed}}{\text{Weight of unprocessed seed}} \times 100$$

3.1.7.2.10 Hundred seed weight (g)

Hundred seed weight in grams was recorded from each treatment combination as per the procedure given by ISTA Rules (Anon., 1985)

3.1.7.3 Seed quality parameters

3.1.7.3.1 Seed germination (%)

Germination percentage was determined by between paper towel method as prescribed in ISTA Rules (Anon., 1985). The 100 seeds with four replication in each treatment were placed in germination towels and kept in agerminator at $25 \pm 1^{\circ}\text{C}$ and 95 per cent relative humidity.

The germination counts were taken on fifth and ninth day (*i.e.* first and second count). The germination percentage was calculated on the basis of number of normal seedlings and expressed as germination percentage.

3.1.7.3.2 Rate of germination

The daily germination counts were recorded up to final count. The germination rate index was worked out by using the formula.

$$RG = \frac{G_1}{T_1} + \frac{G_2}{T_2} + \dots + \frac{G_n}{T_n}$$

Where

G_1 , G_2 and G_n were the number seeds germinated at time T_1 , T_2 and T_n were the days on which the observation was made.

3.1.7.3.3 Root and shoot length (cm)

Five normal seedlings were randomly selected in each replication and root and shoot length were measured in centimeters. The shoot length was measured from collar region to the point of attachment of cotyledons and root length from the collar region to the tip of the primary root. The average root and shoot length of five seedlings were computed and expressed in centimeter.

3.1.7.3.4 Seed vigour index

Seed vigour index was computed by adopting the formula as suggested by Abdul-Baki and Anderson (1973) and expressed in whole number.

$$\text{Vigour index} = \text{Germination (\%)} \times (\text{shoot length} + \text{root length in cm})$$

3.1.7.3.5 Seedling dry weight (mg/seedling)

The randomly selected seedlings used for measuring the seedling length and same seedlings were also utilized for determining the dry weight per seedling. These seedlings were dried in hot air oven at $70 \pm 1^{\circ}\text{C}$ for 24 hours. After drying, seedlings were kept in desiccator for cooling and further weighed and expressed in milligrams.

3.1.7.3.6 Field emergence (%)

A sample of 100 seeds in four replications from each treatment were sown on a well prepared seed bed with optimum soil moisture. The field emergence count was taken on 9 days after sowing and percentage was calculated.

3.1.7.3.7 Electrical conductivity (EC) of seed leachate

Five gram seeds were surface sterilized with 0.1 % HgCl₂ solution and thoroughly washed in distilled water later seeds were soaked in 25 ml distilled water and kept at 25+1°C for 24 hours. The leachate kept at 25+1°C for 24 hours. The leachate was decanted and the electrical conductivity of seed leachate was measured with Electrical Conductivity Bridge and expressed as dSm⁻¹ (Presley, 1958).

3.1.7.3.8 Seed protein (%)

The protein percentage in the seeds was calculated as the product of nitrogen percentage with a factor (6.25). The nitrogen percentage was estimated by modified Kjeldhal's method (Jackson, 1967).

3.1.7.3.9 Statistical analysis

The mean data were statistically analysed by adopting the appropriate methods outlined by Panse and Sukhatme (1978) and Sundarajan *et al.* (1972). The critical differences were calculated at 5 per cent level of probability, wherever 'F' test was significant. The percentage data were transformed into arcsine root transformation, wherever it is applicable.

3.2 EXPERIMENT - II : SCREENING OF FRENCHBEAN VARIETIES FOR SEED YIELD AND QUALITY PARAMETERS

3.2.1 Seed material

Genetically pure and fresh seeds of seven different french bean varieties *viz.*, RSJ-288, IIHR-909 (Arka Suvidha), MFB-1, Contender, Arka Komal, MFB-2 and MFB-3 were collected from the All India Co-ordinated

Research Project on Vegetables, University of Agricultural Sciences, Dharwad.

3.2.2 Experimental details

The experiment was laid out in Random Block Design (RBD) with three replications at Main Agricultural Research Station, Dharwad during *kharif* season of 2002. The plot size was 3 metre long with two rows of each variety spaced at 30 cm apart in which plant to plant distance was at 10 cm (Fig. 2).

3.2.3 Cultural operation

3.2.3.1 Preparation of land

The preparation of land was carried out as explained under item 3.1.4.1.

3.2.3.2 Fertilizer application

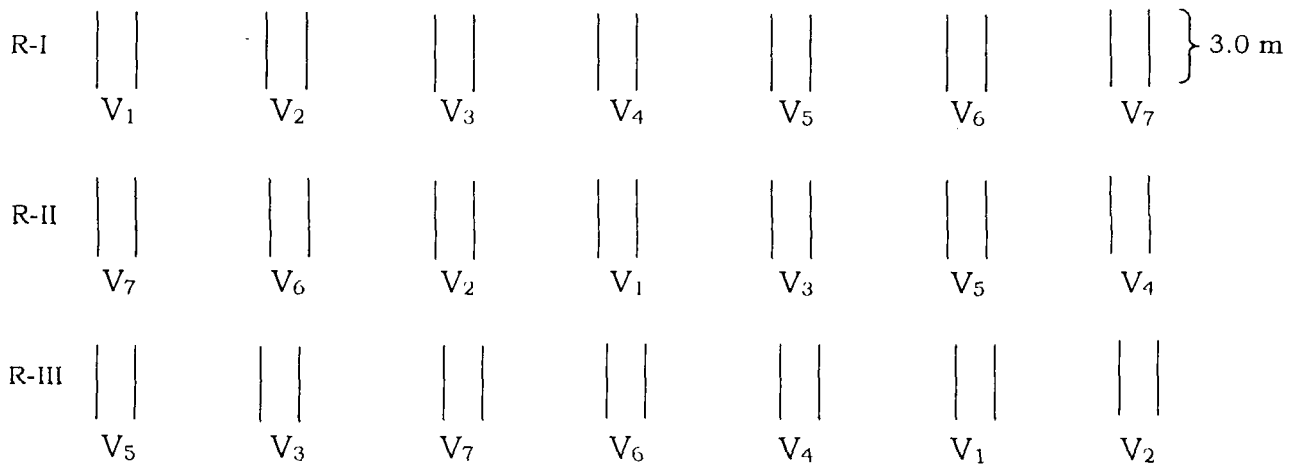
The nitrogen, phosphorus and potassium were applied in the form of urea, single super phosphate and murate of potash at the rate of 62.5, 100 and 75 kg per ha, respectively. 50 per cent of nitrogen and full dose of phosphorus and potassium were applied as basal dose at the time of sowing, the remaining 50 per cent of nitrogen was applied as top dress 30 days after sowing.

3.2.3.3 Seed sowing

The seed sowing was carried out as explained under item 3.1.4.3.

3.2.4 After care

Irrigation was given at critical stages of crop growth and earthing up was done 30 days after sowing. The plots were kept weed free by



LEGEND

- V₁- RSJ-288
- V₂- IIHR-909
- V₃- MFB-1
- V₄- Contender
- V₅- Arka Komal
- V₆- MFB-2
- V₇- MFB-3

Fig. 2 : Plan and lay out of experiment II

interculturing and hand weeding. Recommended fungicidal and insecticidal sprays were given to protect the crop from diseases and pests.

3.2.5 Harvesting, drying and threshing

The pods were harvested as and when they attain physiological maturity (pod showing yellow colour). The harvested pods were sun dried for one day and the seeds separated manually by beating with stick. Then the seeds were cleaned and further dried till the moisture content reduced to 9 per cent.

3.2.6 Details of collection of data

The data was recorded from five randomly selected and tagged plants and all biometric observations were carried out on these plants.

3.2.6.1 Growth parameters

3.2.6.1.1 Days to 50 per cent flowering

Number of days required to attain flowering in 50 per cent of the plants in each replication was taken as the days to 50 per cent flowering in a variety.

3.2.6.1.2 Number of branches per plant

The number of branches were counted in each tagged plant then totalled and average value was calculated and expressed as mean number of branches per plant.

3.2.6.2 Yield parameters

3.2.6.2.1 Number of pods per plant

Number of pods per plant was recorded as per the procedure furnished under 3.1.7.2.3.

3.2.6.2.2 Seeds per pod

Number of seeds per pod was counted as per the procedure furnished under 3.1.7.2.4.

3.2.6.2.3 Pod weight (g)

Pod weight was recorded in gram as explained under item No. 3.1.7.2.2.

3.2.6.2.4 Seed weight per plant (g)

Seed yield (g) per plant was recorded from each variety in replication and grouped as :

High yield : > 15.0 g/plant

Medium yield : 10.0-15.0 g/plant

Low yield : < 10.0 g/plant

3.2.6.3 Seed quality parameters

The observations on 100 seed weight, germination per cent, root length (cm), shoot length (cm), seedling vigour index, seedling dry weight (mg), electrical conductivity of seed leachate (dSm^{-1}) and protein content of seed in per cent were recorded as per the procedures given under item No. 3.1.7.3.

3.2.6.4 Statistical analysis

Statistical analysis were carried out as explained in the item No. 3.1.7.3.9.

3.3 EXPERIMENT - III : VARIETAL IDENTIFICATION OF FRENCH BEAN CULTIVARS

3.3.1 Morphological characters

The studies pertaining to identification of french bean varieties based on seed, seedling and plant characters were carried out at Seed Technology Research Unit, University of Agricultural Sciences, Dharwad. The details of the materials and methods employed are presented below.

3.3.1.1 Seed material

Genetically pure and fresh seeds of seven different french bean varieties *viz.*, RSJ-288, IIHR-909 (Arka Suvidha), MFB-1, Contender, Arka komal, MFB-2 and MFB-3 were collected from All India Co-ordinated Research Project on vegetables, University of Agricultural Sciences, Dharwad.

3.3.1.2 Seed morphology

The various seed attributes like seed coat colour, hilum colour, length, size, shape and 100 seed weight were recorded.

3.3.1.2.1 Seed coat colour

The seed coat colour for the seeds of all the varieties were observed under natural day light condition and classified into following colour groups *viz.*, black, white, cream and brown.

3.3.1.2.2 Hilum colour

The seeds of different varieties were categorised based on this hilum colour *viz.*, white.

3.3.1.2.3 Seed shape

Shape of the seeds were recorded by using 10 X magnifying lens in 25 seeds and they were classified as below categories *viz.*, cylinder oval and kidney.

3.3.1.2.4 Seed length (mm)

Seed length of different varieties were measured from the base to top portion of the seed in millimeter using micrometer and classified into three categories *viz.*, small (10.0 to 11.5 mm), medium (11.5 to 13.0 mm) and large (>13.00 mm).

3.3.1.2.5 Seed girth (mm)

Seed girth of seven french bean varieties was measured in millimetres and classified into three categories *viz.*, low (13.5 to 16.0 mm), medium (16.0 to 18.5 mm) and high (>18.5 mm).

3.3.1.2.6 Seed size (mm)

Seed length, width and thickness were measured by using micrometer then seed size (mm) was calculated by using the formula $(\text{length} \times \text{width} \times \text{thickness})^{1/3}$ and categorized into small (6.0 to 7.0 mm), medium (7.0 to 8.0 mm) and large (> 8.0 mm).

3.3.1.3 Seedling morphology

The fifty seeds were sown in sand media for germination of which ten normal seedlings were taken for recording morphological characters of seedling for seven varieties under study.

3.3.1.3.1 Seedling hypocotyl colour

The hypocotyl colour of the Seedling was observed under natural day light condition five days after sowing in sand media and classified into purple, green and pale green.

3.3.1.3.2 Cotyledon colour

Cotyledon colour was also observed on five day old seedling of different varieties under natural day light condition and classified in to purple, green and pale green.

3.3.1.3.3 Hairiness on seedling

Varieties were grouped based on hairiness on five day old seedling as Dense or Glabrous, Dense means thick hairiness on hypocotyl of the seedling, Glabrous means without hairiness on hypocotyl of the seedling.

3.3.1.4 Plant morphological characters under field conditions

From the tagged plants of the plots of experiment II *i.e.* screening of french bean varieties for seed yield and quality parameters, the morphological characters were recorded.

3.3.1.4.1 Growth habit

Nature of the stem, height of the plants and number of branches were determined. The growth habit of plants at maturity stage was classified as,

A. Erect : The number of branches are less and plant looks erect.

B. Spready : Due to numerous branching plant looks spready in nature.

3.3.1.4.2 Flower colour

The colour of flower from each variety at flowering stage are observed and grouped as dark purple, pale purple and white.

3.3.1.4.3 Constriction on pod

It is a very important character, which was observed on the pods and categorized as :

A : Shallow constricted pods

B : Moderately constricted

C : Deeply constricted

3.3.2 Electrophoretic analysis

3.3.2.1 Electrophoretic analysis of total proteins and Isozyme

The electrophoretic analysis was conducted at National Seed Project Research Laboratory, Seed Unit, University of Agricultural Sciences, Dharwad during 2002-03.

3.3.2.2 Equipment

- a. Pestle and mortar
- b. Centrifugation : Jouan table top micro centrifuge supplied by scientific instruments Private Limited, Bangalore.
- c. Electrophoresis unit : Model GTV 100 of ultra violet products, London.
- d. Power pack : ATTO power station, model AE-8450.
- e. Transilluminator : White/UV transilluminator of ultra violet products, London.

3.3.2.3 Procedure

The analysis of proteins and isozyme, peroxidase was carried out by adapting Poly Acrylamide Gel Electrophoresis (PAGE) technique (Davis, 1964 ; Tanksley, 1983).

Seeds of each variety were germinated on a germination paper. The six day old seedlings were used for extraction. The hypocotyl portions were chopped into small pieces and were ground well by adding chilled phosphate buffer (0.2 M, pH 8.0) using pestle and mortar, kept on ice cubes. About 1 ml of extraction buffer was used per gram of sample. The homogenate was filtered into an eppendorf tube using muslin cloth. The homogenate collected, centrifuged at 10,000 rpm for 15 minutes at 4°C. supernatant so obtained was carefully transferred to another eppendorf tube and used as a source of protein or enzyme.

Stock solutions

1. Acrylamide-bisacrylamide monomer solution : It was prepared by dissolving 30.0 g of acrylamide and 0.8 g bis acrylamide in 199 ml water. The solution was then filtered through whatman No. 1 filter paper and stored at 4°C in brown bottles.
2. Ammonium persulphate
3. Tetramethyl ethylene diamine (TEMED)

Electrophoresis buffers

1. Stacking gel buffer stock

0.5 M tris-HCl (pH 6.8), 6.0 g of Tris base was dissolved in 40 ml of water and pH was adjusted to 6.8 with HCl and final volume was made to 100 ml.

2. Resolving gel buffer stock

3.0 M Tris-HCl (pH 8.3) : 36.3 g of tris base was dissolved in 40 ml of water and pH was adjusted to 8.3 with HCl and final volume was made up to 10 ml.

3. Tank buffer (electrode buffer ; 24.8 mM Tris + 0.19 M Glycine, pH 8.3)

Three gram of Tris base and 14.4 g of Glycine were dissolved in water and pH was adjusted to 8.3 and volume was made up to 1000 ml.

4. Extraction buffer

Phosphate buffer pH 8.0 with 1% PVP

Recipe for gel preparation

Component	Resolving gel (20 ml)		Stacking gel (10 ml) (2.5%)
	(7.5%)	(12.5%)	
Monomer solution	5.0 ml	8.4 ml	1.25 ml
Resolving gel buffer	2.5 ml	2.5 ml	-
Stacking gel buffer	-	-	2.5 ml
Distilled water	12.5 ml	9.16 ml	6.25 ml
TEMED @ 0.5 µl/ml	10.0 µl	10.0 µl	5.0 µl
APS @ 0.75 mg/ml	15 mg	15 mg	7.5 mg

Preparation of slab gels

Glass plate sandwich was prepared using clean glass plates with spacing coated with a thin film of Vaseline and gel sandwich was finally assembled using the adhesive tape.

Appropriate resolving gel mixture was prepared by adding correct volumes of all components except TEMED and APS, which were added just before use. The assembled plate was held vertically in the gel casting unit and the resolving gel mixture was poured into the space between the glass plate up to 3/4th level and was overlaid with water to avoid contact with atmospheric oxygen. The assembly was left undisturbed upto polymerisation.

After the polymerisation, the top water layer was blotted and the stacking gel solution was poured and the comb was inserted avoiding air bubbles. For isozyme analysis, stacking gel was not used. After polymerization, the comb was carefully removed and wells were washed with the reservoir buffer and fixed to electrophoresis apparatus (Hames and Rickwood, 1984).

Tank buffer (200 ml) was poured in the gel apparatus. 50-100 μ g of protein sample along with 5 μ l of Bromophenol blue (Tracking dye) was mixed well and loaded into the wells. After loading of samples was complete, voltage lamps were attached and the gel apparatus was connected to power pack set at 70 volts. The gel was run till the tracking dye was 1/2-1cm away from bottom of the gel.

Once the run was completed, gel sandwich detached and placed in distilled water to avoid exposure to air. Right hand side was marked by

cutting piece of gel at the end. Later, the gel was transferred to suitable staining solution.

Staining

- i) **Protein** : 0.1% coumassie brilliant blue R 250 in methanol :
Acetic acid : water (5:2:5 v/v/v)

Staining recipe

Methanol	100 ml
Acetic acid	40 ml
Distilled water	100 ml
CBBR 250	240 mg

CBBR 250 was dissolved completely and filtered through whatman No. 1 filter paper.

Destainer solution (1000 ml)

Methanol 30% v/v	300 ml
Acetic acid 10% v/v	100 ml
Distilled water	600 ml

Gel was incubated in stain for 8 hours and non specific stain was removed by destaining.

ii) Isoenzyme- peroxidase

A. Na- Acetate buffer (0.2 M pH 5.6)	100 ml
B. Benzidine	100 mg
C. H ₂ O ₂	0.1 ml

Solution A was boiled with B and cooled and filtered. H₂O₂ was added just before, incubation. Gel was incubated (in dark) till brown bands appear and rinsed in 7% acetic acid. Gel was stored in 2% acetic acid.

Later Rm (Relative mobility) values of the bands were recorded and zymogram was drawn considering the thickness of band. Interpretation was done based on polymorphism.

3.3.2.4 RAPD Analysis

This technique uses a Polymerase Chain Reaction (PCR) method to amplify DNA with random primers. Using PCR, a random primer of arbitrary nucleotide sequence binds to the DNA at two different sites on opposite strands of the DNA template. If the primer sites are located at a close amplifiable distance from each other, this results in a DNA product by a thermocyclic reaction. This technique detects the genetic polymorphism.

Procedure followed

i) DNA isolation:

DNA from individual genotypes was isolated using slightly modified CTAB maxi-prep method (Doyle and Doyle, 1990).

1. Aliquot of 15 ml CTAB buffer was taken in the sterile tubes, added appropriate amount of β -mercaptoethanol and incubated in 65°C water bath.
2. 1-2 g fresh of young leaf from shoot apex was taken. Tissue was ground in a pre cooled pestle and mortar under liquid nitrogen to aid grinding some amount of sterile sand was used.

3. The ground tissue was transferred to polypropylene tube containing 15 ml extraction buffer pre-treated to 65°C (2-3 % w/v CTAB, 1.5 M NaCl, 20 mM EDTA, 100 mM Tris-HCl. pH-8, 0.03% β -mercapto ethanol).

Samples were incubated for 30 minutes, with intermittent shaking at every 15 minutes.

4. Equal volume of chloroform-iso amyl alcohol (24:1 v/v) was added and gently shaken for 10 minutes to form an emulsion.
5. Balanced tubes were centrifuged for 10 minutes at 6000 x g at room temperature.
6. The final aqueous phase was transferred to sterile tubes and DNA was precipitated with 0.7 volume of cold isopropanol and was hooked out into sterile eppendorf tube.
7. Pellet was washed once in 2 ml of 70% ethanol.
8. Pellet was air dried for 30-45 min.
9. Later DNA was suspended in 500-700 μ l high salt TE (10 mM Tris HCl, 1 mM EDTA, 2 M NaCl pH8) using eppendorf tubes and left for 45 minutes at room temperature or incubated in water bath at 65°C for 15 min.
10. When DNA was fully suspended RNase was added to a final concentration of 100 μ g/ml, mixed well and incubated at 37°C for 1 hour.
11. Equal volume of cold phenol : chloroform : isoamyl alcohol (25:24:1 v/v/v) mix was added and phases were separated by centrifugation (5 minutes at 14000 rpm at ambient temperature). Again the step is repeated with cold chloroform : isoamyl alcohol.

12. 0.5 volume of 7.5 mM ammonium acetate was added followed by 2 volumes of absolute ethanol DNA was hooked out.

