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Survey, collection and seed morphometric characterization of French bean (*Phaseolus vulgaris* L.) landraces of Himachal Pradesh

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ABSTRACT

The study was conducted to survey and collect various French bean genotypes from the main hot spot regions of Himachal Pradesh like Shimla, Chamba, Sirmour, Kullu, Kinnaur and some areas of Mandi and Solan district. Different bean genotypes were collected in a 15-day exploration trip from various geographical regions. The collected seed samples were evaluated and characterized for intra-specific seed morpho-metric characteristics like seed coat colour, hilum colour, seed shape, hypocotyls pigmentation, cotyledon colour pubescence on hypocotyls, 100 seed weight (g), seed length (mm) and seed width (mm). The evaluated genetic material was then deposited in the short term seed bank of Department of Seed Science and Technology UHF Nauni for future correspondence. The genetic diversity found among the landraces is of great importance in the utilization of the species for food and nutrition and more importantly for genetic improvement. The study would prove a basic primary step in germplasm recognisition activity for future breeding programmes and will project the local available genetic footprints to entire agro associated enterprises for necessary consideration.

Key words: French bean genotypes, Seed morphometric characteristics, Survey, Variability.

INTRODUCTION

French bean (*Phaseolus vulgaris* L. 2n=2x=22) belongs to family Leguminosae and sub family Papoilionacae. It has many synonyms like snap bean, kidney bean, haricot bean and also called "Rajmash" in Hindi. French bean originated from Central America and Peruvian Andes in South America, from where it spread to Europe during 16th and 17th centuries. It was introduced to India during 17th century form Europe (Prakash and Ram, 2014). French bean is consumed as immature tender pods, green grains as vegetables and dry grains. Beans, the "meat of the poor", contribute essential protein to the under-nourished people of the developing countries. Besides this, dry beans are also rich in calcium required for bone structure and general health, in iron required for blood making, in different types of vitamin B which are effective on the nervous system (Karasu and Oz, 2010). Medicinal properties include control of diabetes, cardiac problems and natural cure for bladder burn. It has both carminative and reparative properties against constipation and diarrhoea respectively (Duke, 1981). As per the FAO estimates, it is grown in the world in an area of 28 million hectares with annual production of 20 million tons and productivity 729 Kg/ha (Prakash and Ram, 2014).The total area in India under bean cultivation is 134 thousand hectares with annual production of 1165 thousand tons (Anonymous, 2015).

Himachal Pradesh is endowed with varied agro-climatic zones from sub tropical to dry temperate desert and possesses a distinct position among other states with regard to bean germplasm. Anonymous (2013-14) reported nearly 3.44 thousand hectares area under bean cultivation with the production of 40.88 thousand tons in Himachal Pradesh. The main hot spot regions of French bean cultivation in Himachal Pradesh are Shimla, Chamba, Sirmour, Kullu, Kinnaur and some areas of Mandi and Solan district.

In Himachal Pradesh, French bean is mainly grown as an intercrop in temperate orchards or as mixed crop with maize. Thus it provides a sustainable opportunity to the farmers to gain more from the same piece of land and economic assurance against one crop failure under severe biotic and abiotic stress conditions. The importance of bean to the small scale farmers of Himachal can also be attributed to the fact that it is popularly marketed as 'Pahari Rajmash' which fetch more price in the market.

Nature and evolution are responsible for variation at species and genetic level. Climate has direct influence on species distribution, establishment and adaptation. Landraces are the outcome of such biological phenomenon. Landraces have been defined as dynamic populations of a cultivated plant with a historical origin, distinct identity, often genetically diverse and locally adapted, and associated with a set of farmers' practices of seed selection and field

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management as well as with a knowledge base (Bellon and Etten, 2014). For thousands of years, farmers have adapted their crops and their cropping systems in accordance to environmental change. Now, the world is under severe pressure from climate adversities, mainly global warming, desertification, drought, floods, landslides, storm surge, soil erosion, and saline water intrusion. Moreover, the atrocities brought about by the growing human pollution, urbanization and industrialization are likely to present unprecedented challenges to the agriculture sector and future plant breeders.

French bean germplasm of Himachal Pradesh that possess tremendous genetic polymorphism is also at the verge of facing the above stated consequences. Further, the modern day priority towards mono-cropping agriculture mainly concentrates on major cash crops like apple, pea, cauliflower, cabbage, capsicum, tomato, stone fruits from mid hill to high hill zones of Himachal Pradesh which has limited the growing space for diverse genetic resource of French bean. Therefore, a comprehensive conservation strategy has to be developed to protect the diverse genetic resource. This is because these locally adapted genotypes can provide a cost-effective way to manage pests and diseases and climate risks. These landraces have a potential to contribute to health and nutrition, provide genes for breaking yield plateau and sustain improvement in varietal development programmes. Climate change and food security are the two important drivers that enforces agronomist to realize the importance of diversified sustainable agriculture. The precedence of scientific personals needs to be reallocated towards saving the precious local available genetic footprints for future breeders.