13. Hooked out DNA was washed twice in 250 ml of 70% ethanol, late supernatant was poured off and pellet was air dried for 10 minutes. Later pellet was suspended in 200 ml of TE buffer (10 mM Tris, 1 mM EDTA pH 8).

ii) DNA quantification and quality analysis

0.7-0.8% Agarose gel in 1xTAE buffer in the presence of Ethidium bromide was used for quality checking. Presence of intact band is suitable for Polymerase Chain Reaction (PCR).

The molecular weight of the known DNA was used to quantify the DNA based on the intensity of the band. Serial dilutions were carried out to get desired quantity of DNA for PCR.

iii) PCR mix and conditions

PCR was carried out in primes thermal cycler using 0.2 ml PCR tubes.

PCR mix:

Tag DNA polymerase (3U) (Bangalore Genei)	33 μ l (Bangalore Genei)
(NH ₄) ₂ SO ₄ Taq assay buffer (10x)	2.5 μ l
dNTP mix (2.5 mM) B'lore Genei)	1.0 μ l
Primer (5 pm) operon technologies) Inc. USA	1.0 μ l
Template (20.5 ng)	1.0 μ l
SDDW	19.17 μ l
Total	25.0 μ l

PCR conditions are as follows

- 94°C for 8 min - Initial denaturation
- 94°C for 1 min - Denaturation
- 36°C for 1 min - Primer annealing
- 72°C for 2 min - Primer extension
- 72°C for 10 min - Final extension
- } 40 cycles

Primers used

Code	5' To 3'
OPF-07	CCGATATCCC
OPF-10	GGAAGCTTGG
OPF-16	GGAGTACTGG
OPF-20	GGTCTAGAGG

After completion of the reaction, amplified DNA was separated using 1.2% Agarose gel in 1 x TAE and 0.5 mg/μl Ethidium bromide. The contents were electrophoresed and visualized with UV documentation system (Herolab).

Experimental Results

IV EXPERIMENTAL RESULTS

The results of the field experiment conducted to study the effect of spacing and phosphorus levels on french bean cv. Arka Komal (IIHR 909) on seed yield and quality at the Main Agricultural Research Station and varietal identification studies made at Laboratory of NSP/BSP Unit and Seed Science and Technology Department, University of Agricultural Sciences, Dharwad during *kharif* season of 2002 are presented in this chapter as below.

4.1 EXPERIMENT - I : EFFECT OF SPACING AND PHOSPHORUS LEVELS ON CROP GROWTH, SEED YIELD AND QUALITY OF FRENCH BEAN cv. ARKA SUVIDHA (IIHR-909)

4.1.1 Growth parameters

4.1.1.1 Plant height (cm)

The results on plant height at 30, 60 days after sowing (DAS) and at harvest in french bean as influenced by spacing and phosphorus levels are presented in Table 3.

Spacing levels differed significantly at 30 DAS for plant height. Significantly highest plant height (19.67 cm) was obtained at spacing of 30x10 cm, (S₁) where as the lowest (16.26 cm) was recorded at 45x20 cm (S₄). The spacing of 30x10 cm (S₁) was on par with 45x10 cm (S₂) (19.23 cm), plant height at 30x20 cm spacing (S₃) was on par with spacing of 45x10 cm (S₂).

At 60 DAS maximum plant height was recorded at spacing of 30x10 cm S₁ (28.92 cm) which was significant over other spacing levels

Table 3. Effect of spacing and phosphorus levels on plant height at 30, 60 DAS and at harvest in French bean

Spacing (S)	Plant height (cm)											
	30 DAS				60 DAS				At harvest			
	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	18.20	19.73	21.07	19.67	27.07	29.63	30.07	28.92	27.73	31.30	32.80	30.94
S ₂ - 45 x 10 cm	18.40	19.33	19.96	19.23	26.07	26.63	27.80	26.83	27.93	28.70	29.80	28.81
S ₃ - 30 x 20 cm	16.80	18.53	19.33	18.22	24.80	25.70	26.30	25.60	26.93	27.77	28.37	27.69
S ₄ - 45 x 20 cm	12.87	17.33	18.57	16.26	22.73	24.80	25.47	24.33	25.27	27.03	27.97	26.76
Mean	16.57	18.72	19.73	18.34	25.17	26.69	27.41	26.42	27.22	28.70	29.73	28.55
For comparing means of	S				P				‘P’ at same ‘S’			
	S.Em±	CD at 5%			S.Em±	CD at 5%			S.Em±	CD at 5%		
	0.35	1.21	0.96	NS	0.63	1.21	0.96	NS	0.37	1.27	0.49	0.98
	0.32	0.96	NS	NS	0.18	0.36	NS	NS	0.16	0.33	0.98	1.36
	0.64	NS	NS	NS	0.36	0.49	NS	NS	0.33	0.45	1.36	
	0.63	NS	NS	NS	0.49	NS	NS	NS	0.45	NS	NS	

NS- Non significant

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

S- Spacing levels

of 45x10 cm, S₂ (26.83 cm) 30x20 cm, (25.60 cm) and 45x20 cm S₄ (24.33 cm), whereas S₂ and S₃ were on par with each other. similar trend in plant height was also observed at harvest stage.

There was significant effect of phosphorus levels on plant height at 30 DAS. Phosphorus level (P₃) of 125 kg per ha recorded significantly higher plant height (19.73 cm) over 100 kg, P₂ (18.72 cm) and 75 kg per ha, P₁ (16.57 cm). The minimum plant height was recorded at phosphorus level of 75 kg per ha. Similar trend in plant height was also recorded at 60 DAS and at harvest.

The interaction of phosphorus at same level of spacing on plant height was found to be non significant at 30 DAS and 60 DAS, whereas it exhibited significant at harvest. The combination of P₃S₁ recorded maximum plant height (32.80 cm) and minimum plant height of 28.73 cm was recorded by P₁ at S₁ level. The P₃ at S₂, P₃ at S₃ and P₃ at S₄ recorded maximum plant height of 29.80, 28.37 and 27.97 cm, respectively.

The interaction of spacing levels at same or different levels of phosphorus on plant height was non significant at 30 and 60 DAS, where as it showed significant differences at harvest. The P₃S₁ recorded maximum plant height of 32.80 cm and minimum was found at P₁S₄ (25.27 cm) combination.

4.1.1.2 Number of branches per plant

The data on number of branches per plant at 30, 60 days after sowing (DAS) and at harvest in french bean as influenced by spacing and phosphorus levels are presented in Table 4.

There was no significant difference in number of branches per plant due to spacing levels at 30 DAS, whereas, it was found significant

Table 4. Effect of spacing and phosphorus levels on number of branches per plant at 30, 60 DAS and at harvest in French bean cv. Arka Suvidha

Spacing (S)	Number of branches per plant											
	30 DAS				60 DAS				At harvest			
	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	4.64	5.00	5.13	4.92	6.23	6.78	8.03	7.02	6.57	7.81	8.70	7.69
S ₂ - 45 x 10 cm	4.77	4.96	5.33	5.02	6.61	7.32	9.45	7.79	7.24	8.11	10.36	8.57
S ₃ - 30 x 20 cm	4.83	5.10	5.32	5.08	8.99	9.18	9.88	9.35	10.18	10.50	11.08	10.59
S ₄ - 45 x 20 cm	4.62	5.48	5.49	5.20	8.69	9.78	10.41	9.63	10.83	11.16	12.20	11.40
Mean	4.71	5.13	5.32	5.05	7.63	8.26	9.44	8.45	8.71	9.39	10.59	9.56
For comparing means of	S.Em±			CD at 5%	S.Em±			CD at 5%	S.Em±			CD at 5%
S	0.10		NS		0.19		0.67		0.26		0.91	
P	0.07		0.21		0.13		0.38		0.21		0.62	
'P' at same 'S'	0.14		NS		0.25		0.76		0.42		NS	
'S' at same or diff. 'P'	0.15		NS		0.28		0.85		0.43		NS	

NS- Non significant

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

S- Spacing levels

at 60 DAS and at harvest. At 60 DAS, spacing of 45x20 cm (S₄) recorded the maximum branches (9.63) followed by 30x20 cm (S₃) (9.35) spacing which was on par with that of spacing. Minimum branches per plant (7.02) were recorded at spacing of 30x10 cm (S₁). Similar trend was also observed at harvest.

Phosphorus levels varied significantly at 30 DAS, 60 DAS and at harvest. At 60 DAS maximum branches (9.44/plant) were recorded at 125 kg P₂O₅ per ha (P₃) followed by 100 kg P₂O₅ per ha (P₂) (8.26) and least number of branches (7.63) were recorded at 75 kg P₂O₅ per ha (P₁). Similar trend was also observed at harvest.

The interaction of phosphorus levels at same spacing levels differed significantly at 60 DAS and non-significant at harvest. At 60 DAS, the P₃ at S₄ recorded maximum number of branches (10.41) and minimum branches at P₁ (6.23) at S₁ level.

The interaction of spacing at same or different phosphorus levels varied non significantly at 30 DAS. At 60 DAS branches were observed at P₃S₄ (10.41) followed by P₃S₃ (9.88) and minimum number of branches were recorded at P₁S₁ (6.23). The interaction was non-significant at harvest.

4.1.1.3 Number of leaves per plant

The observations on number of leaves per plant at 30 and 60 DAS are presented in Table 5.

The spacing levels varied significantly at 30 DAS. Maximum number of leaves per plant were observed at spacing 45x20 cm (8.49) followed by 30x20 (S₃) and minimum number of leaves per plant were recorded at 30x20 cm (7.54).

At 60 DAS 45x20 cm (S₄) recorded maximum number of leaves (13.58) followed by 30x20 cm (S₃) spacing (11.70), while 30x10 cm (S₁) spacing recorded the leaves per plant (9.82).

There was non-significant differences due to phosphorus levels at 30 DAS, whereas it was found significant at 60 DAS. Application of 125 kg P₂O₅ per ha (P₃) recorded maximum number of leaves (12.59) followed by 100 kg P₂O₅ per ha (11.89) and minimum number of leaves were observed at 75 kg P₂O₅ per ha (10.18).

The interaction of spacing and phosphorus levels did not vary significantly at 30 DAS for number of leaves, but varied significantly at 60 DAS. Application of 125 kg P per ha recorded maximum number of leaves (14.81) at spacing of 45x20 cm whereas 75 kg per ha at 30x10 cm spacing recorded minimum value (8.25). The results on leaves per plant in spacing at same or different levels of spacing, the combination of P₃S₄ recorded maximum number of leaves (14.81), which was on par with S₄P₂ (14.28) and minimum number of leaves were recorded at P₁S₁ (8.25) spacing.

4.1.1.4 Plant dry weight at harvest

The data on dry weight at harvest per plant are presented in Table 5.

There was significant differences among the spacing levels for plant dry weight at harvest. Spacing of 45x20 cm recorded maximum dry weight per plant (11.43 g) followed by spacing of 30x20 cm (9.99 g) and minimum dry weight per plant was recorded 30x10 cm (7.35 g).

The phosphorus levels varied significantly for plant dry weight at harvest. Significantly higher dry weight (11.42 g) was recorded at

Table 5. Effect of spacing and phosphorus levels on number of leaves per plant at 30, 60 DAS and plant dry weight at harvest in French bean cv. Arka Suvidha

Spacing (S)	Number of leaves per plant										Plant dry weight (g)				
	30 DAS					60 DAS					At harvest				
	P ₁	P ₂	P ₃	Mean	S.E.m _±	P ₁	P ₂	P ₃	Mean	S.E.m _±	P ₁	P ₂	P ₃	Mean	S.E.m _±
S ₁ -30 x 10 cm	7.37	7.58	7.66	7.54	0.10	8.25	9.87	11.35	9.82	0.35	5.42	7.56	9.07	7.35	0.25
S ₂ - 45 x 10 cm	7.96	7.83	7.91	7.90	0.09	9.59	11.70	12.03	11.11	0.17	7.27	8.73	9.76	8.59	0.13
S ₃ - 30 x 20 cm	8.31	8.09	7.63	8.01	0.18	11.20	11.73	12.17	11.70	0.35	7.66	9.66	12.67	9.99	0.27
S ₄ - 45 x 20 cm	8.30	8.54	8.61	8.49	0.17	11.67	14.28	14.81	13.58	0.45	9.24	10.86	14.18	11.43	0.33
Mean	7.99	8.01	7.95	7.98		10.18	11.89	12.59	11.55		7.40	9.20	11.42	9.34	
For comparing means of	S.E.m _±					S.E.m _±					S.E.m _±				
S	0.10					0.35					0.25				
P	0.09					0.17					0.13				
'p' at same 'S'	0.18					0.35					0.27				
'S' at same or diff. 'P'	0.17					0.45					0.33				
	CD at 5%					CD at 5%					CD at 5%				
	0.34					1.19					0.86				
	NS					0.52					0.40				
	NS					1.03					0.80				
	NS					1.34					0.99				

NS- Non significant

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

S- Spacing levels

125 kg per ha followed by 100 kg per ha (9.20 g) and least dry weight was seen at 75 kg P₂O₅ per ha (7.40 g).

The phosphorus levels at same spacing varied significantly. Application of 125 kg P₂O₅ per ha recorded maximum dry weight (14.18 g) at 45x20 cm whereas, 75 kg P₂O₅ per ha recorded least dry weight (5.42 g) at 30x10 cm.

The spacing levels at same or different levels of phosphorus varied significantly. Significantly higher dry weight was recorded at 125 kg per ha at 45x20 cm (14.18 g) followed by P₃S₃ (12.67 g), P₂S₄ (10.86 g) and the lowest dry weight was recorded at P₁S₁ (5.42 g/plant).

4.1.2 Yield parameters

4.1.2.1 Number of pods per plant

The results on number of pods per plant due to spacing and phosphorus levels are presented in Table 6 and depicted in Fig. 3.

Spacing levels varied significantly for number of pods per plant. The spacing of 45x20 cm recorded maximum number of pods per plant (15.34) followed by 30x20 cm (13.03) and minimum number of pods per plant was seen at 30x10 cm spacing (8.75).

There was significant difference among phosphorus levels for number of pods per plant. The application of 125 kg P₂O₅ per ha produced maximum pods (13.43/plant) followed by 100 kg (12.07) and least was observed at 75 kg P₂O₅ per ha (10.17/plant).

The interaction of phosphorus levels at same spacing also found to be significant. The P₃ at S₄ recorded maximum pods per plant (16.96)

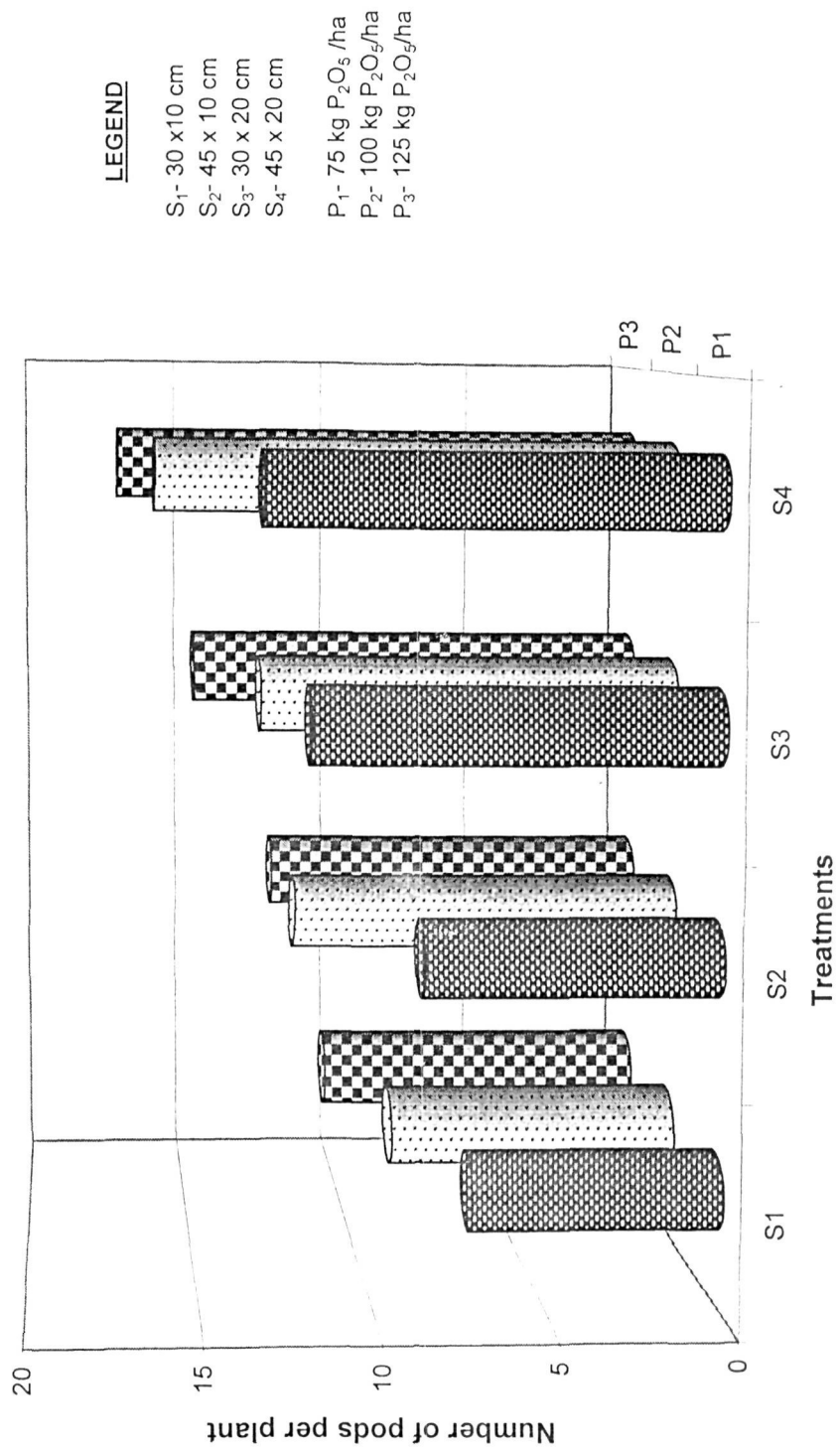


Fig. 3 : Effect of spacing and phosphorus levels on number of pods per plant

followed by P₂ at S₄ (15.96), P₃ at S₃ (14.51) and least was observed at P₁ at S₁ combination.

The interactions of spacing at same or different phosphorus levels were also significant. Maximum number of pods per plant recorded at P₃S₄ and least was seen at P₁S₁ (7.22/plant).

4.1.2.2 Pod weight (g)

The data on pod weight (g) as influenced by spacing and phosphorus levels are presented in Table 6.

The spacing levels varied significantly for individual pod weight. Maximum pod weight (2.44) was recorded at 45x20 cm spacing followed by 30x20 cm (2.32 g) and least pod weight was recorded at 30x10 cm spacing (2.28 g).

There was significant difference among the phosphorus levels for individual pod weight. Maximum pod weight of 2.40 g was observed at 125 kg P₂O₅ and minimum was observed at 75 kg P₂O₅ per ha (2.28 g).

Interaction of phosphorus levels at same spacing level found to be significant. The combination of P₃ at S₁, S₂, S₃ and S₄ recorded maximum pod weight of 2.34, 2.33, 2.42 and 2.99 g, respectively. Whereas P₁ at S₁, S₂, S₃ and S₄ recorded minimum pod weight of 2.24, 2.26, 2.23 and 2.39, respectively.

Interaction of spacing levels at same or different phosphorus level was found to be significant. The P₃S₄ recorded maximum pod weight of 2.49 g, which was on par with P₂S₄ (2.45 g) and P₃S₅ (2.42 g) and least pod weight of 2.24 g was recorded at P₁S₁.

4.1.2.3 Pod length (cm)

The observations on pod length (cm) at different spacing and phosphorus levels are presented in Table 6.

The spacing levels varied significantly for individual pod length. Maximum pod length (14.27 cm) was observed at 45x20 cm which was on par with 30x20 cm (13.86 cm) and minimum was seen at 30x10 cm spacing (12.99 cm).

There was significant difference among the phosphorus levels for pod length. Maximum pod length (13.99 cm) was observed with 125 kg P₂O₅ per ha and minimum pod length (12.98 cm) was observed at 75 kg P₂O₅ per ha.

The interaction between phosphorus levels at same spacing was found to be significant. Application of 125 kg P₂O₅ per ha recorded maximum of (14.73 cm), pod length at 45x20 cm spacing followed 75 kg P₂O₅ per ha recorded least pod length (13.70 cm) at 45x20 cm. The P₁ recorded least pod length (12.40 cm) at S₁.

The spacing at same or different phosphorus levels varied significantly. The combination of P₃S₄ recorded maximum pod length (14.73 cm), which was on par with P₂S₄ (14.37 cm), P₄S₃ (14.36 cm) and least pod length was observed at P₁S₁ (12.40 cm).

4.1.2.4 Number of seeds per pod

The data on the number of seeds per pod are presented in Table 7.

The spacing levels varied significantly for number of seeds per pod. The maximum number of seeds per pod (5.22) was observed at 45x20 cm

Table 6. Effect of spacing and phosphorus levels on number of pods per plant, weight per pod and pod length in French bean cv. Arka Suvidha

Spacings (S)	Number of pods per plant				Weight per pod (g)				Pod length (cm)			
	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	7.22	8.78	10.24	8.75	2.24	2.26	2.34	2.28	12.40	13.13	13.43	12.99
S ₂ - 45 x 10 cm	8.56	11.77	12.02	10.45	2.26	2.32	2.33	2.30	12.57	13.23	13.43	13.08
S ₃ - 30 x 20 cm	11.80	12.78	14.51	13.03	2.23	2.30	2.42	2.30	13.27	13.93	14.36	13.86
S ₄ - 45 x 20 cm	13.11	15.96	16.96	15.34	2.39	2.45	2.49	2.44	13.70	14.37	14.73	14.27
Mean	10.17	12.07	13.43	11.89	2.28	2.33	2.40	2.34	12.98	13.67	13.99	13.55
For comparing means of	S.E.m _±				S.E.m _±				S.E.m _±			
S	0.15				0.02				0.25			
P	0.19				0.01				0.09			
'P' at same 'S'	0.38				0.02				0.18			
'S' at same or diff. 'P'	0.34				0.03				0.29			
	CD at 5%				CD at 5%				CD at 5%			
	0.52				0.07				0.87			
	0.56				0.02				0.27			
	1.02				0.05				0.54			
	1.12				0.07				0.87			

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

S- Spacing levels

spacing by 45x10 cm (4.78), which was on par with 30x20 cm (4.77) and least number of seeds per pod (4.50) were observed at 30x10 spacing.

Significant difference among the phosphorus levels was recorded. Application of 125 kg P₂O₅ per ha recorded the highest number of seeds per pod (5.50) and 75 kg P₂O₅ per ha recorded least number of seeds per pod (4.44).

The interaction of phosphorus levels at same spacing was found to be significant. Application of 125 kg P₂O₅ per ha recorded maximum number of seeds (5.50) at 45x20 cm, whereas P₁ at S₄ recorded minimum number of seeds per pod (4.82).

The interaction among the spacing levels at same or different phosphorus levels were also varied significantly. The more number of seeds per pod (5.61) were recorded at S₃P₃, which was on par with S₄P₃ (5.50), S₄P₂ (5.23) and least numbers were recorded at S₁P₁ (4.25 /pod).

4.1.2.5 Seed weight per pod (g)

The results of seed weight per pod at different spacing and phosphorus levels are given in Table 7.

Spacing levels varied significantly. The highest seed weight per pod (1.74 g) was observed at 45x20 cm spacing followed by (1.65 g) at 30x20 cm and lowest was recorded at 30x10 cm (1.57 g).

Phosphorus levels also varied significantly for seed weight per pod. Maximum of 1.70 g was recorded at 125 kg P₂O₅ per ha and minimum of 1.56 g was seen at 75 kg P₂O₅ per ha.

There was significant difference among the phosphorus at same spacing. The P₃ recorded maximum seed weight per pod at S₁, S₂, S₃ and

S₄ (1.64, 16.3, 1.74 and 1.80 g, respectively) followed the same trend by P₂ at all 'S' levels and P₁ recorded minimum seed weight per pod at S₁, S₂, S₃ and S₄ (1.51, 1.48, 1.57 and 1.67 g/pod, respectively) spacing.

Spacing levels at same or different phosphorus levels interacted significantly. The P₃S₄ recorded maximum of 1.80, which was on par with P₂S₄ (1.74 g) and P₃S₃ (1.74 g) and least seed weight per pod was recorded at P₁S₁ (1.51 g).

4.1.2.6 Seed weight per plant (g)

The data on seed weight per plant as influenced by spacing and phosphorus levels are presented in Table 7.

Spacing levels varied significantly for the seed weight per plant. The spacing of 45x20 cm recorded maximum seed weight per plant (26.14 g), followed by 30x20 cm (22.14 g) and least seed weight per plant was recorded at 30x10 cm spacing (13.58 g).

There was significant difference among the phosphorus levels. Significantly highest seed weight per plant was recorded at 125 kg P₂O₅ per ha (21.33 g) and minimum was observed at 75 kg P₂O₅ per ha (17.70 g).

The interaction among the phosphorus levels at same spacing level was differed significantly. The P₃ at S₁, S₂, S₃ and S₄ recorded maximum seed weight per plant, whereas P₁ at S₁, S₂, S₃ and S₄ recorded minimum seed weight per plant.

Spacing levels at same or different phosphorus levels differed significantly. The P₃S₄ recorded the highest seed weight (26.90 g) and

Table 7. Effect of spacing and phosphorus levels on number of seeds per pod, seed weight per pod and seed weight per plant in French bean cv. Arka Suvidha

Spacings (S)	Number of seeds per pod				Seed weight per pod (g)				Seed weight per plant (g)			
	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	4.25	4.49	4.75	4.50	1.51	1.55	1.64	1.57	12.13	12.90	15.70	13.58
S ₂ - 45 x 10 cm	4.41	4.75	5.17	4.78	1.48	1.56	1.63	1.56	12.60	16.50	19.60	16.23
S ₃ - 30 x 20 cm	4.26	4.43	5.61	4.77	1.57	1.64	1.74	1.65	20.77	22.57	23.10	22.14
S ₄ - 45 x 20 cm	4.82	5.23	5.50	5.22	1.67	1.74	1.80	1.74	25.30	26.23	26.90	26.14
Mean	4.44	5.28	5.50	4.81	1.56	1.63	1.70	1.63	17.70	19.55	21.33	19.53
For comparing means of	S.Em±				S.Em±				S.Em±			
S	0.07				0.01				0.14			
P	0.07				0.01				0.17			
'P' at same 'S'	0.13				0.02				0.34			
'S' at same or diff. 'P'	0.13				0.02				0.31			
	CD at 5%				CD at 5%				CD at 5%			
	0.24				0.04				0.47			
	0.20				0.03				0.51			
	0.39				0.06				0.92			
	0.40				0.07				1.02			

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

S- Spacing levels

minimum seed weight per plant was seen in P_1S_1 combination (12.13 g).

4.1.2.7 Seed yield per plot (kg)

The data on seed yield per plot differed due to spacing and phosphorus levels are presented in Table 8.

There was significant differences among the spacing levels. The maximum seed yield per plot (3.84 kg) was recorded at spacing of 30x20 cm, which was on par with 30x10 cm i.e. (3.67 kg) and minimum seed yield per plot was recorded at 30x20 cm spacing (3.26 kg).

Phosphorus levels were also varied significantly. Application of 125 kg P_2O_5 per ha recorded maximum seed yield per plot (3.88 kg) and minimum at 75 kg P_2O_5 per ha (3.20 kg).

The interaction of phosphorus level at same spacing level was found to be significant. The combination of P_3 at S_1 , S_2 , S_3 and S_4 recorded maximum seed yield of 4.06, 3.51, 4.41 and 3.52 kg per plot. While the P_1 recorded minimum yield at all spacing levels.

The interaction between spacing levels at same or different phosphorus levels showed significant values. Maximum seed yield per plot was recorded at P_3S_3 (4.41 kg), which is on par with P_3S_1 (4.06 kg) and minimum was seen at P_1S_2 (3.02 kg).

4.1.2.8 Seed yield per ha (kg)

The data on seed yield per ha as influenced by spacing and phosphorus levels are presented in Table 8 and depicted in Fig. 4.

The spacing levels varied significantly for seed yield per ha. The highest seed yield was obtained at 30x20 cm spacing (2138 kg/ha), which

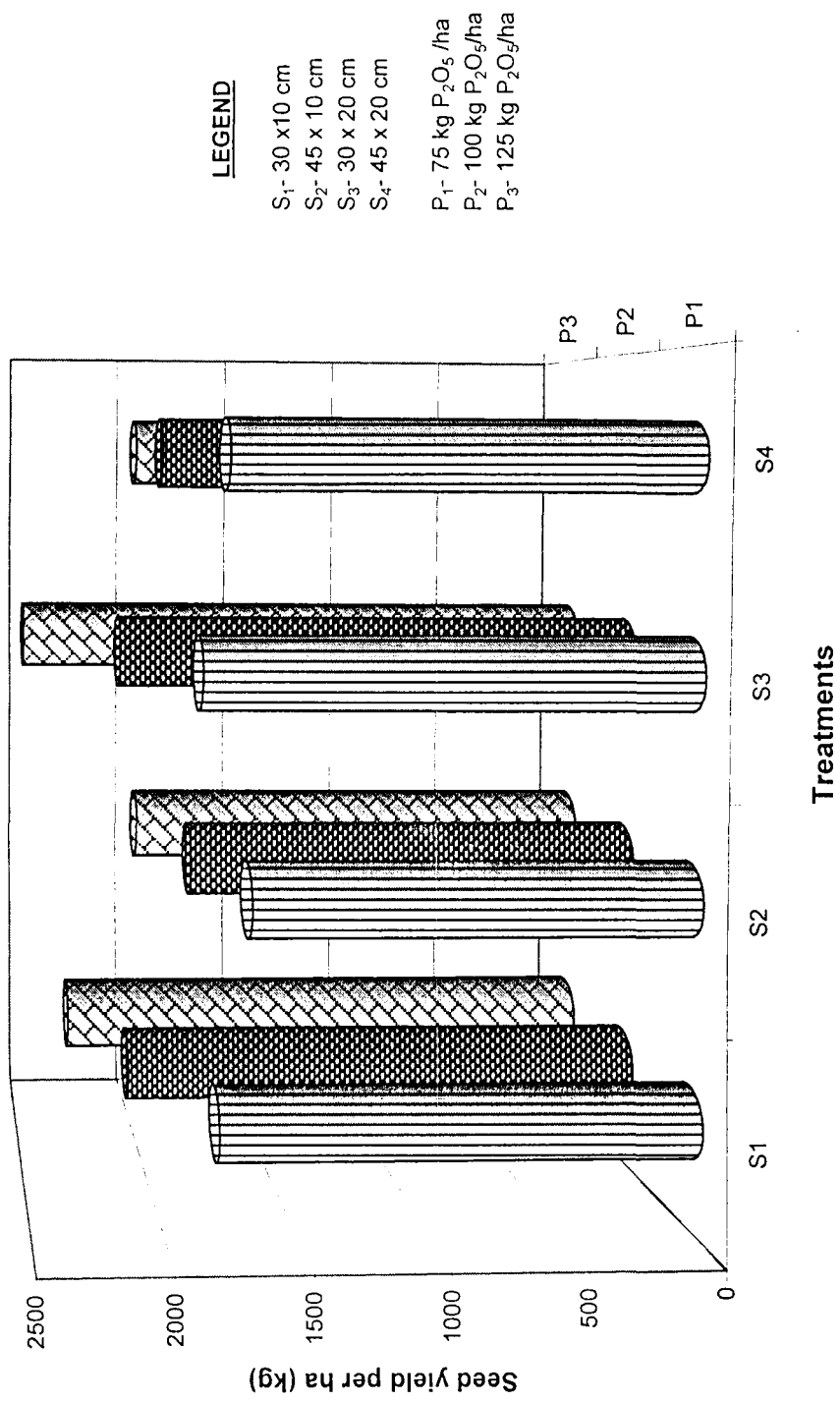


Fig. 4 : Effect of spacing and phosphorus levels on seed yield per ha

was on par with 30x10 cm (2043 kg/ha) and minimum was noticed at 30x20 cm (1813 kg/ha).

There was significant difference were observed among the phosphorus levels for seed yield per ha. The application of P_2O_5 125 kg per ha recorded maximum seed yield 2157 kg per ha and minimum yield per ha was noticed at 75 kg P_2O_5 per ha (1779 kg/ha).

The interaction among the phosphorus levels at same spacing found to be significant. The P_3 at S_3 recorded maximum of 2454 kg per ha and P_1 at S_3 recorded minimum of 1863 kg per ha.

The interaction between spacing levels at same or different phosphorus levels differed significantly. The P_3S_3 recorded the maximum seed yield per ha (2454 kg), which was on par with P_3S_1 (2259 kg/ha) and minimum seed yield was recorded P_1S_2 (1680 kg/ha).

4.1.2.9 Seed recovery (%)

The data on seed recovery per cent are presented in Table 8.

Spacing levels did not vary significantly, whereas phosphorus levels differed significantly for seed recovery per cent. Application of 125 kg P_2O_5 per ha recorded maximum seed recovery per cent (94.69) and minimum was obtained at 75 kg P_2O_5 per ha (91.62).

The interaction of phosphorus and spacing levels found to be non-significant.

Table 8. Effect of spacing and phosphorus levels on seed yield per plot, seed yield per ha and seed recovery (%) in French bean cv. Arka Suvidha

Spacing (S)	Seed yield per plot (kg)				Seed yield per ha (kg)				Seed recovery (%)			
	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	3.24	3.72	4.06	3.67	1805	2067	2259	2043	91.13	93.31	94.50	92.98
S ₂ - 45 x 10 cm	3.02	3.24	3.51	3.26	1680	1807	1953	1813	91.30	92.69	94.26	92.75
S ₃ - 30 x 20 cm	3.35	3.77	4.41	3.84	1863	2097	2454	2138	91.95	92.97	95.44	93.45
S ₄ - 45 x 20 cm	3.18	3.47	3.52	3.39	1768	1930	1961	1886	92.09	92.50	94.52	93.03
Mean	3.20	3.55	3.88	3.54	1779	1975	2157	1971	91.62	92.87	94.69	93.10
For comparing means of	S.Em±				S.Em±				S.Em±			
S	0.07		0.20		51.02		156.03		0.43		NS	
P	0.05		0.14		25.29		75.69		0.30		0.91	
'P' at same 'S'	0.09		0.28		50.51		151.38		0.61		NS	
'S' at same or diff. 'P'	0.17		0.50		92.63		277.58		0.66		NS	

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

S- Spacing levels

4.1.3 Quality parameters

4.1.3.1 100 seed weight (g)

The results on 100 seed weight as influenced by spacing and phosphorus levels are presented in Table 9 and depicted in Fig. 5.

There was significant difference among the spacing levels for 100 seed weight. The spacing of 45x20 cm recorded maximum 100 seed weight of 36.23 g, which was on par with 30x20 cm (35.61 g) and minimum 100 seed weight of 33.60 g was recorded at 30x10 cm spacing.

Phosphorus levels showed significant difference for 100 seed weight. Application of 125 kg P₂O₅ per ha recorded maximum 100 seed weight (36.89 g) and minimum 100 seed weight (33.45 g) was obtained at 25 kg P₂O₅ per ha.

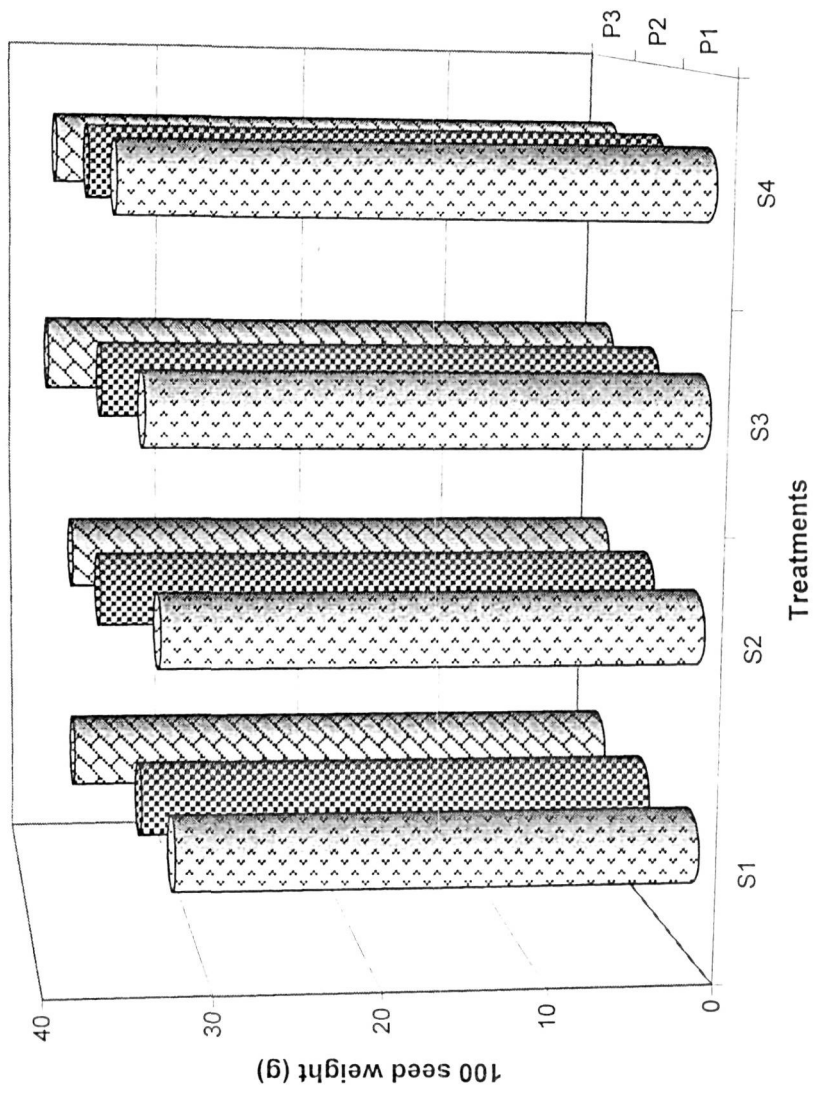
The interaction among the phosphorus levels at same spacing was found to be significant. The P₃ at S₄, P₃ at S₃ and P₃ at S₁ recorded maximum 100 seed weight of 37.30, 37.83 and 36.17 g, respectively.

The interaction between spacing levels at same or different phosphorus levels was found to be significant. The maximum 100 seed weight (37.83 g) was observed at P₃S₃, which was on par with P₃S₄ (37.30 g) and minimum 100 seed weight was noticed at P₁S₁ (31.87 g).

4.1.3.2 Seed germination per cent

The data on germination per cent as influenced by different spacing and phosphorus levels are presented in Table 9 and depicted in Fig. 6.