In consideration to the above facts, the research was designed with two broad objectives. First to conduct survey of bean germplasm in different districts of Himachal Pradesh and record its geo ecological position. Second to conduct seed morphometric characterization for assessment of intra - specific variability.

MATERIALS AND METHODS

The present investigation was carried out in the Department of Seed Science & Technology, Dr. Y.S. Parmar University of Horticulture and Forestry, Nauni, Solan, H.P. during the year 2015. Himachal Pradesh has four agro climatic zones *viz.*, mid hills sub humid, high hills temperate wet, high hills temperate dry and sub mountain low hills sub tropical and nine different agro-ecological zones based upon two major determinants elevation and precipitation (Table 1).

Table 1: Agro ecological zones of Himachal Pradesh

Agro ecological zones	Altitude range (m)	Rainfall (mm)
Zone 1.1	240-1000	< or =1500
Zone 1.2	240-1000	>1500
Zone 2.1	1001-1500	< or =1500
Zone 2.2	1001-1500	>1500
Zone 3.1	1501-2500	< or = 1500
Zone 3.2	1501-3250	> 1500
Zone 4.1	2501-3250	< 700 (Dry)
Zone 4.2	3251-4250	Dry/snow
Zone 4.3	>4250	Dry/snow

Collection and acquisition of seed samples: During the survey and exploration trip, 36 sites from districts viz., Shimla, Chamba, Sirmour, Kullu, Kinnaur and some areas of Mandi and Solan were visited and 60 indigenous genotypes of French bean were collected (Table 2). Samples collected comprised of freshly harvested seeds (1 Kg each or less depending upon the availability). The collection was made from farm store, farmer's field and threshing yards. Each collected sample was brought to the laboratory where

Table 2: Survey and Collection of French bean germplasm

District	Agro ecological zone	Agro climatic zone	Number of germplasm	Genotype collection no.	Place of Collection
Kinnaur	3.2, 4.1, 4.2, 4.3	High hills, Temperate dry	10	RL-1 to RL-10	Sapni; Kanai (Karcham); Kalpa; Poweri; Kanam (Poo); Sangla
Chamba	3.1, 3.2, 4.1, 4.2	High hills, Temperate dry	10	RL-11 to RL-20	Satnala (Bharmaur); Pangi; Panjsei (Bharmaur)
Shimla	3.1, 3.2, 4.1, 4.2	High hills, Temperate wet	17	RL-21 to RL-37	Nerwa (Chopal); Sunarly (Chopal); Kotgarh; Arunakshi (Rampur); Dhar Gaura (Rampur); Gassopul (Rampur);Tangru-Jhakri (Rampur); Khashdhar (Chirgaon); Dhanwari (Chirgaon); Sindasali (Chirgaon); Khaltudhar (Kumarsain)
Kullu	3.1, 3.2, 4.1, 4.2	High hills, Temperate wet	9	RL-38 to RL- 46	Khanag; Anni; Banjar; Bhuthi
Mandi	2.1, 2.2, 3.1	Mid hills, Sub humid	7	RL-47 to RL-53	Ghaniya; Neeru; Somgad; Rahidhar (Thunag)
Solan	1.1, 1.2, 2.1, 2.2	Sub mountain, Low hills, Sub tropical	4	RL-54 to 57	Kothi (Dharja); KotlaNagali; Nauni
Sirmaur	1.1, 1.2, 2.1, 2.2, 3.1	Mid hills, Sub humid	3	RL-58 to RL-60	SerJaghas(Rajgarh); Ratoli(Rajgarh) ; Phabiana (Rajgarh)

it was cleaned, assigned the collection number constituting of alpha numeric character like Rajmash Local 1 (RL-1), Rajmash Local 2 (RL-2) until every sample had its own identification number and stored in air tight plastic containers. The seeds were evaluated for different qualitative and quantitative characteristics. Qualitative traits included seed coat colour, hilum colour, seed shape, hypocotyls pigmentation, cotyledon colour and pubescence on hypocotyls whereas, quantitative traits comprised of 100 seed weight (g), seed length (mm) and seed width (mm) to distinguish the different strains. A single common strain was then stored in the seed bank of the department for future crop improvement studies.

Seed morpho-metric characterization: The laboratory experiment was laid out in completely randomized design (CRD) using three replications. Qualitative characters were observed on the basis of visual evaluation while quantitative characters were measured and recorded in SI units. The seed coat colour of all the genotypes were observed under natural day light using Royal Horticulture Society (RHS) colour charts. Similarly, the seeds of different genotypes were observed for hilum colour under natural day light. The seeds of collected genotypes were observed under magnifying lens of purity work board and grouped as cuboid, oval, truncate and kidney shaped depending on their seed shape. The hypocotyl colour, cotyledon colour and pubescence on hypocotyl was observed under natural day light condition after five days of seed sowing in the paper towel at 25^0C and 85 to 90% relative humidity inside seed germinator.