Spacing levels varied significantly. The spacing of 30x20 cm recorded maximum germination per cent (92.08), which was on par with



LEGEND

- S₁- 30 x 10 cm
- S₂- 45 x 10 cm
- S₃- 30 x 20 cm
- S₄- 45 x 20 cm
- P₁- 75 kg P₂O₅ /ha
- P₂- 100 kg P₂O₅/ha
- P₃- 125 kg P₂O₅/ha

Fig. 5 : Effect of spacing and phosphorus levels on hundred seed weight

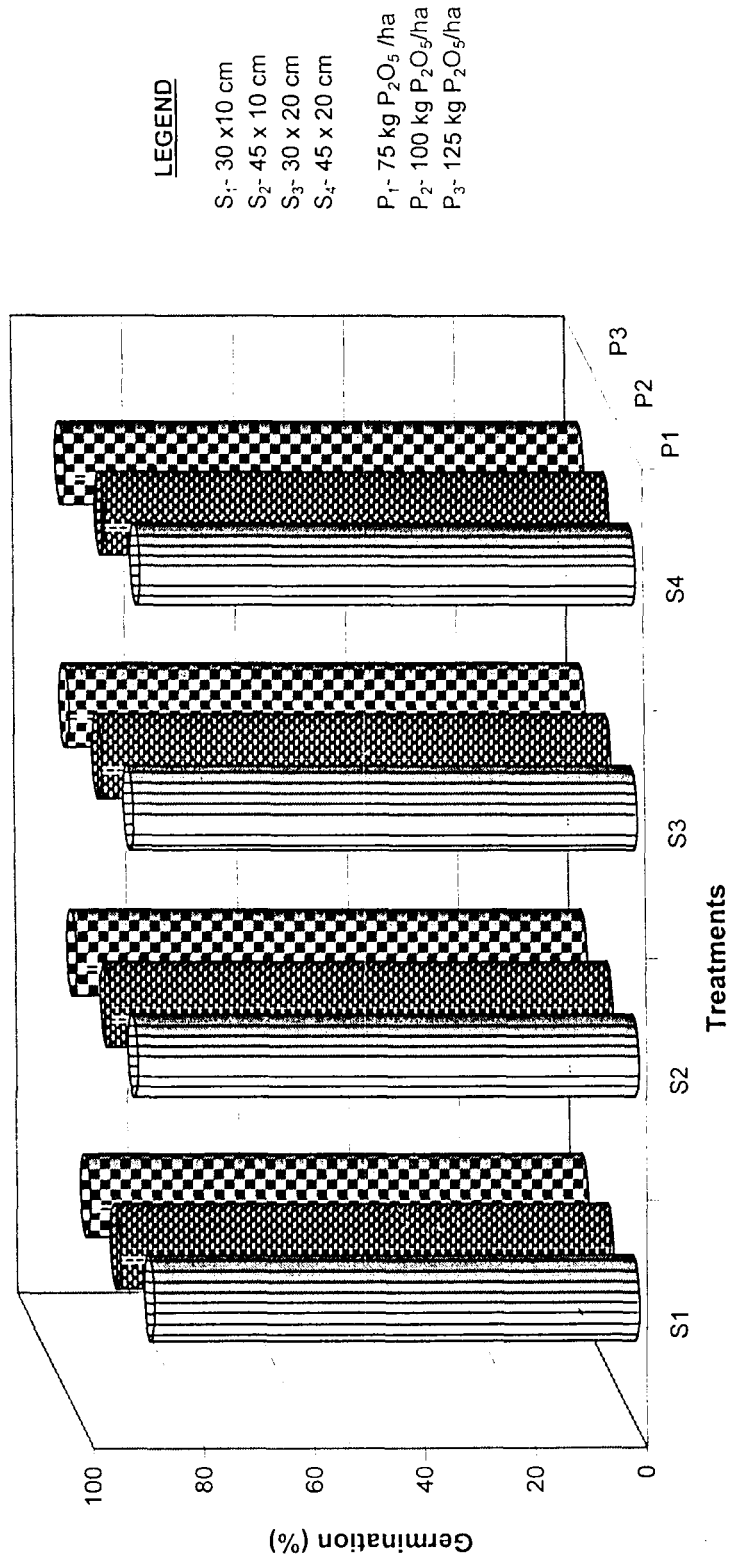


Fig. 6 : Effect of spacing and phosphorus levels on germination percentage

45x20 cm (91.50%) and 45x10 cm (91.16%). The least per cent of germination was recorded at 30x10 cm spacing (89.00%).

There was significant difference among the phosphorus levels for germination per cent. Application of 125 kg P_2O_5 per ha recorded maximum of 92.31 per cent and minimum was noticed at 75 kg P_2O_5 per ha (89.68%).

The interaction between phosphorus levels at same spacing differed significantly. The P_3 at S_4 recorded maximum germination per cent (93.75%) and P_1S_1 recorded minimum germination per cent (87.75%).

The interaction among the spacing levels at same or different phosphorus levels were significant. The P_3S_4 recorded maximum germination per cent (93.75%), which was on par with P_2S_5 (91.25%), P_3S_3 (93.25%) and P_2S_3 (92.00%) and minimum germination percentage was recorded at P_1S_1 (87.75%).

4.1.3.3 Rate of germination

The data on rate of germination are presented in Table 9.

There was a non-significant difference among the spacing levels.

Phosphorus levels varied significantly for rate of germination. Significantly highest rate of germination (31.12) was found at 125 kg P_2O_5 per ha, followed by 100 kg P_2O_5 per ha (29.50) and minimum was obtained at 75 kg P_2O_5 per ha (28.53).

The interaction of phosphorus and spacing levels found to be non-significant.

Table 9. Effect of spacing and phosphorus levels on 100 seed weight, seed germination per cent and rate of germination in French bean cv. Arka Suvidha

Spacing (S)	100 seed weight (g)				Seed germination (%)				Rate of germination			
	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	31.87	32.77	36.17	33.60	87.75* (69.49)	89.25 (70.81)	90.00 (71.56)	89.00 (70.63)	26.52	29.23	31.52	29.09
S ₂ - 45 x 10 cm	32.80	35.40	36.27	34.92	90.50 (72.08)	90.75 (72.31)	92.25 (73.90)	91.16 (72.76)	28.15	29.00	30.26	29.13
S ₃ - 30 x 20 cm	33.73	35.26	37.83	35.61	91.00 (72.61)	92.00 (73.63)	93.25 (75.01)	92.08 (73.75)	29.14	30.60	31.22	30.32
S ₄ - 45 x 20 cm	35.40	36.10	37.30	36.23	89.50 (71.09)	91.25 (72.74)	93.75 (75.46)	91.50 (73.05)	30.34	29.20	31.50	30.34
Mean	33.45	34.88	36.89	35.90	89.68 (71.16)	90.81 (72.34)	92.31 (73.89)	90.43 (72.44)	28.54	29.50	31.12	29.72
For comparing means of	S.Em±				S.Em±				S.Em±			
S	0.20				0.60				0.48			
P	0.18				0.43				0.41			
'P' at same 'S'	0.36				0.86				0.82			
'S' at same or diff. 'P'	0.35				0.92				0.82			
	CD at 5%				CD at 5%				CD at 5%			
	0.69				1.90				NS			
	0.54				1.25				1.21			
	1.06				2.50				NS			
	1.07				2.68				NS			

NS- Non significant

* Figures in the parenthesis are arc sine transformed values

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

S- Spacing levels

4.1.3.4 Per cent field emergence

The data on per cent field emergence as influenced by spacing and phosphorus levels are given in Table 10 and depicted in Fig. 7.

Spacing levels did not vary significantly. While phosphorus levels varied significantly for per cent of field emergence. Application of 125 kg P_2O_5 per ha recorded the highest per cent field emergence (89.62) whereas 75 kg P_2O_5 per ha recorded minimum field emergence of 84.37 per cent.

The interaction between spacing and phosphorus levels did not vary significantly.

4.1.3.5 Seedling dry weight (mg)

The results of seedling dry weight as influenced by spacing and phosphorus levels are presented in Table 10.

Spacing levels varied significantly for seedling dry weight. The maximum seedling dry weight (27.4 mg) was recorded at 45x20 cm spacing, which was on par with 30x20 cm (26.4 mg) and least was noticed at 30x10 cm spacing (25.2 mg).

Phosphorus levels also varied significantly. Maximum seedling dry weight (27.1 mg) was recorded at 125 kg phosphorus per ha but minimum was obtained at 75 kg phosphorus per ha (25.0 mg).

The interaction among the phosphorus levels at same spacing found to be significant. The P_3 and P_1 recorded maximum and minimum seedling dry weight at S_4 , S_3 , S_2 and S_1 , respectively.

The interaction between spacing levels at same or different phosphorus levels were significant. The P_3S_4 recorded maximum

seedling dry weight (27.9 mg), which was on par with P_3S_3 (27.9 mg), P_2S_4 (27.5 mg) and P_2S_3 (27.0 mg).

4.1.3.6 Electrical conductivity of seed leachate (dSm^{-1})

The data on electrical conductivity as influenced by spacing and phosphorus levels are shown in Table 10.

Spacing levels recorded significant differences for Electrical conductivity of seed leachate. The least value for Electrical conductivity of seed leachate was recorded at 45x20 cm spacing ($1.209 dSm^{-1}$) and maximum was seen at 30x10 cm spacing ($1.314 dSm^{-1}$).

Phosphorus levels varied significantly for Electrical conductivity values of seed leachate. Application of 125 kg P_2O_5 per ha recorded least value of Electrical conductivity ($1.148 dSm^{-1}$) and highest Electrical conductivity was noticed at 75 kg P_2O_5 per ha ($1.398 dSm^{-1}$).

The interaction between phosphorus levels at same spacing varied significantly. The P_3 at S_4 , S_3 , S_2 and S_1 recorded least values for Electrical conductivity of seed leachate.

The interaction among the spacing levels at same or different phosphorus levels found to be significant. The P_3S_4 recorded the least value for Electrical conductivity values of $1.127 dSm^{-1}$ and maximum value was noticed in P_1S_1 ($1.532 dSm^{-1}$) combination.

4.1.3.7 Root length (cm)

The data on seedling root length (cm) are presented in Table 11

Spacing and phosphorus levels varied non-significantly for seedling root length. However, the higher root length was recorded at S_1 (12.07 cm) and minimum was observed in S_4 (11.64). Among the phosphorus levels

Table 10. Effect of spacing and phosphorus levels on per cent field emergence, seedling dry weight and electrical conductivity in French bean cv. Arka Suvidha

Spacing (S)	Per cent field emergence				Seedling dry weight (mg)				Electrical conductivity (dSm ⁻¹)			
	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	83.50 (66.04)*	88.00 (69.75)	88.50 (70.29)	86.66 (68.69)	24.1	25.3	26.3	25.2	1.532	1.244	1.166	1.314
S ₂ - 45 x 10 cm	79.50 (63.16)	88.50 (70.22)	90.00 (71.77)	86.00 (68.38)	25.3	25.5	26.4	25.6	1.429	1.296	1.151	1.292
S ₃ - 30 x 20 cm	87.00 (68.92)	86.00 (68.35)	89.50 (71.14)	87.50 (69.47)	24.4	27.0	27.9	26.4	1.341	1.308	1.151	1.266
S ₄ - 45 x 20 cm	87.50 (69.30)	86.00 (68.08)	90.50 (72.10)	88.00 (69.83)	26.7	27.5	27.9	27.4	1.290	1.212	1.127	1.209
Mean	84.37 (66.85)	87.12 (69.10)	89.62 (71.32)	87.03 (69.08)	25.0	26.3	27.1	26.1	1.398	1.268	1.148	1.271
For comparing means of	S.Em±				S.Em±				S.Em±			
S	1.50				0.32				0.020			
P	0.94				0.07				0.020			
'P' at same 'S'	1.88				0.15				0.030			
'S' at same or diff. 'P'	2.15				0.35				0.036			
	CD at 5%				CD at 5%				CD at 5%			
	NS				1.12				0.060			
	3.07				0.23				0.050			
	NS				0.45				0.090			
	NS				1.04				0.100			

NS- Non significant

* Figures in the parenthesis are arc sine transformed values

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

S- Spacing levels

P₃ produced highest root length (12.31 cm), lowest was noticed at P₁ (11.18 cm) and interactions were also found to be non significant.

4.1.3.8 Shoot length (cm)

The data on seedling shoot length (cm) are presented in Table 11.

The spacing levels did not varied significantly whereas, phosphorus levels varied significantly for shoot length. Application of 125 kg P₂O₅ per ha recorded maximum shoot length (21.52 cm) and minimum was seen in 75 kg P₂O₅ per ha (18.51 cm).

The interaction between spacing and phosphorus levels were non-significant.

4.1.3.9 Seedling vigour index (SVI)

The data revealing seed vigour index of French bean as influenced by spacing and phosphorus levels are presented in Table 11 and depicted in Fig. 7.

The spacing levels did not differ significantly for seed vigour index. Phosphorus levels showed significant difference for seed vigour index. The application of 125 kg P₂O₅ per ha recorded maximum seed vigour index per ha (3122), which was on par with 100 kg phosphorus per ha (2951) and least seed vigour index was obtained at 75 kg P₂O₅ per ha (2661). Interaction between 'P' levels and 'S' levels found to be non-significant.

4.1.3.10 Seed protein per cent

The data on seed protein per cent as influenced by spacing and phosphorus are presented in Table 12.

Table 11. Effect of spacing and phosphorus levels on root length, shoot length and seedling vigour index in French bean cv. Arka Suvidha

Spacing (S)	Root length (cm)				Shoot length (cm)				Seedling vigour index (SVI)			
	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	11.23	12.33	12.73	12.07	18.20	19.60	19.90	19.23	2582	2840	2936	2786
S ₂ - 45 x 10 cm	11.00	11.77	12.33	11.70	18.83	22.00	23.23	21.35	2699	3064	3280	3014
S ₃ - 30 x 20 cm	11.07	11.60	12.37	11.68	18.03	20.47	21.60	20.03	2648	2950	3167	2921
S ₄ - 45 x 20 cm	11.40	11.73	11.80	11.64	18.97	20.60	21.33	20.30	2718	2951	3105	2924
Mean	1.18	11.83	12.31	11.77	18.51	20.67	21.52	20.23	2661	2951	3122	2911
For comparing means of	S.Em±				S.Em±				S.Em±			
S	0.38				0.65				70.00			
P	0.40				0.61				67.00			
'P' at same 'S'	0.80				1.21				135.00			
'S' at same or diff. 'P'	0.76				1.19				130.00			
	CD at 5%				CD at 5%				CD at 5%			
	NS				NS				NS			
	NS				1.82				202.00			
	NS				NS				NS			
	NS				NS				NS			

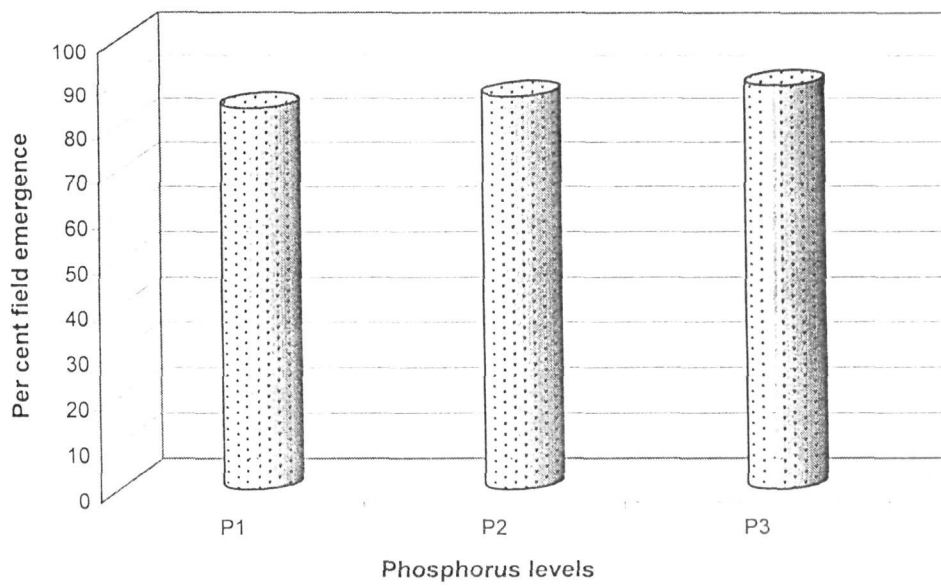
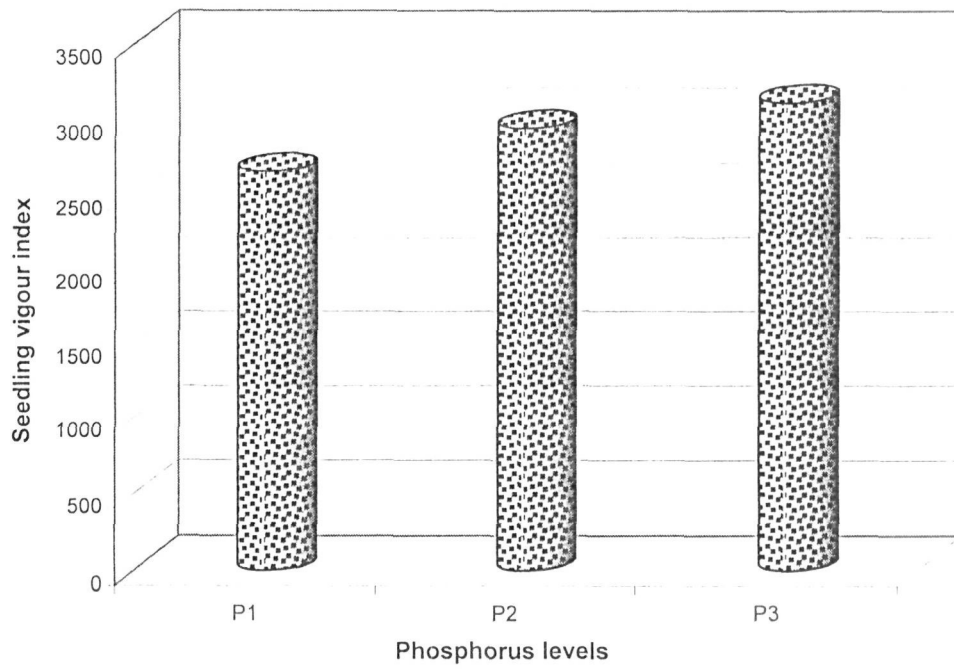
NS- Non significant

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha

P₂- 100 kg P₂O₅/ha

P₃- 125 kg P₂O₅/ha

S- Spacing levels



P₁- 75 kg P₂O₅ /ha P₂- 100 kg P₂O₅/ha P₃- 125 kg P₂O₅/ha

Fig. 7 : Effect of phosphorus levels on seedling vigour index and per cent field emergence

Table 12. Effect of spacing and phosphorus levels on seed protein per cent in French bean cv. Arka Suvidha

Spacing (S)	Seed protein per cent			
	P ₁	P ₂	P ₃	Mean
S ₁ -30 x 10 cm	17.83	18.43	19.37	18.54
S ₂ - 45 x 10 cm	18.97	19.43	20.83	19.74
S ₃ - 30 x 20 cm	21.33	22.53	22.73	22.20
S ₄ - 45 x 20 cm	23.13	23.50	24.17	23.60
Mean	20.32	20.98	21.76	21.02
For comparing means of	S.Em _±		CD at 5%	
S	0.14		0.47	
P	0.09		0.26	
'P' at same 'S'	0.17		0.51	
'S' at same & diff. 'P'	0.20		0.59	

P - Phosphorus levels : P₁- 75 kg P₂O₅ /ha
P₂- 100 kg P₂O₅/ha
P₃- 125 kg P₂O₅/ha

S- Spacing levels

Spacing levels varied significantly for seed protein per cent. The maximum seed protein per cent was recorded at 45x20 cm (23.60) followed by 30x20 cm (22.20%) and minimum was noticed at 30x10 cm spacing (18.54%).

There was significant difference among the phosphorus levels for seed protein per cent. Application of 125 kg P₂O₅ per ha recorded maximum of 21.76 per cent and minimum of 20.32 per cent found in the seeds harvested from the treatment of 75 kg P₂O₅ per ha.

The interaction among the phosphorus levels at same spacing found to be significant. The P₃ recorded maximum protein per cent at S₁, S₂, S₃ and S₄ spacing (19.37 ; 20.83, 22.73 and 24.17%, respectively),

The interaction between spacing levels at same or different phosphorus levels were found to be significant. The P₃S₄ recorded maximum seed protein per cent (24.17), followed by P₂S₄ (23.50) and minimum was recorded at P₁S₁ (17.83%) combination.

4.2 EXPERIMENT- II : SCREENING OF FRENCH BEAN VARIETIES FOR SEED YIELD AND QUALITY PARAMETERS

4.2.1 Growth parameters

4.2.1.1 Days to 50 per cent flowering

The data on days to 50 per cent flowering in french bean varieties are presented in Table 13.

Contender took least days to 50 per cent flowering (37-38), whereas MFB-3 recorded maximum days (41.42) to 50 per cent flowering.

4.2.1.2 Number of branches per plant

The data on number of branches per plant in french bean varieties are presented in Table 13.

Branches per plant varied significantly among the varieties. RSJ-288 recorded maximum number of branches (11.20 /plant) which is followed by IIHR-909 and least was observed with variety Arka Komal and MFB-2 (6.0). Irrespective of varieties, the mean number of branches per plant was 7.85.

4.2.2 Yield parameters

4.2.2.1 Number of pods per plant

The data on number of pods per plant at different french bean varieties are presented in Table 13.

The varieties varied significantly for number of pods per plant. RSJ-288 variety recorded maximum pods per plant (12.00), followed by MFB-3 (10.33) Contender (10.20), MFB-1 (10.00) and IIHR-909 (9.60) which are on par with each other. The least number of pods per plant was observed with the variety Arka Komal (7.66). Irrespective of varieties, the average number of pods per plant was 9.68.

4.2.2.2 Number of seeds per pod

The data on number of seeds per pod in different varieties are presented in Table 13.

IIHR-909 recorded maximum seeds per pod (6.40) followed by MFB-2 (5.13), MFB-3 (4.86) and RSJ-288 (4.60) are on par with each other. The least number was observed in MFB-I (4.40). The overall average value in the varieties is 4.96.

4.2.2.3 Pod weight (g)

The data on pod weight of different varieties of french bean are given in Table 13.

IIHR-909 recorded maximum pod weight (2.56 g) followed by Contender (2.38 g) and RSJ-288 (2.31 g), which were on par with each other. The lowest pod weight (1.71 g) is recorded with MFB-I.

4.2.2.4 Seed weight per plant (g)

Seed weight per plant in different french bean varieties are presented in Table 13.

The varieties are grouped into three classes as low (<10.0 g), medium (10.0 to 15.0 g) and high (>15.0 g) based on seed weight per plant.

Varieties were grouped under low seed weight per plant are MFB-2 (8.26 g), Arka Komal (8.70 g) and MFB-1 (9.86 g), medium class is MFB-3 (12.73 g) variety whereas RSJ-288 (18.90 g), Contender (16.23 g), IIHR-909 (15.26 g) are grouped under high seed weight per plant category. The mean seed weight per plant was 12.84 g.

4.2.3 Seed quality parameters

4.2.3.1 100 seed weight

Hundred seed weight varied significantly due to varieties (Table 14).

RSJ-288 recorded significantly highest 100 seed weight (38.93 g) followed by Contender (35.20 g), IIHR-909 (27.37 g) and MFB-2 (26.33 g).

Table 13. Yield parameters of different french bean varieties

Varieties	Days to 50% flowering	Branches per plant	Number of pods per plant	Number of seeds per pod	Pod weight (g)	Seed weight per plant (g)	Class
1. RSJ-288	39-40	11.20	12.0	4.60	2.31	18.90	High
2. IHR-909	37-39	10.20	9.60	6.40	2.56	15.26	High
3. MFB-1	40-41	7.80	10.00	4.40	1.71	9.86	Low
4. Contender	37-38	6.20	10.20	4.53	2.38	16.13	High
5. Arka Komal	38-41	6.00	7.66	4.80	1.75	8.70	Low
6. MFB-2	39-42	6.00	7.93	5.13	2.12	8.26	Low
7. MFB-3	41-42	7.60	10.33	4.86	1.90	12.73	Medium
Mean		7.85	9.68	4.96	2.11	12.84	<10.0 g Low
S.E.m _t		0.082	0.344	0.187	0.024	0.289	10.0-15.0 g Medium
CD at 5%		0.868	1.06	0.578	0.073	0.891	> 15.0 g High

The lowest hundred seed weight was observed with the variety of MFB-3 (21.00 g).

Irrespective of varieties, the mean 100 seed weight was 27.30 g.

4.2.3.2 Germination per cent

The data on germination per cent of the varieties are presented in Table 14.

The germination per cent varied non-significantly among the varieties.

4.2.3.3 Root length (cm)

The root length varied non-significantly among the varieties, the data of which is presented in Table 14.

However, the higher root length was recorded with MFB-3 (13.70 cm) and lowest was seen in Contender (11.65 cm).

4.2.3.4 Shoot length (cm)

Shoot length varied non-significantly due to varieties, the data of which is presented in Table 14.

However, the higher shoot length was recorded with Arka Komal (20.37 cm) and lowest was seen in MFB-1 (15.62 cm).

4.2.3.5 Seedling vigour index

There was non-significant difference among the varieties for vigour index, the data of which is presented in Table 14.

However, the higher seedling vigour index was recorded with MFB-3 (3023) and lowest was seen in IIHR-909 (2463).

4.2.3.6 Seedling dry weight (mg)

Seedling dry weight varied significantly among the varieties (Table 14).

Variety RSJ-288 recorded significantly highest seedling dry weight (283.3 mg) followed by IIHR-909 (24.43 mg) and Contender (23.53 mg), which were on par with each other. The least dry weight was recorded in the variety MFB-1 (11.30 mg). The average dry weight among the varieties was 19.91 mg.

4.2.3.7 Electrical conductivity (dSm^{-1}) of seed leachate

The data on electrical conductivity of french bean variety are presented in Table 14.

The lowest value for electrical conductivity (0.696 dSm^{-1}) was recorded in the variety RSJ-288 followed by Contender (1.073), IIHR-909 (1.096) and the maximum value was seen in MFB-2 (2.360 dSm^{-1}). The average values of electrical conductivity was (1.40 dSm^{-1}).

4.2.3.8 Protein per cent

The data on protein per cent of different french bean varieties are presented in Table 14.

MFB-2 recorded maximum protein per cent (22.06), followed by Contender (18.6), RSJ-288 (18.63) which are on par with each other. The least per cent of protein is recorded in variety MFB-3 (16.03).

Table 14. Seed quality parameters of french bean varieties

Varieties	100 seed weight (g)	Germination (%)	Root length (cm)	Shoot length (cm)	Vigour index	Seedling dry weight (mg)	EC (dSm ⁻¹)	Protein per cent
1. RSJ-288	38.93	90.25 (73.55)*	13.00	16.95	2750	28.33	0.696	18.63
2. IHR-909	27.37	88.25 (70.09)	12.45	15.65	2463	24.43	1.096	16.83
3. MFB-1	20.63	92.00 (73.64)	12.97	15.62	2633	11.30	1.459	17.53
4. Contender	35.20	88.25 (72.83)	11.65	17.50	2662	23.53	1.073	18.76
5. Arka Komal	21.70	92.25 (73.90)	12.00	20.37	2984	17.67	1.493	16.23
6. MFB-2	26.33	88.00 (69.94)	13.25	15.82	2562	19.27	2.360	22.06
7. MFB-3	21.00	92.25 (73.90)	13.70	19.10	3023	14.63	1.617	15.03
Mean	27.30	87.90 (72.55)	12.72	17.29	2726	19.91	1.40	18.01
S.Em _±	0.69	1.37	0.78	1.66	176	0.83	0.04	0.26
CD at 5%	2.13	NS	NS	NS	NS	2.54	0.12	0.82

* Figures in the parenthesis are arc sine transformed values

4.3 EXPERIMENT – III : VARIETAL IDENTIFICATION OF FRENCH BEAN CULTIVARS

The results of morphological and electrophoretic identification of french bean varieties are presented below.

4.3.1 Morphological characters

4.3.1.1 Seed morphology

Seed characters like seed coat colour, hilum colour, seed length, seed size, seed shape and 100 seed weight were studied and varieties were grouped. The details of the specific variety with respect to characters are presented in different Plates.

4.3.1.1.1 Seed coat colour

Based on the seed coat colour, the seven varieties were grouped individually as Black, Light brown, creamy white, Pale brown, Brown, Mottled and Dark brown, which are presented in Table 15 and depicted in Plate 2.

4.3.1.1.2 Seed hilum colour

All the seven varieties under study were recorded with white hilum colour and presented in Table 15.

4.3.1.1.3 Seed shape

The different french bean varieties based on seed shape were grouped into cylinder, kidney and oval on visual basis and are presented in Table 15.

Table 15. Seed morphological characters of french bean varieties

Varieties	Seed coat colour	Hilum colour	Seed shape
1. RSJ-288	Black	White	Cylinder
2. IIHR-909	Light brown	White	Kidney
3. MFB-1	Creamy white	White	Oval
4. Contender	Pale brown	White	Kidney
5. Arka Komal	Brown	White	Kidney
6. MFB-2	Mottled	White	Oval
7. MFB-3	Dark brown	White	Oval

Varieties IIHR 909, Contender, Arka Komal were grouped under kidney shape, MFB-1, MFB-2 and MFB-3 had oval shape, where as RSJ-288 found to be cylinder in shape.

4.3.1.1.4 Seed length (mm)

The data on seed length recorded in millimeter of seven varieties of frenchbean are presented in Table 16 and are grouped into small (10.0 to 11.5 mm), medium (11.5 to 13.0 mm) and large (>13.0 mm).

Varieties RSJ-288, Contender and MFB-2 were categorised in to large; IIHR-909 was of medium length whereas MFB-1, Arka Komal and MFB-3 were under the category of small. Irrespective of varieties, the mean seed length was 12.59 mm.

4.3.1.1.5 Seed girth (mm)

Seed girth varied with varieties and are grouped as low (13.5 to 16.0 mm), medium (16.0 to 18.5 mm) and high (> 18.5 mm), and are presented in Table 16.

The varieties *viz.*, MFB-1, Arka Komal and MFB-3 were grouped under low girth seeds. Whereas medium girth was observed in the varieties of IIHR-909 and MFB-2, and the varieties of RSJ-288, Contender were having high girth seeds. The mean seed girth for all the varieties was 17.58 mm.

4.3.1.1.6 Seed size (mm)

The varieties exhibited different seed sizes, are categorized as small (6.0 to 7.0 mm), medium (7.0 to 8.0 mm), large (>8.0 mm) and are presented in Table 16.

Table 16. Seed morphometric measurements of french bean varieties

Varieties	Seed length (mm)	Class	Seed girth (mm)	Class	Seed size (mm)	Class
1. RSJ-288	13.93	Large	19.73	High	7.95	Medium
2. IHR-909	11.66	Medium	17.46	Medium	7.75	Medium
3. MFB-1	10.36	Small	15.96	Low	6.16	Small
4. Contender	15.43	Large	21.63	High	8.80	Large
5. Arka Komal	10.83	Small	13.86	Low	7.15	Medium
6. MFB-2	14.86	Large	18.40	Medium	8.10	Large
7. MFB-3	11.06	Small	15.83	Low	6.74	Small
Mean	12.59	10.0-11.5 mm small	17.58	13.5-16.0 mm low	7.61	6.0 to 7.0 mm small
S.Em _±	0.315	11.5-13.0 mm medium	0.462	16.0-18.5 mm medium	0.092	7.0 to 8.0 mm medium
CD at 5%	0.97	>13.0 mm large	1.42	>18.5 mm High	0.284	> 8.0 mm large

Varieties *viz.*, MFB-1, Arka Komal and MFB-3 were grouped under small sized seeds. IIHR-909 is under medium size and RSJ-288, Contender and MFB-2 are grouped under large sized seeds. On average, seed size of all the varieties was 7.61 mm.

4.3.1.2 Seedling morphology

4.3.1.2.1 Hypocotyl colour

The varieties under study are varied for the seedling hypocotyl colour and the data are presented in Table 17 and depicted in Plate 2.

RSJ-288, IIHR-909, contender, Arka Komal and MFB-3 varieties were having hypocotyl colours of purple, green, light green, yellowish green and light purple, respectively whereas, MFB-1 and MFB-2 were having pale green colour.

4.3.1.2.2 Cotyledon colour

Based on cotyledon colour, varieties are grouped as purple, green and light green, the data is presented in Table 17.

Purple cotyledon, was seen with RSJ 288, whereas, IIHR-909, MFB-1, Arka Komal, MFB-2, MFB-3 were having green cotyledon and Contender with light green cotyledon.

4.3.1.2.3 Nature of pubescence on hypocotyl

Based on pubescence on hypocotyl, varieties were grouped as Dense hairiness and Glabrous (without hairiness), and presented in Table 17.

MFB-3 was having with Dense Pubescence whereas MFB-1, Contender and MFB-3 are glabrous (without hairs on hypocotyl).

Table 17. Seedling morphological characters of french bean varieties

Varieties	Hypocotyl colour	Cotyledon colour	Pubescence on hypocotyl
1. RSJ-288	Purple	Purple	Dense
2. IIHR-909	Green	Green	Dense
3. MFB-1	Pale green	Green	Glabrous
4. Contender	Light green	Light	Glabrous
5. Arka Komal	Yellowish green	Green	Dense
6. MFB-2	Pale green	Green	Glabrous
7. MFB-3	Light purple	Green	Dense

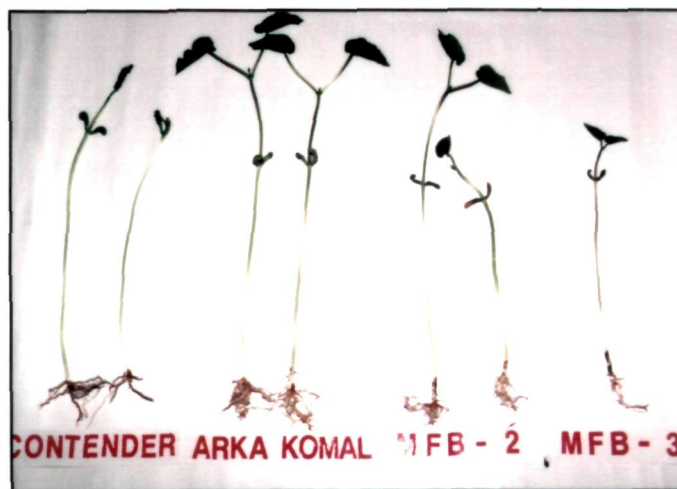
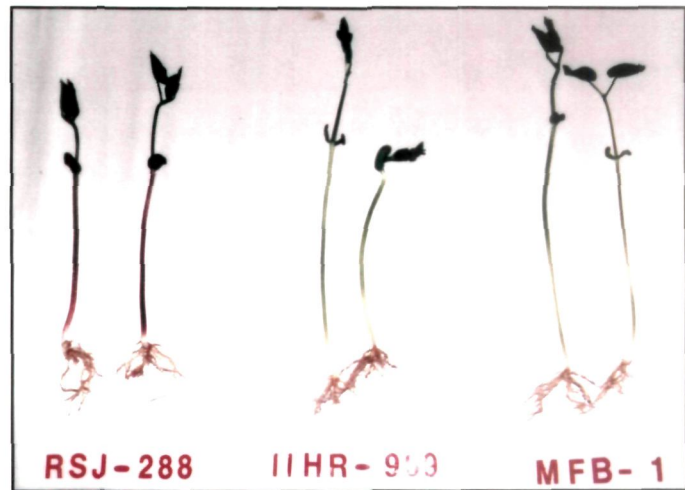
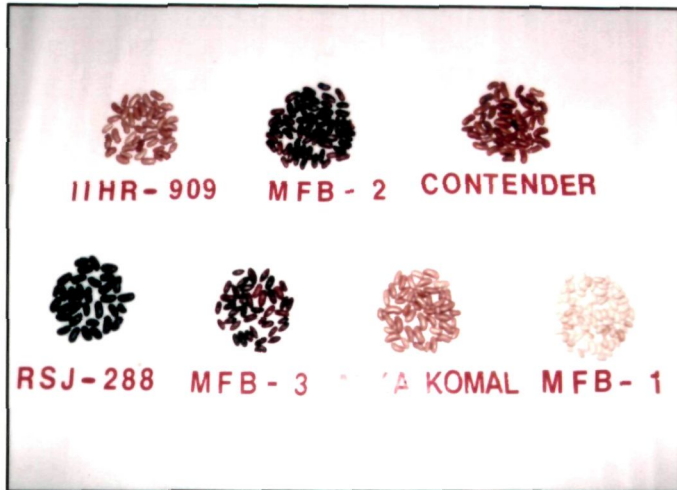


Plate 2: Variation in seed coat colour and seedling morphology in French bean varieties

4.3.1.3 Plant morphology

4.3.1.3.1 Growth habit

Growth habit for the french bean varieties was categorized as as spready and Erect. The observations on which are presented in Table 18.

Spready growth habit was seen with RSJ-288 and Contender and the other varieties like IIHR-909, MFB-1, Arka Komal, MFB-1 and MFB-2 were having the erect growth habit.

4.3.1.3.2 Leaf colour

Based on leaf colour, varieties are grouped into light green, green and dark green and observations are presented in Table 18.

Arka Komal was categorized under light green. RSJ-288, MFB-1, Contender and MFB-2 were categorized under green, whereas, IIHR-909 and MFB-3 are observed with dark green leaves.

4.3.1.3.3 Flower colour

French bean varieties classified into five classes based on flower colour. The observations on flower colour are presented in Table 18 and depicted in Plate 3.

RSJ-288 produced dark purple flower, colour, IIHR-909 and Arka Komal having with purplish white flower, colour, whereas, white flowers in MFB-1, purple flowers in Contender and MFB-2. While, MFB-3 found to produce light purple flowers.

4.3.1.3.4 Constriction on pod

The varieties were classified into shallow, moderate, deep and very deep on the basis of constriction on pod, the readings are presented in Table 18.

Varieties *viz.*, RSJ-288 and MFB-2 are categorized into shallow constriction whereas IIHR-909, MFB-1 grouped under moderate constriction and contender, Arka Komal varieties with deep pod constriction. While, MFB-3 is having very deep pod constrictions.

4.3.2 Electrophoretic characterisation

4.3.2.1 Native total proteins of hypocotyl

The native total protein profiles of seven genotypes are presented in the form of zymogram (Fig. 8). A total of 15 bands with Rm value ranging from 0.018 to 0.973 were resolved of these three bands were polymorphic corresponding to Rm values 0.045, 0.161 and 0.536. Based on the presence or absence of these bands, three electrophoretic phenotypes (H₁-H₃) were identified, as described below.

Electrophoretic phenotype	Characteristics	Varieties
H ₁	The polymorphic band of Rm value 0.045 was absent	RSJ-288
H ₂	The band at Rm value 0.161 was absent	MFB-1, Contender, Arka Komal, MFB-2
H ₃	The bands at Rm value 0.161 and 0.536 were absent	IIHR-909, MFB-3

Table 18. Plant morphological characters of french bean varieties

Treatments	Growth habit	Leaf colour	Flower colour	Constriction on pod
1. RSJ-288	Spready	Green	Dark purple	Shallow
2. IHR-909	Erect	Dark green	Purplish white	Moderate
3. MFB-1	Erect	Green	White	Moderate
4. Contender	Spready	Green	Purple	Deep
5. Arka Komal	Erect	Light green	Purplish white	Deep
6. MFB-2	Erect	Green	Purple	Shallow
7. MFB-3	Spready	Dark green	Light purple	Very deep

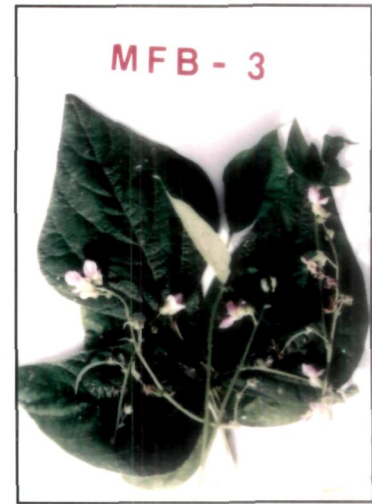
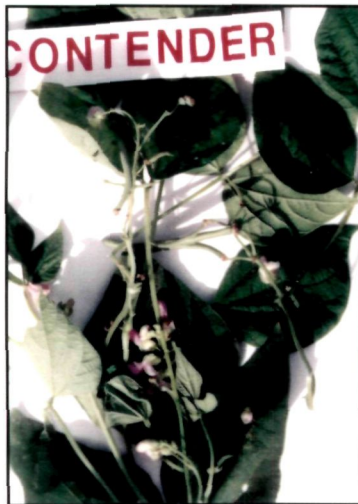
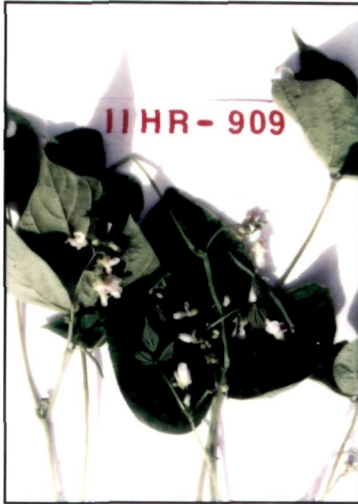
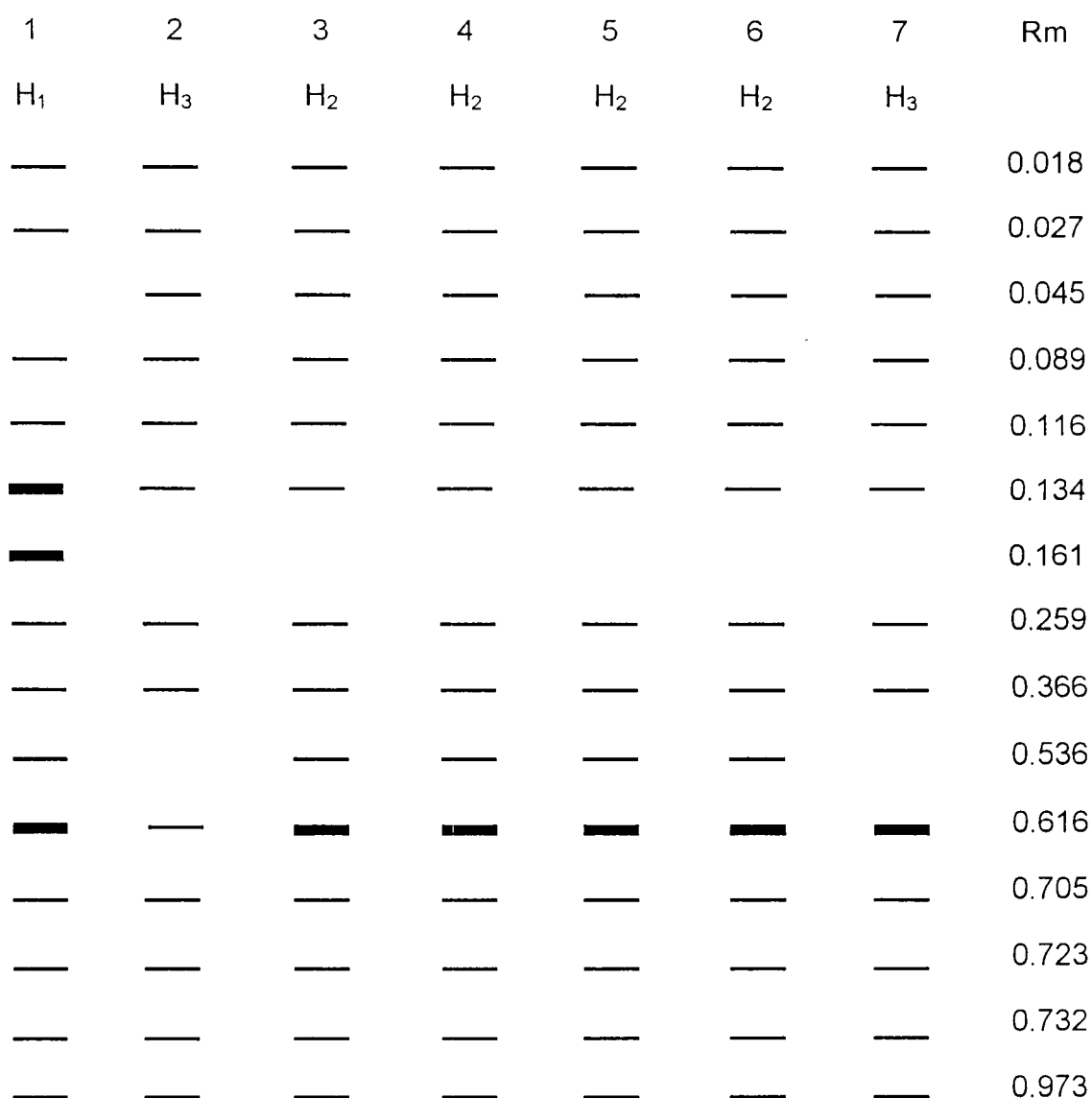


Plate 3: Variation in flower colour of French bean varieties



H1 – H3 : Electrophoretic phenotype for hypocotyl protein
Rm : Relative mobility values

1. RSJ-288 2. IIHR-909 3. MFB-1 4. Contender
5. Arka Komal 6. MFB-2 7. MFB-3

Figure 8 : Hypocotyl protein profiles of frenchbean varieties

4.3.2.2 Isozyme profiles of genotypes

The isozyme- banding of peroxidase for seven genotypes is presented in the form of zymograms (Fig. 9). Single band with Rm value 0.285 was observed. So only one monomorphic band and Rm value 0.285 was observed in all the seven varieties of french bean.

4.3.2.3 Random Amplified Polymorphic DNA analysis

RAPD analysis was carried out for using OPF series primers and identification of varieties can be done using four primers namely OPF-7, OPF-10, OPF-16 and OPF-20.

RAPD profiles of seven french bean varieties using OPF-7 primer is depicted in Plate 4.

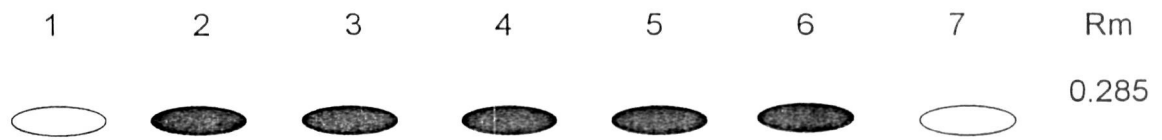
Polymorphic bands was noticed in variety Contender and MFB-2. These two varieties exhibited extra band of 4000 bp, while other varieties are having a single band of 2000 bp.

RAPD profiles of french bean varieties using OPF-10 primer is depicted in Plate 5.

Frenchbean varieties exhibited polymorphic banding pattern. The varieties *viz.*, IHR-909, MFB-1, Arka Komal, MFB-2 and MFB-3 showed a distinct band of 600 bp. While this band is absent in RSJ-288 and Contender variety.

RAPD pattern of french bean varieties using OPF-16 primer are presented in Plate 6.

Varieties *viz.*, MFB-1 and MFB-2 showed polymorphic banding pattern, these varieties possessed an extra band of 1750 bp whereas other



Rm : Relative mobility value

1. RSJ-288 2. IIHR-909 3. MFB-1 4. Contender

5. Arka Komal 6. MFB-2 7. MFB-3

Fig. 9 : Hypocotyl peroxidase isozyme profile of frenchbean varieties

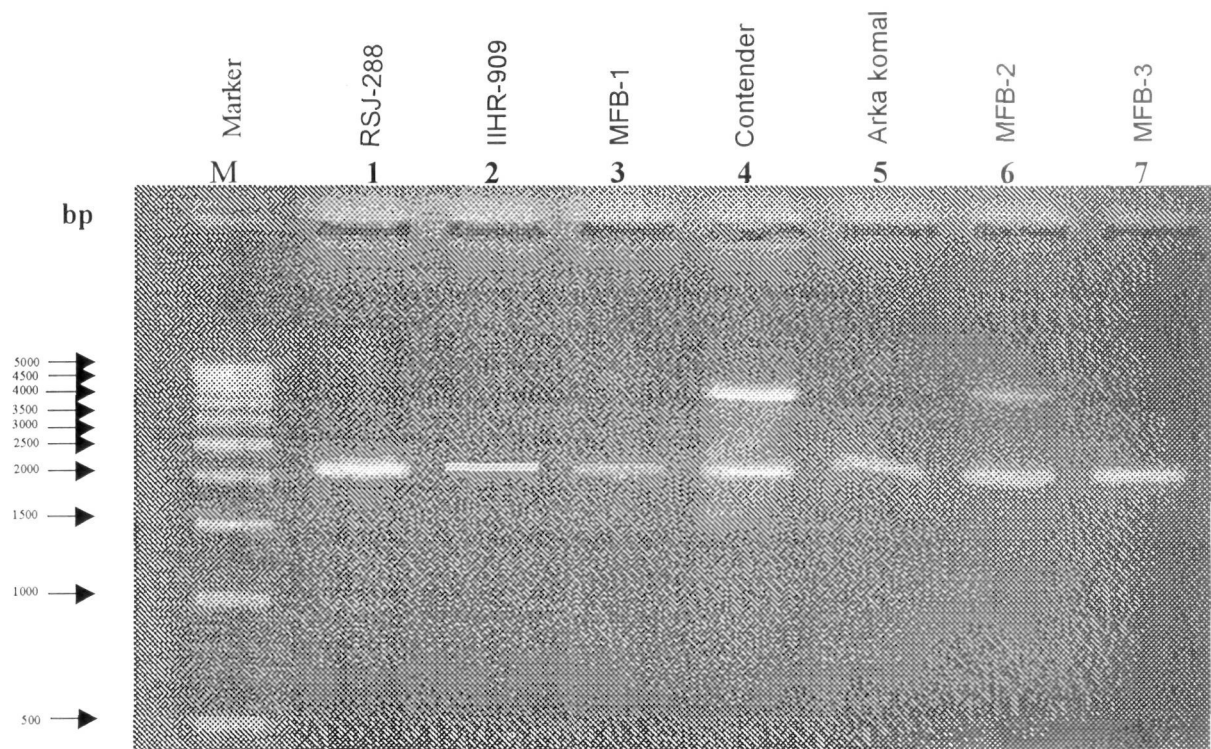


Plate 4 : RAPD profiles of Frenchbean varieties using OPF 7 primer

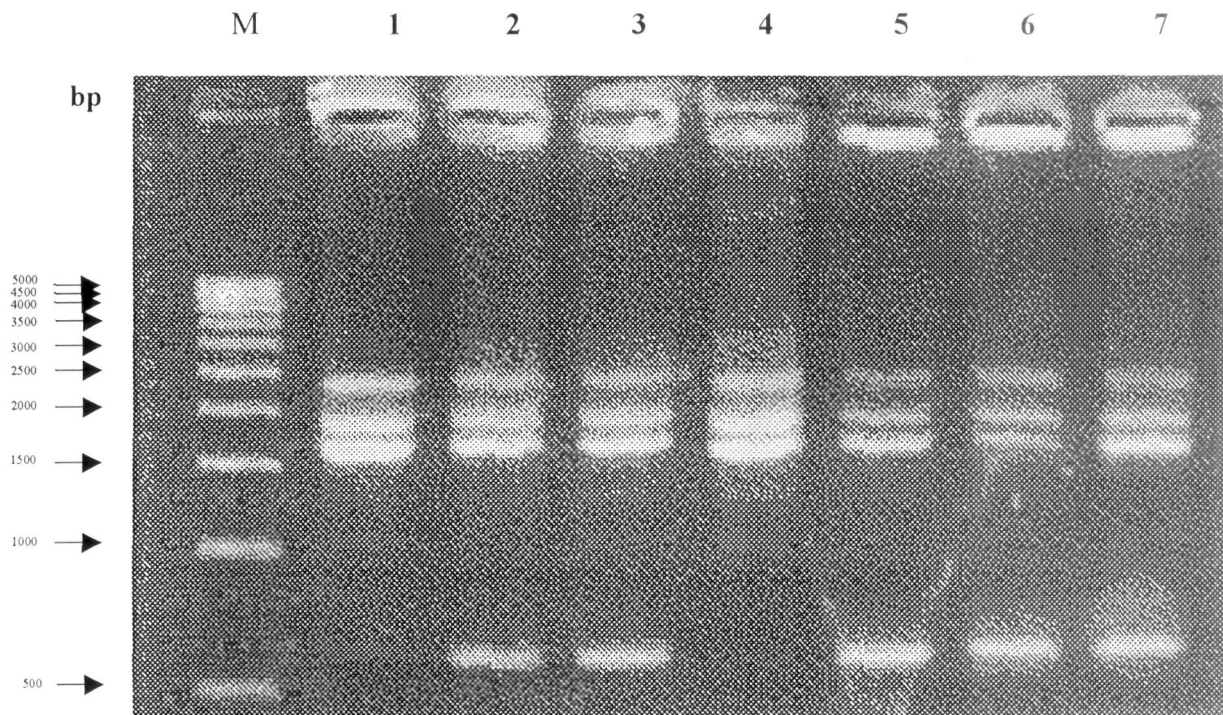


Plate 5 : RAPD profiles of Frenchbean varieties using OPF 10 primer

varieties *viz.*, RSJ-288 IIHR, contender, Arka Komal and MFB-3 are lacking the band of 1750 bp.

RAPD profiles of french bean varieties using OPF-20 primer are presented in Plate 7.

Compared to OPF-7, 10 and 16 primers, OPF-20 showed higher polymorphic banding pattern. Contender variety possesses three extra band of 1400, 2200 and 3000 bp. While, the variety MFB-3 has only one extra band of 1400 bp, but the variety MFB-1 is lacking with the band of 700 bp. Whereas the other varieties *viz.*, RSJ-288, IIHR-909, Arka Komal and MFB-2, are having the similar bands of 1500, 1000, 700 and 600 bp.

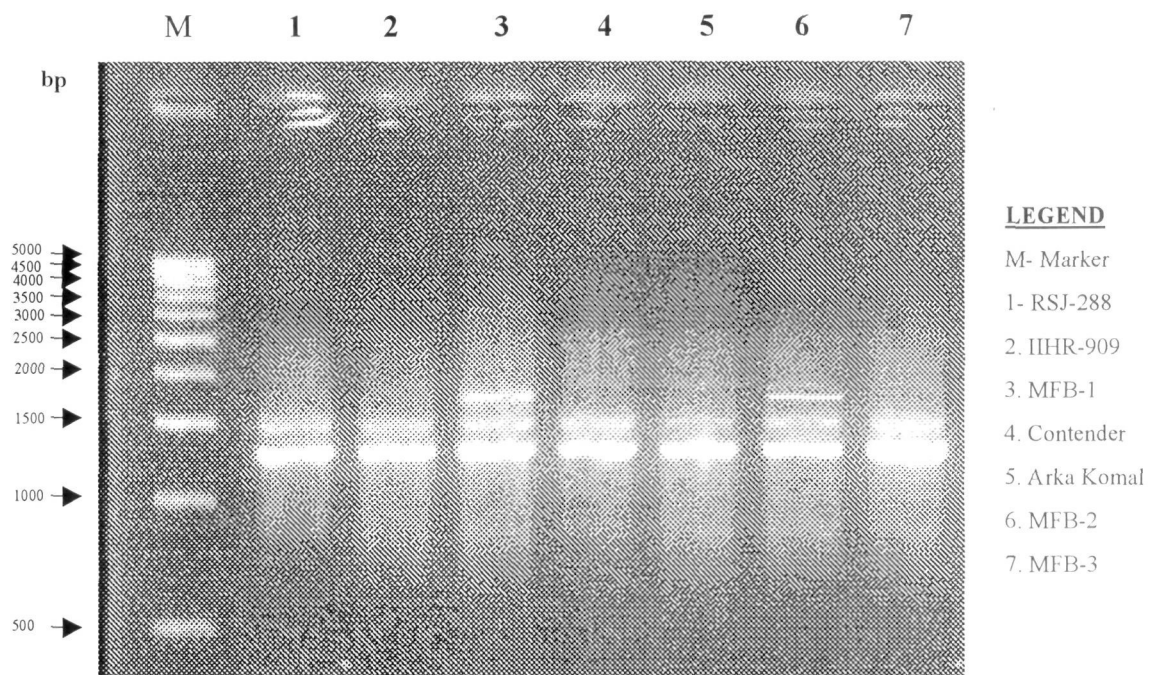


Plate 6 : RAPD profiles of Frenchbean varieties using OPF 16 primer

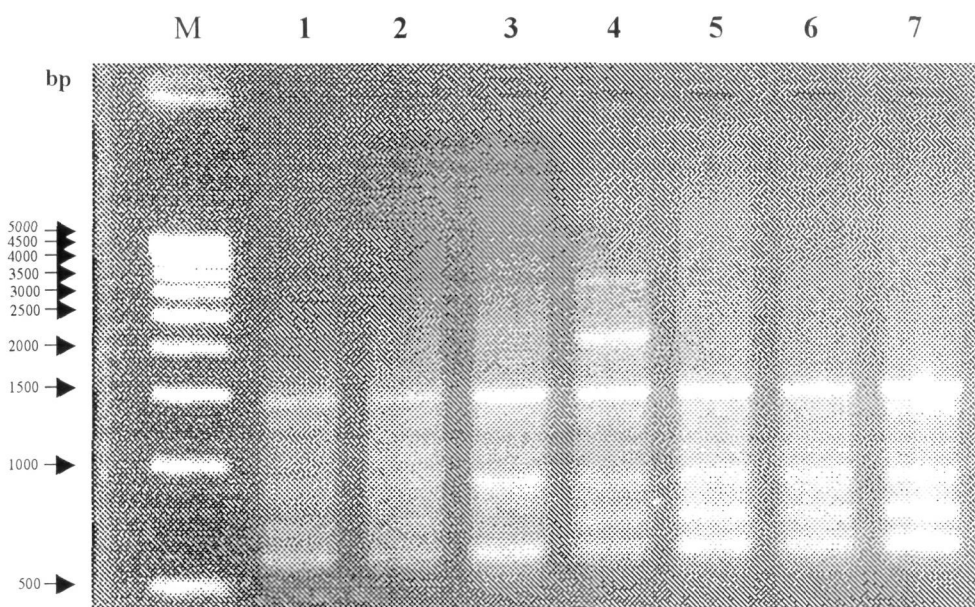


Plate 7 : RAPD profiles of Frenchbean varieties using OPF 20 primer

Discussion

V DISCUSSION

A field experiment was conducted to study the effect of spacing and phosphorus levels on crop growth, seed yield and quality of french bean variety Arka Suvidha (IIHR-909) at the Main Agricultural Research Station during *kharif* season of 2002 and varietal identification studies were conducted at NSP/BSP Unit, University of Agricultural Sciences, Dharwad. The results of the investigations are discussed in this chapter.

5.1 EXPERIMENT - I : EFFECT OF SPACING AND PHOSPHORUS LEVELS ON GROWTH, SEED YIELD AND QUALITY OF FRENCH BEAN cv. ARKA SUVIDHA (IIHR-909)

5.1.1 Effect of spacing

Seed yield and quality are the net result of an interplay of diverse metabolic activity taking place in different plant parts, which are largely influenced by the environmental factors. The maximum seed yield in a particular cultivar/crop under a given set of environment can be obtained with the plant density where competition among the plants is minimal. This can be achieved with optimum plant density which not only help to utilize light, moisture and nutrients more effectively but also avoids excess competition among the plants. Hence, efforts were made to the investigate the optimum plant density on growth, seed yield and quality parameters.

5.1.1.1 Growth parameters

The plant dry weight (Table 5) differed significantly. Maximum plant dry weight (11.43 g) at harvest was observed at spacing of 45x20 cm which was followed by the spacings of 30x20 cm (9.99 g), 45x10 cm

(8.59 g) and minimum dry weight was observed at 30x10 cm (7.35 g), which is 32 per cent lesser than that of 45x20 cm spacing. This is because of maximum number of branches (11.40) per plant recorded at wider spacing of 45x20 cm which is 48.2, 33.0 and 7.6 per cent higher over spacing levels of 30x10 cm, 45x10 cm and 30x20 cm, respectively. The low number of branches and lesser number of leaves per plant were observed at closer spacing of 30x10 cm. At 60 DAS, 30x10 cm spacing recorded minimum number of leaves (9.82) per plant, while wider spacings of 45x10, 30x20 and 45x20 cm recorded in increasing order as 11.11, 11.70 and 13.58 leaves per plant, respectively. These results are in agreement with the findings of Chatterjee and Som (1991), Jadhavo (1993), Dwivedi *et al.* (1995a), Shivakumar *et al.* (1996) and Dhanjal *et al.* (2001) in french bean.

The spacing had significant influence on plant height at all growth stages. Maximum plant height at all stages of growth was recorded at closer spacing of 30x10 cm. At harvest 30x10 cm spacing recorded the plant height of 30.94 cm followed by 45x10 cm (28.81 cm), 30x20 cm (27.69 cm) and minimum height was recorded at wider spacing of 45x20 cm (26.76 cm). This may be due to inter and intra plant competition for sunlight among the plants at closer spacing. It also encouraged self thinning of branches because of the non availability of light that led to enhancement of vertical growth rather than horizontal growth. Hence the plant height was more at closer spacing. The similar results were also reported by Pande *et al.* (1974), Shivakumar *et al.* (1996) and Dwivedi *et al.* (1995a) in french bean.

5.1.1.2 Yield parameters

Higher seed yield per ha (2138 kg) was recorded at the spacing of 30x20 cm which was on par with 30x10 cm (2043 kg) than wider spacings of 45x10 cm (1813 kg) and 45x20 cm (1886 kg). The wider spacing of 45x20 cm recorded maximum values of yield attributing characters like more number of pods per plant (15.34), more seeds per pod (5.22), higher seed weight per pod (1.74 g) and seed weight per plant (26.14 g) followed by spacing levels of 30x20 cm and 45x10 cm as (13.03, 4.77, 1.65 g, 22.14 g and 10.45, 4.78, 1.56 g, 16.23 g, respectively). Whereas the minimum values were recorded at closer spacing of 30x10 cm as (8.75, 4.50, 1.57 g and 13.58 g). The increase in seed yield and its attributing characters at wider spacing because of increased nutrient uptake, better light penetration and space availability for individual plant that increase branches, and leaves in wider spaced plants in turn increased the seed yield and its attributing parameters. Though the yield attributing parameters per plant were higher at 45x20 cm spacing but, the yield per ha was minimum because of less population density per ha (1.1×10^5 plants), while at 30x20 cm spacing per ha population was 1.6×10^5 plants which was recorded maximum seed yield per ha. This was on par with 30x10 cm spacing (3.3×10^5 plants per ha). The increase in seed yield at closer spacing was mainly attributed to increased plant population, average yield per plant rather than better individual plant performance as was evident in the study. These results are in agreement with the reports of Pande *et al.* (1974), Masood Ali and Tripathi (1988) in french bean cultivars, 'PDR 14' and 'VL 63' Chatterjee and Som (1991), Dhanjal *et al.* (2001) and Das *et al.* (1996) in french bean.

5.1.1.3 Seed quality parameters

The differences in seed germination was significant among the spacings.

Maximum seed germination percentage (92.08) was observed with the spacing level of 30x20 cm, which was on par with 45x10 cm (91.16) and 45x20 cm (91.50) and minimum germination per cent was recorded at 30x10 cm spacing (89.00). The other seed quality parameters such as 100 seed weight (g), rate of germination, root length (cm), seed vigour index and seedling dry weight (g) were maximum at the spacing level of 45x20 cm as (36.23, 30.34, 11.64, 20.30, 2924, 27.4). Followed by 30x20 cm and 45x10 cm and minimum values are recorded at closer spacing of 30x10 cm, (33.60x 29.10, 12.07, 19.23, 2786 and 25.2) respectively.

The seedling root length, shoot length and vigour index were non-significant between spacing levels.

Electrical conductivity of seed leachate was low at wider spacing of 45x20 cm compared to 30x10 cm because the wider spacing increased nutrient uptake resulted with intact seed coat.

The seed protein per cent was maximum at the wider spacing of 45x20 cm (23.60) followed by 30x20 cm (22.20), 45x10 cm (19.74). Minimum was seen at closer spacing of 30x10 cm (18.54%). Significant increase in seed quality parameters noticed at wider spacing of 45x20 cm was mainly due to increase in the seed size and better filling of individual seed. This can be explained on the basis of efficient photosynthetic activity, uptake of nutrients, and better translocation of photosynthates from source to sink (seed) (Hazra and Som, 1999). The similar increase in

seed quality parameters were also reported by Masood Ali and Tripathi (1988) and Loato *et al.* (1982) in french bean.

The spacing of 30x20 cm recorded maximum seed yield per ha (2138 kg) which was on par with 30x10 cm (2043 kg). Although the wider spacing of 45x20 cm recorded maximum values for seed quality parameters but on par with 30x20 cm. Therefore, by looking into above discussion, it can be concluded that the spacing of 30x20 cm found to be optimum for getting higher seed yield with better quality seeds in french bean cv. IHR 909.

5.1.2 Effect of phosphorus

Phosphorus is a major plant nutrient known to have role in synthesis of proteins, which could increase vertical growth (plant height) and lateral growth in terms of number of branches (Noggle and Fretz, 1979). Phosphorus plays an important role in crop growth and seed yield of any pulse crop, by manipulating the morphology and physiology of plants it brings optimum source-sink relationship which results in higher seed yield coupled with better seed quality attributes.

5.1.2.1 Growth parameters

Phosphorus had significant influence on plant growth which was evidenced by plant dry weight at harvest.

The maximum plant dry weight at harvest (11.42 g) was observed at 125 kg P₂O₅ per ha followed by 100 kg P₂O₅ per ha (9.20 g) and minimum dry weight at harvest (7.40 g) was obtained at 75 kg P₂O₅ per ha. This may be attributed to increased plant height (29.73 cm), branches per plant (10.59) and number of leaves at 60 DAS (12.59) at 125 kg P₂O₅ per ha followed by 100 kg P₂O₅ per ha (28.70 cm, 9.39 and 11.89, respectively)

and minimum values were observed at 75 kg P₂O₅ per ha (27.22 cm, 8.71 and 10.18, respectively).

These results are in agreement with the reports on french bean by Srinivas and Naik (1988), Roy and Parthasarathy (1999) Singh and Singh (2000) found increased vegetative growth up to 120 kg P₂O₅ per ha compared to 80 kg P₂O₅ per ha.

5.1.2.2 Yield parameters

The phosphorus showed significant influence on seed yield per ha.

Maximum seed yield (2157 kg/ha) was recorded at 125 kg P₂O₅ per ha followed by 100 kg P₂O₅ per ha (1975 kg/ha) and minimum seed yield at 75 kg P₂O₅ per ha (1779 kg/ha). The seed yield at 125 kg P₂O₅ per ha was 21.24 and 9.2 per cent more over 75 and 100 kg P₂O₅ per ha respectively.

This may be due to maximum values of yield attributing parameters like number of pods per plant (13.43), number of seeds per pod (5.50), pod length (13.99 cm) at higher level of phosphorus (125 kg/ha). Seed weight per pod (1.70 g) and seed weight per plant (21.33 g), followed by 100 kg P₂O₅ per ha and minimum values were seen at 75 kg P₂O₅ per ha as (10.17, 4.44, 12.98 cm, 1.56 g, and 17.70 g, respectively). The increased phosphorus level had increased photosynthetic activity, uptake of nutrients, nitrogen fixation, photosynthate synthesis and translocation from source (leaf) to sink (seed), that enhanced reproductive activity resulting in high seed setting and filling. These results are in confirmity with the reports on french bean by Srinivas and Prabhakar (1985), Srinivas and Naik (1988), Deshpande *et al.* (1995) in the variety HP4-35.

Tomar (2001) in cv. PDR-14 and Dwivedi *et al.* (1995b) in cultivar Arka Komal.

5.1.2.3 Seed quality parameters

The differences in seed quality parameters were found to be significant.

Maximum germination per cent of 92.31 was recorded at 125 kg P_2O_5 per ha followed by 100 kg (90.81) and 75 kg P_2O_5 per ha (89.68). Higher level of phosphorus (125 kg/ha) recorded maximum values of quality attributing parameters like 100 seed weight (36.89 g), rate of germination (31.12), root length (12.31 cm), shoot length (21.52 cm), seed vigour index (3122) and seedling dry weight (27.13 mg) followed by 100 kg phosphorus per ha (34.88, 29.79, 11.83, 20.67 and 2951, 26.34) and minimum values at 75 kg phosphorus per ha, (33.45, 28.25, 11.18, 18.51 and 2661, 25.09, respectively). Electrical conductivity of seed leachate was low at highest phosphorus dose of 125 kg per ha (1.149 dSm^{-1}) followed by 75 kg P_2O_5 per ha (1.398 dSm^{-1}). Seed protein was significantly more at high dose of phosphorus (21.76%) followed by medium level (20.98%) and least was seen at low level of phosphorus (20.32%).

The increased phosphorus level has increased the seed quality parameters because of the fact that phosphorus application to french bean (legume crop) stimulated root growth, symbiotic N- fixation, uptake of other essential nutrients and translocation of photosynthates from source to sink. Higher seed germination at high 'P' application is due to higher 100 seed weight which inturn was supplied adequate food reserves during germination. The results are in accordance with the findings of Viera (1986), Khyad (1996), Singh and Singh (2000) in french bean,

Handovizadeh and George (1988) in pea and Meena *et al.* (2001) in chickpea from the above discussion, it can be concluded that increased application of phosphorus increased growth, yield and seed quality parameters.

5.1.3 Interaction of spacing and phosphorus levels

5.1.3.1 Growth parameters

The interaction was significant for the growth parameters. The combined effect of spacing (45x20 cm) and phosphorus (125 kg P₂O₅/ha) recorded maximum plant dry weight per plant at harvest (14.18 g) and least dry weight was obtained in the spacing of 30x10 cm at 75 kg P₂O₅ per ha (5.42 g). This may be attributed to wider spacing of 45x20 cm and phosphorus level of 125 kg P₂O₅ per ha recorded maximum branches per plant at harvest (12.20) and more number of leaves at 60 DAS (14.81), where as closer spacing of 30x10 cm at 75 kg P₂O₅ per ha produced minimum values (6.57 and 8.25, respectively). The plant height was maximum at combined effect of spacing of 30x10 cm at 125 kg P₂O₅ per ha. These results are in accordance with the reports of Singh (2000) and Chatterjee and Som (1991) in french bean.

5.1.3.2 Yield and yield parameters

The combined effect of spacing of 30x20 cm coupled with phosphorus 125 kg per ha recorded maximum seed yield per ha (2454 kg), which was on par with the combination of 30x10 cm spacing at 125 kg P₂O₅ per ha (2259 kg). The yield attributing factors were maximum in the combined effect of wider spacing of 45x20 cm at 125 kg P₂O₅ per ha ; like number of pods per plant (16.96), seeds per pod (5.50), seed weight per pod (1.89 g) and seed weight per plant (26.90 g), among them number of

seeds per pod and seed weight per pod were on par with combination of 30x20 cm spacing at 125 kg P₂O₅ per ha (5.61 and 1.74 g).

Minimum values of yield attributing parameters were recorded in the spacing of 30x10 cm at 75 kg P₂O₅ per ha (7.22, 4.25, 1.51 and 12.13). Such results were also reported on french bean by Ahlawat (1996) and Chatterjee and Som (1991) in french bean.

5.1.3.3 Seed quality parameters

The maximum germination per cent (93.75) was recorded in the combined effect of 45x20 cm spacing at 125 kg P₂O₅ per ha, which was on par with spacing level of 30x20 cm at 125 kg P₂O₅ per ha (93.25), 30x20 cm at 100 kg P₂O₅ per ha (92.99) and 45x20 cm at 125 kg P₂O₅ per ha (91.25). The seed quality attributing characters like root length, shoot length and vigour index were not varied significantly due to interaction.

The reduced Electrical conductivity (1.127 dSm⁻¹) in the wider spacing of 45x20 cm coupled with application of 125 kg P₂O₅ per ha indicating better seed quality.

The wider spacing of 45x20 cm at 125 kg P₂O₅ per ha recorded maximum seed protein of 24.17 per cent and minimum protein (17.83%) was seen at 30x10 cm at 75 kg P₂O₅ per ha treatment combination.

By looking into the above results, it can be concluded that combined effect of 30x20 cm spacing coupled with application of 125 kg P₂O₅ per ha resulted in maximum seed yield with better quality in french bean cv. IHR 909.

5.2 EXPERIMENT - II : SCREENING OF FRENCH BEAN VARIETIES FOR SEED YIELD AND QUALITY PARAMETERS

In the present study seven french bean varieties *viz.*, RSJ-288, IHR-909, MFB-1, Contender, Arka Komal, MFB-2, and MFB-3 were screened for seed yield and quality parameters. The results of which are discussed below.

5.2.1 Growth parameters

The variety Contender recorded minimum number of days for 50 per cent flowering (37-38), whereas MFB-3 took maximum days to 50 per cent flowering (41.42). Significantly highest number of branches per plant was recorded in the variety, RSJ-288 (11.20) followed by IHR-909 (10.20) and least was at Arka Komal and MFB-2 (6.00) compared to other varieties *viz.*, MFB-1, Contender, and MFB-3 highest number of branches found in RSJ-288 may be due to bushy /spreadly nature of genotypes, whereas others showed either semi spreadly or erect type.

5.2.2 Yield parameters

Higher seed weight per plant (18.90 g) was recorded with the variety RSJ-288 compared to other varieties. Increase in seed yield per plant in RSJ-288 can be attributed due to more number of branches per plant (11.20) and pods per plant (12.0). Though, IHR-909 recorded maximum number of seeds per pod (6.40) and pod weight (2.56 g), it did not give more seed yield per plant because of less number of pods per plant (9.60) as it was observed. Such varietal differences in seed yield and its components also reported by other research workers (Singh *et al.*, 1997) in

broad bean, (Dhanjal *et al.*, 2001) and (Roy and Parthasarathy, 1999) in french bean.

5.2.3 Seed quality parameters

Seed quality parameters like germination per cent, root length (cm), shoot length (cm) and seedling vigour index did not vary significantly among the varieties. However, seedling dry weight was maximum at RSJ-288 (28.33 mg) because this variety recorded maximum 100 seed weight (38.93 g) and minimum electrical conductivity values (0.696 dSm⁻¹), highest protein per cent was recorded in the variety MFB-2 (22.06 %) compared to other varieties.

5.3 EXPERIMENT - III : VARIETAL IDENTIFICATION OF FRENCH BEAN CULTIVARS

5.3.1 Morphological identification

In the present investigation, attempts were made to characterize some of the promising varieties of french bean based on certain seed, seedling and plant morphological characters in order to develop varietal identification keys which are reliable and easy to perform at field and laboratory level. The results generated from the above said characters have been discussed below.

5.3.1.1 Seed morphology

In the present study seven french bean varieties *viz.*, RSJ-288, Contender, Arka Komal, IIHR-909, MFB-2, MFB-1 and MFB-3 were grouped into seven seed coat colour as black, pale brown, brown, light brown, mottled, creamy white and dark brown. Seed coat colour differences is mainly due to the heritable characters of the variety. Seed

colour is influenced by environmental conditions during ripening in addition to the genetic effect (Pascual Villalobos *et al.*, 1993).

Seed hilum colour found to be white in all the varieties, reason could be ascertained as, if no mutation has been occurred that results in seeds having identical hila (Taylor and Cavness, 1982).

Seed shape for different french bean varieties were grouped into cylinder, kidney and oval shape. The variety RSJ-288 is grouped as cylinder Contender, Arka Komal and IIHR-909 grouped into kidney. Whereas, MFB-1, MFB-2, MFB-3 are grouped as oval shape. The variety RSJ-288 could be identified individually from other varieties. This may be due to the position of the seed in the pod and may be influenced by environmental conditions during the pod filling stage. Mainly it is a genetic character (Richard and Payne, 1979). Similarly in pea, 22 cultivars are grouped into round or flattened seed shape (Anon., 1992).

Based on seed size, the varieties were grouped into small (6.0 to 7.0 mm), medium (7.0 to 8.0 mm) and large (> 8.0 mm). The varieties MFB-1, Arka Komal and MFB-3 are grouped under small, whereas IIHR-909 is categorized as medium and remaining RSJ-288, Contender, MFB-2 are categorized into large size. The variation may be due to the influence of embryo size and other food resource materials with respect to varieties. This is in agreement with the work done by Mitranov (1983).

Generally, seed size varied with the varieties. The normal seed size is controlled by polygenes and extra-ordinary large or small seed size is controlled by major gene or genes (Takeda, 1991).

5.3.1.2 Seedling morphology

Based on pubescence on hypocotyl, varieties are categorized into Dense (hairy) and Glabrous (non-hairy). The varieties like RSJ-288, IIHR-909, Arka Komal are grouped into dense hairy group, whereas MFB-1, Contender and MFB-2 are grouped under Glabrous. This variation is due to genetic characters of the varieties, as reported by Pathak and Singh (1961). The similar results were also reported by Edgar *et al* (1970) in soybean.

The varieties are categorized into purple, green and light green based on cotyledon colour, RSJ-288 is purple cotyledon, whereas IIHR-909, MFB-3, MFB-1, MFB-2 and Arka Komal are categorized under green. Contender is grouped under light green group.

Hypocotyl colour varied with the varieties *viz.*, RSJ288 (purple), IIHR-909 (green), Arka Komal (Yellowish green), MFB-3 (Light purple), MFB-1 and MFB (Pale green) and Contender (light green). This character is under genetic control (Jana and Rao, 1974). These observations are in accordance with the reports of Chakrabarthy and Agarwal (1989b) in Blackgram.

5.3.1.3 Plant morphology

French bean varieties exhibited two types of plant growth habit like erect and spready. Erect growth habit varieties are IIHR-909, MFB-1, Arka Komal, MFB-2, and others, RSJ-288, Contender, MFB-3 are categorized under spready type. This character is governed by the genetic factor of the varieties. Similar results were reported by Edgar *et al.* (1970), Millions *et al.* (1984) and Ravikumar (1998) in soybean.

Leaf colour at maturity is grouped into light green, green and dark green. Arka Komal is categorized under light green, RSJ-288, Contender, MFB-1 and MFB-2 are Green whereas as MFB-3 and IIHR-909 under Dark green. This variation may be due to soil and environmental conditions during crop growth and inherited genes. Similar results were reported by (Anon., 1992) in chickpea and Ravikumar (1998) in soybean.

The varieties varied for flower colour as RSJ-288 (Dark purple), Contender and MFB-2 (purple) MFB-3 (light purple), Arka Komal and IIHR-909 (Purplish whiter) and MFB-1 (white). The variation in flower colour among the varieties is due to genotypic character of the varieties. Similar work was carried out by various workers (Edgar *et al.*, 1970, Kozykowski and Burgoon, 1983 and Agarwal, 1984 in soybean).

Based on pod constriction the varieties RSJ-288 and MFB-2 are grouped under shallow, Contender, MFB-1 and IIHR-909 as moderate and Arka Komal under deep constriction. While, MFB-3 is having very deep pod constriction.

5.3.1.4 Development of varietal identification keys

The present investigation reveals the seed, seedling and plant morphological characters based on these characters varietal identification keys for seem french bean varieties are developed. In the development of these keys, the characters having high stability and least influenced by the environment are taken as the major diagnostic characters to form broad clarifications (Fig. 10, 11 &12).

Fig. 10 : French bean varieties identification key on the basis of seed morphological characteristics

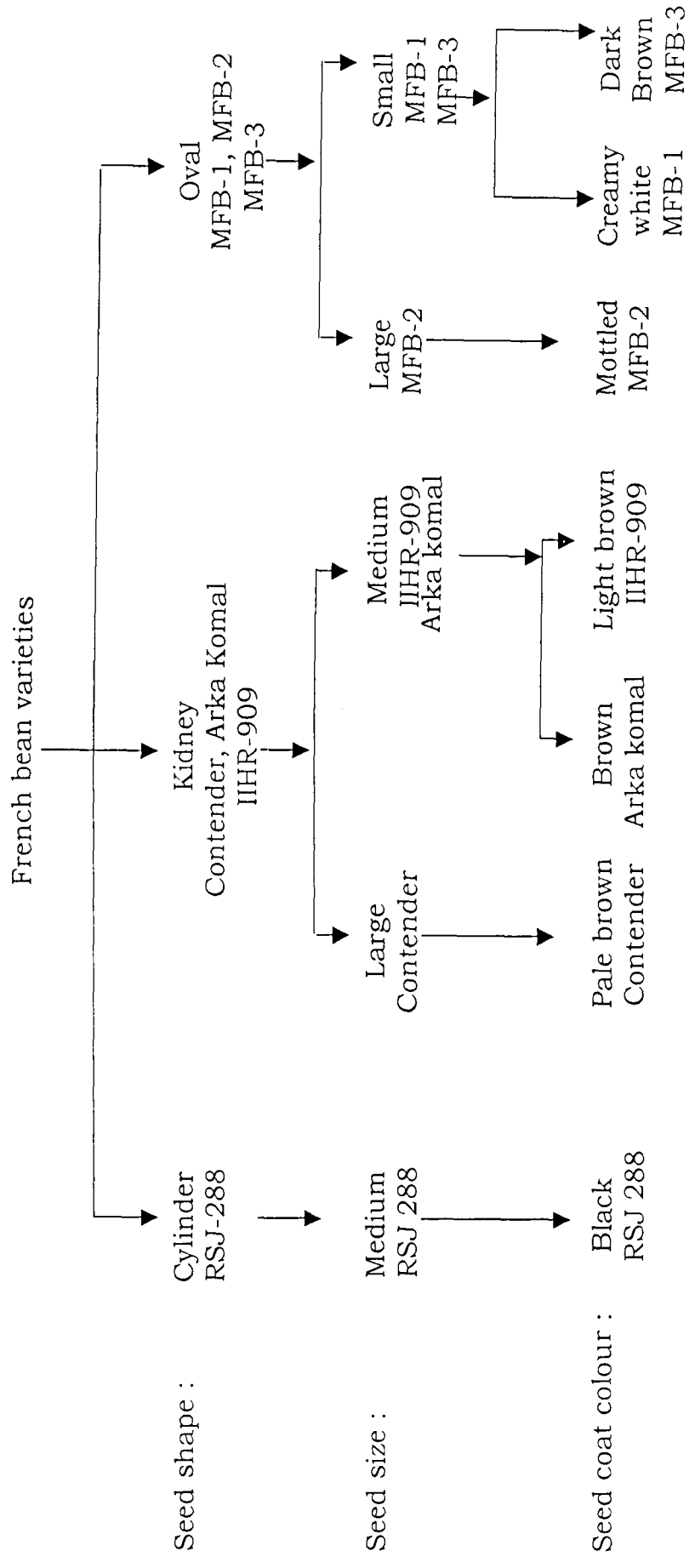


Fig. 11 : Identification keys on the basis of seedling morphological characteristics in french bean varieties

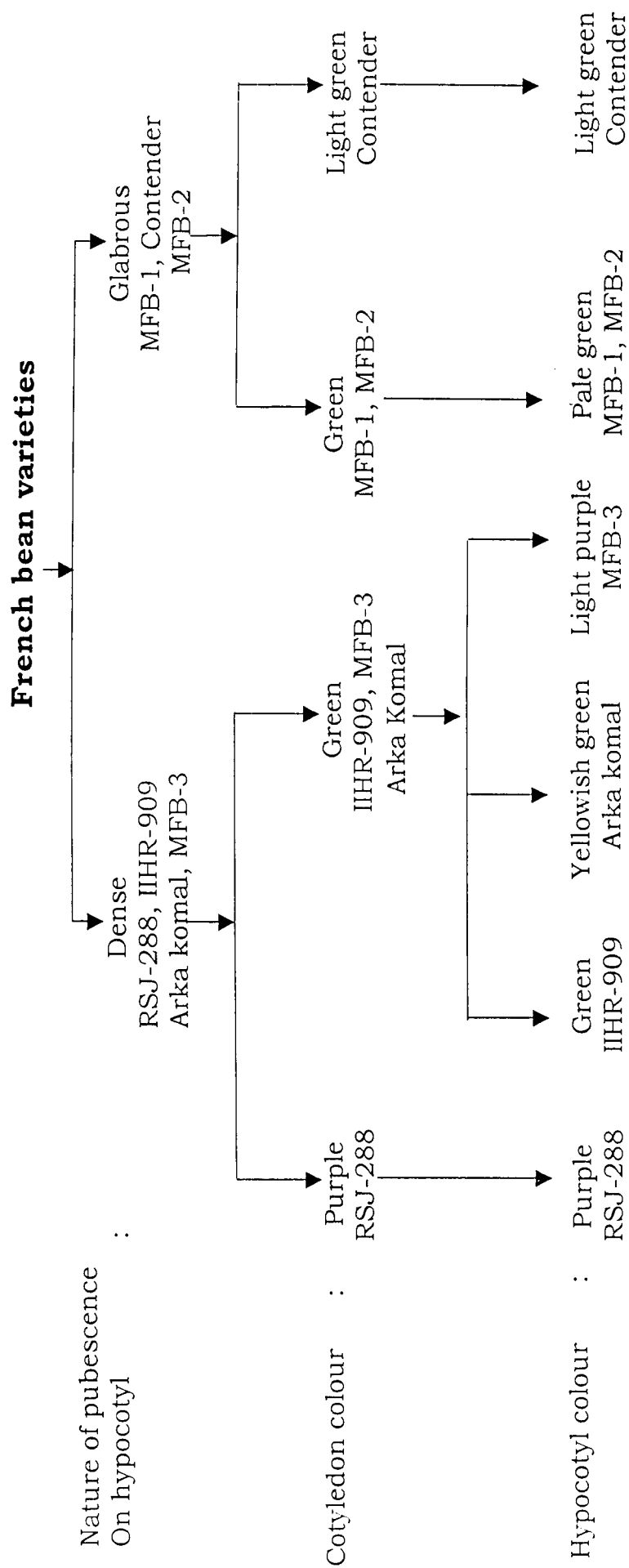
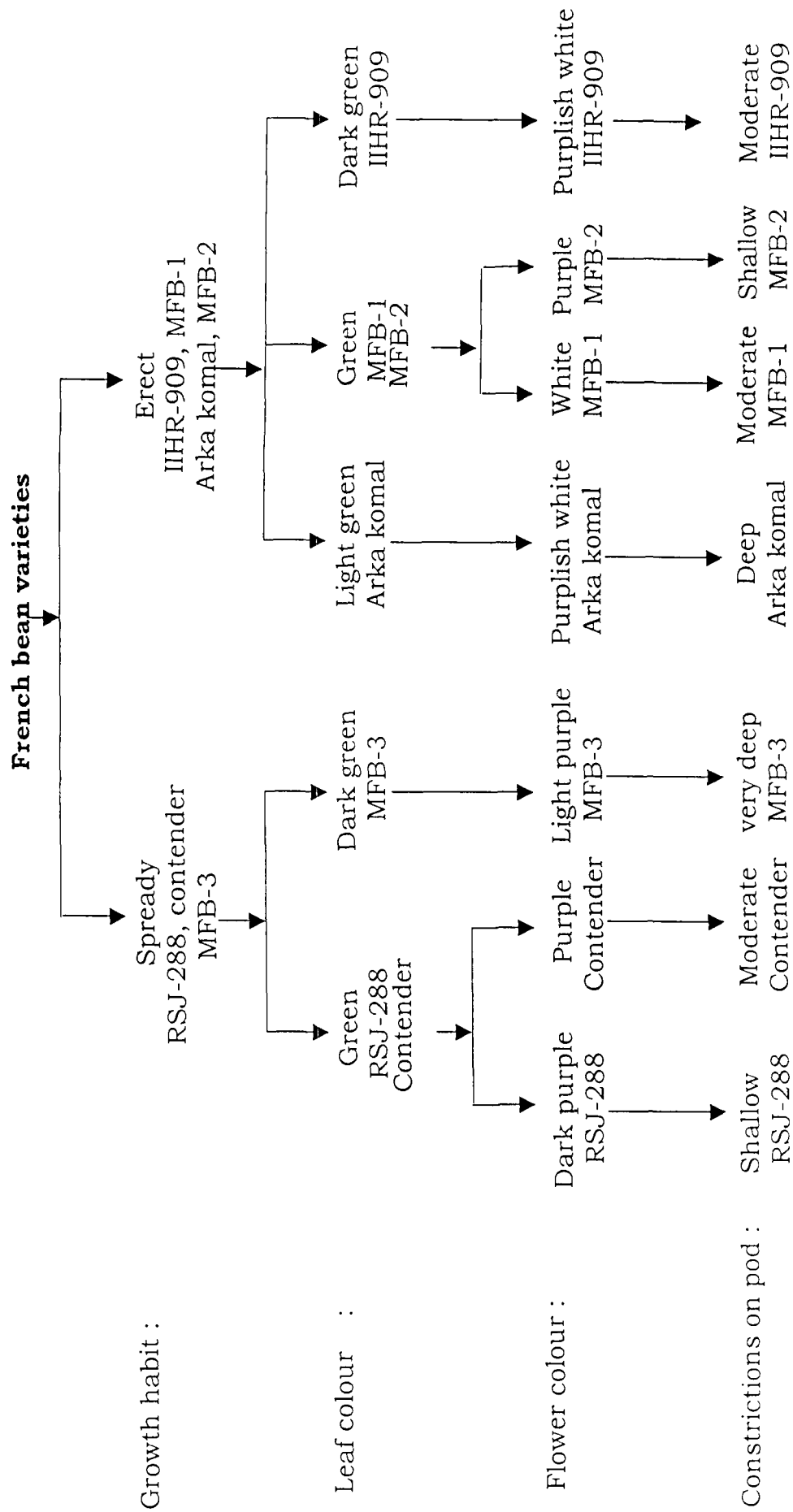


Fig. 12 : Identification keys based on plant morphological characteristics in french bean varieties



5.3.2 Electrophoretic identification

5.3.2.1 Protein and isozyme polymorphism for varietal identification

The rapid pace of new crop varieties has pointed out the need of cultivar identification and protection of proprietary rights of both old and newly developed cultivars to meet this end. Electrophoresis of proteins has emerged as an efficient, single and reliable tool supplementing traditionally used morphological parameters (Picket and Jerman, 1994). Proteins and isozymes because of their stability and reproducibility have potential to serve as tools for fingerprinting of genotypes. In the present study, seven genotypes were characterized for the protein and isozyme profiles.

Native poly acrylamide gel electrophoresis profile of hypocotyl proteins resulted with three electrophoretic phenotypes. RSJ-288 was differentiated by other varieties as it lacked a band at Rm value of 0.045. varieties namely MFB-1, Contender, Arka Komal and MFB-2 were categorized under one class as they lacked a band at Rm value of 0.161 IIHR-909 and MFB-3 were categorised under third group as they lacked a band at Rm value of 0.563 using a combination of band number and band location. This is in agreement with seven cultivars of frenchbean discriminated by following a native PAGE electrophoresis by Hussain *et al.* (1986) and electrophoretic evaluation of french bean cultivars done by Bassiri *et al.* (1978).

Single monomorphic band observed with peroxidase isozyme at Rm value of 0.285 could not differentiate the undertaken french bean varieties. Hence, there is a need to go for some more isozymes.

5.3.2.2 RAPD analysis for cultivar identification

The morphological and protein markers have employed in identifying the crop varieties of a species. The accurate identification of varieties is questionable because of availability of low number of morphological and biochemical markers, their poor or unknown genetic control and environmental influence in the phenotypic expression of the variety that lead to confusion and inconclusive. Since then an advanced technique, the identification at DNA level is preferred now a days.

The use of RAPD markers as a tool to identify french bean varieties was explored to circumvent the laborious classical approach involving the study of morphogenetical characters. This method has a high discriminatory potential as it probes the nucleotide composition of a gene rather than its products. Another advantage of this method lies in the availability of innumerable primers for screening purposes. RAPD's has been extensively used in identification of crop varieties (Baum *et al.*, 1998) and Mc. Donald *et al.*, 1994).

In the present study seven varieties of french bean were analysed using RAPD techniques, the results of RAPD are discussed in this chapter.

The primers of OPF series are screened. Among them, OPF-7, OPF-10, OPF-16 and OPF-20 exhibited polymorphic banding pattern. When OPF-7 primer is used, Contender and MFB-2 varieties exhibited an extra band of 4000 bp. These two varieties having a common seedling morphogenetic character like without pubescence (Glabrous) on hypocotyl.

The varieties such as, RSJ-288 and Contender are lacking a band of 600 bp, when primer OPF 10 is used for screening. RSJ-288 is similar with other varieties like IIHR-909, Arka Komal and MFB-3 in morphoogical characters like dense pubescence on hypocotyl and spready

growth habit. By RAPD analysis, RSJ 288 is uniquely identified based on absence of 600 bp band.

When the primer OPF-16 used it distinctly identified the varieties namely MFB-1 and MFB-2, which are having an extra band of 1750 bp.

When RAPD analysis is done using primer OPF-20, maximum varieties exhibited polymorphic banding pattern. Three varieties namely Contender, MFB-3 and MFB-1 can be distinctly identified using this primer though these varieties are similar for the character of glabrous hypocotyl (no hairs on hypocotyl).

The similar RAPD analysis method was employed by (Sengupta *et al.*, 2002) to identify seven rice varieties using three primers, the banding pattern (or profiles) is the key basis for identification of varieties, such techniques were tried by Noli *et al.* (1999) in tomato varieties, Nkongolo, (2003) in cowpea and Scott *et al.* (1994) in common bean varieties.

PRACTICAL APPLICATION OF RESULTS

The information generated with the present investigation the following recommendations can be made.

1. The spacing level of 30x20 cm could be adopted to get maximum seed yield with better quality seeds.
2. The phosphorus level of 125 kg P₂O₅ per ha produced maximum seed yield of 21.57 q per ha by recording 21.24 per cent more over 75 kg P₂O₅ per ha.
3. The spacing 30x20 cm coupled with application of 125 kg P₂O₅ per ha resulted in maximum seed yield of 24.54 q per ha with high quality seeds.

4. Prominent variety could be screened out using seed yield and quality parameters.
5. French bean varieties were identified with unique morphological character, *viz.*,
Variety RSJ-288- Black seed coat colour,
IIHR-909- Dense hairiness on hypocotyl and
MFB-1 – Erect plant growth habit.
6. French bean varieties could be identified by using RAPD technique as a DNA marker, *viz.*,
Variety Contender-identified with polymorphic bands of 1400,
2200 and 300 bp, using primer OPF-20 and
MFB-2 – identified with polymorphic bands of 1750 bp using primer OPF-16.

FUTURE LINE OF WORK

1. Studies on the effect of micronutrients through soil and foliar application on performance of french bean can be taken up.
2. Effect of seed treatment of growth regulators on seed yield and quality can be studied.
3. Storability studies may be taken up to know the storability of seeds with different seed treatments and packaging materials.
4. Effect of various organic manures on seed yield and quality may be studied.
5. The repeatable DNA markers like SSRs and ISSRs could be used for the varietal identification.

Summary

VI SUMMARY

A field experiment was conducted at the Main Agricultural Research Station, College of Agriculture, Dharwad during *kharif* season of 2002, to study the “Effect of spacing and phosphorus levels on growth, seed yield and quality of French bean cv. Arka Suvidha (IIHR 909)” and varietal identification studies were conducted at NSP/BSP Unit. The results of the present investigation were summarised below.

6.1 EFFECT OF SPACING AND PHOSPHORUS LEVELS ON GROWTH, SEED YIELD AND QUALITY OF FRENCH BEAN cv. ARKA SUVIDHA (IIHR-909)

The experiment consisted of 12 treatment combinations with four levels of spacing (30x10, 45x10, 30x20 and 45x20 cm) and three levels of phosphorus (75, 100 and 125 kg P₂O₅ /ha).

The wider spacing of 45x20 cm recorded significantly higher number of branches, number of leaves and dry weight of plant compared to other spacing of 30x10, 45x10 and 30x20 cm. But the closer spacing of 30x10 cm recorded maximum plant height.

There was significant increase in number of pods, number of seeds per pod, seed weight per pod and seed weight per pant at 45x20 cm spacing, whereas maximum seed yield (21.4 q/ha) was recorded at the spacing of 30x20 cm which is 13.36 per cent more than 45x20 cm spacing (18.8 q/ha) and was on par with the 30x10 cm spacing (20.4 q/ha).

Seed quality parameter *viz.*, germination per cent (92.08%) was maximum at 30x20 cm spacing which was on par with 45x20 cm (91.50%) whereas parameters like root length, shoot length, vigour index not varied

significantly. The seed protein per cent was maximum at the wider spacing of 45x20 cm (23.60) followed by 30x20 cm spacing (22.20).

Application of 125 kg P₂O₅ per ha recorded significantly higher plant height, number of branches, number of leaves and plant dry weight compared to the application of 100 and 75 kg P₂O₅ per ha.

Significant increase in number of pods per plant, number seeds per pod, seed weight per pod, seed recovery and seed yield per ha with 125 kg P₂O₅ per ha. Seed yield was 21.24 and 9.23 per cent higher than 75 (17.80 q), 100 kg P₂O₅ per ha (19.75 q).

The seed quality parameters *viz.*, germination percentage, rate of germination, root length, shoot length, seed vigour index, seed protein per cent, seedling dry weight were higher at 125 kg P₂O₅ per ha compared to 100 and 75 kg P₂O₅ per ha.

The spacing 45x20 cm spacing with application at 125 kg P₂O₅ per ha recorded maximum number of branches per plant (12.20), leaves per plant (14.81) and plant dry weight (14.18 g) at harvest. Whereas, plant height was maximum with combination of 30x10 cm spacing at 125 kg P₂O₅ per ha.

There was significant increase in number of pods per plant (16.96), seeds per pod (5.61), seed weight per pod (1.80 g), seed weight per plant (26.90 g), with interaction of wider spacing 45x20 cm and 125 kg P₂O₅ per ha. Whereas, seed yield per ha was maximum (2454 kg) in the interaction level of 30x20 cm spacing with 125 kg P₂O₅ per ha, which was on par with 30x10 cm with 125 kg P₂O₅ per ha (2259 kg).

Seed quality parameters like germination per cent, seedling dry weight and 100 seed weight were maximum at the combined levels of

45x20 cm spacing with 125 kg P₂O₅ per ha, which are on par with the interaction level of 30x20 cm spacing with 125 kg P₂O₅ per ha whereas other parameters like root length, shoot length and seed vigour index did not vary significantly among the interactions.

6.2 SCREENING OF FRENCH BEAN VARIETIES FOR SEED YIELD AND QUALITY PARAMETERS

Among the french bean varieties used for screening, RSJ-288 recorded maximum pods per plant (12.0) and seed weight per plant (18.90 g) which in turn recorded maximum yield per ha. In seed quality parameters also RSJ-288 recorded maximum seedling dry weight (28.3 mg), 100 seed weight (38.93 g) and low EC value (0.696 dSm⁻¹) with better germination per cent. Looking into all seed yield and quality characters RSJ-288 can be considered as prominent variety among the varieties of french bean studied.

6.3 VARIETAL IDENTIFICATION OF FRENCH BEAN CULTIVARS

6.3.1 Morphological characters

Varietal identification and cultivar purity testing is very important aspect in seed production. All sectors of seed industry benefit from the ability to assess cultivar purity and identity. Therefore, information on well expressed and distinct characters of frenchbean variety should be made available to the seed producing and certification agencies in order to monitor the genetic purity of seeds.

Therefore, a study was undertaken at the Main Agricultural Research Station, College of Agriculture, Dharwad reveal the objectives of varietal differences in seed, Seedling and plant morphological characters of french bean under field conditions.

Based on seed shape, varieties were categorised as cylinder kidney and oval shape. Seeds also categorised them on size as large, medium and small. Prominent seed coat colours observed were black, brown and white and some more classes on the different intensities of these colours are also done. Based on the seed shape RSJ-288 was individually categorised as cylinder, large size with black seed coat colour.

Based on seedling characters like pubescence on hypocotyl, varieties are categorised into two classes Dense (hairy) and Glabrous (without hairs). Seven french bean varieties are categorised into purple, green and light green based on cotyledon colour, hypocotyl colour like purple, green and their different intensities are used to classify the french bean varieties into six classes.

The plant morphological characters under field conditions are exhibited widely. Varieties are grouped on the basis of following plant morphological characters like growth habit (spread, erect), leaf colour different intensities of white and purple leading to five classes) and constrictions on pod (shallow, moderate, deep and very deep).

Varietal identification keys for french bean varieties are also developed based on different combinations of various characters studied. These identification keys can be used as ready reckonor for determination of genetic purity of seed lot.

6.3.2 Protein and isosyme polymorphism

Seven genotypes of french bean were evaluated for protein and isozyme polymorphism. The hypocotyl protein profile had three electrophoretic phenotypes. RSJ-288 was individually categorized with the absence of polymorphic band at Rm value 0.045 whereas, MFB-1,

Contender, Arka Komal and MFB-2 categorised with the absence of polymorphic band at Rm value 0.161 under second class. MFB-2, MFB-3 and IIHR-909 were grouped under third phenotypic class having no bands at Rm value 0.161 and 0.536.

The only one isozyme namely peroxidase was tried. It resulted with monomorphic bands in all the varieties at Rm value 0.285 and could not categorise the varieties.

6.3.3 RAPD analysis

RAPD analysis is one of the advanced technique can be used to identify the varieties in french bean. In the present study four primers such as OPF-07, 10, 16 and 20 are used to identify the varieties. The Contender and MFB-2 varieties exhibited an extra band of 4000 bp, when OPF-7 primer is used. The varieties RSJ-288 and Contender are identified based on the absence of 600 bp band, when OPF-10 primer is used.

Using OPF-16 primer, the varieties MFB-1 and MFB-2 are identified based on the presence of an extra band of 1750 bp. Using OPF-20 primer in the RAPD analysis. It is indicated that the presence of three extra bands of 1400, 2200 and 3000 bp in Contender, only one extra band of 1400 bp in MFB-3 and the absence of 700 bp band in MFB-1.

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STUDIES ON SPACING AND PHOSPHORUS LEVELS ON SEED YIELD AND QUALITY, AND VARIETAL IDENTIFICATION IN FRENCH BEAN

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ABSTRACT

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Three experiments were carried out at University of Agricultural Sciences, Dharwad. In first experiment, the spacing levels of 30 x 10, 45 x 10, 30 x 20 and 45 x 20 cm were combined with three phosphorus levels of 75, 100 and 125 kg P₂O₅ per ha to study the seed yield and quality. Maximum plant dry weight (14.18 g) at harvest was observed at spacing level of 45 x 20 cm in combination with 125 kg P₂O₅ per ha. Maximum seed yield (2454 kg/ha) was obtained with 30 x 20 cm in combination with 125 kg P₂O₅ per ha which was on par with 30 x 10 cm coupled with 125 kg P₂O₅ per ha (2259 kg/ha). The seed quality parameters like hundred seed weight (37.83 g), germination percent (93.75) and reduced electrical conductivity (1.127 dSm⁻¹) were recorded with spacing of 45 x 20 cm combined with 125 kg P₂O₅ per ha which was on par with 30 x 20cm and 125 kg P₂O₅ per ha.

In second experiment, seven french bean varieties namely RSJ-288, IIHR-909, MFB-1, Contender, Arka Komal, MFB-2, and MFB-3 were screened for seed yield and quality parameters. Variety RSJ-288 produced maximum number of branches per plant (11.20), seed weight per plant (18.90 g), hundred seed weight (38.93 g) and low electrical conductivity value (0.696 dSm⁻¹) over other studied varieties.

In third experiment, the above mentioned varieties were identified through morphological analysis and electrophoretic techniques. Seed, seedling and plant morphological characters were recorded. Total protein electrophoresis resulted with three electrophoretic phenotypes due to banding pattern at R_m values 0.045, 0.161 and 0.563. There was a monomorphic band at R_m value of 0.285 for peroxidase isozyme for all french bean varieties. RAPD analysis for the same varieties was tried with OPF primer series. The four primers namely OPF-7, 10, 16 and 20 resulted with polymorphic bands identifying each varieties uniquely.