The hundred seeds from each genotype were counted and weight was recorded by using electronic balance as per procedure by ISTA (Anonymous, 1999). The average weight was recorded in grammes. Seed length of 10 seeds of different genotypes was measured using vernier calliper from the base to tip portion of the seed i.e. parallel to the hilum. Similarly seed width of same 10 seeds used for length measurement was measured using vernier caliper from hilum to opposite side of seed. The mean was recorded in millimeter.

RESULTS AND DISCUSSIONS

Qualitative traits: The sixty French bean genotypes were grouped individually into eleven categories as greyed orange, greyed purple, greyed yellow, greyed red, greyed white, orange white, red purple, purple white, black and orange (Table 3 and Plate 1 and 2). Chattopadhyay and Dutta (2010) studied the seed colour of 15 genotypes of Dolichos bean. One genotype showed brown seed, five genotypes had reddish purple seed, five genotypes had white seed, three genotypes had greyish yellow seed and one genotype had greyish orange seed. Stoilova *et al*. (2013), Bode *et al*. (2013) also reported different seed colours in French bean.

The hilum colour of all the French bean genotypes was creamish white (Table 3). Rania *et al*. (2010) studied the various seed characters of *Phaseolus vulgaris.*They observed that seeds were white in colour, large in size, kidney shape with white hilum.

The seeds of different genotypes were categorized as cuboid, oval, truncate and kidney shaped depending on their seed shape (Table 3 and Plate 1 and 2). Singh *et al*. (2014) estimated the morphological variability of 18 French bean varieties collected from ICAR institutes and SAUs for seed morphological traits. The varieties, *viz.*, Pusa Himalaya showed circular to elliptic seed shape, Kentucky Wonder and Swarna Lata showed elliptic and rest 15 varieties showed kidney shaped seed however, none of the variety had round seed shape.

The data on hypocotyl pigmentation revealed that all the 60 French bean genotypes exhibited creamish white hypocotyl pigmentation (Table 3). Chandrashekhar (2005) indicated expression of hypocotyl colour in different French bean genotypes as purple, light green, light purple and pale green.

French bean genotypes were classified based on cotyledon colour as yellow, yellow with red streaks and yellow with purple streaks (Table 3). Genotype (RL – 20) showed yellow with red streaks cotyledon colour, genotype (RL – 46) exhibited yellow with purple streaks while the rest of the genotypes had yellow coloured cotyledon. Neupane *et al*. (2005) investigated 100 accessions of local and exotic bean germplasm for agro morphological characteristics at Agriculture Research Station Jumla. Examination of seedlings revealed that hypocotyl pigmentation of germplasm ranged between purple, green and others colors. 63 accessions had green, 25 purple and 6 had other pigments in hypocotyls. Fifty-four accessions showed green color of the emerging cotyledons whereas two had purple, 15 pale green and the rest had red color of the emerging cotyledon.

Based on presence of pubescence on hypocotyl, all the French bean genotypes were categorized as glabrous (without hairs) and data is presented in Table 3. Prashanth (2003) characterized seven French bean genotypes based on seedling pubescence (glabrous and dense).

Quantitative traits: The perusal of data on 100 seed weight categorized 60 indigenous French bean genotypes into light (<32.5 g), medium (32.5 to 42.5 g) and heavy (>42.5 g) category (Table 4). Twenty two genotypes were grouped in light weight, eleven in medium weight and twenty seven in heavy weight category.

The data on seed length classified 60 French bean genotypes into three classes (Table 4). The seeds which measured less than 14.0 mm were grouped as small (34), the seeds that measured between 14.0 to 15.5 mm were

Table 3: Qualitative seed morpho-metric characteristics of indigenous French bean (*Phaseolus vulgaris* L.) landraces.

Genotype	Seed Colour	Hilum Colour	Seed Shape	Hypocotyl Pigmentation	Cotyledon Colour	Pubescence on hypocotyls
RL-1	a) Greyed Orange group (164 – C) b)Greyed Purple group (185 – A	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-2	White group (155 – D)	Cremish white	Oval	Creamish White	Yellow	Absent
RL-3	Greyed Orange group (165 – D)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-4	Greyed Purple group (187 – B)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-5	a) Greyed Orange group (165 – B) b) Greyed Purple group (183 – B)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-6	a) Greyed Orange group (167 – A) b) Greyed Orange group (166 – B)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-7	Greyed Yellow group (162 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-8	Greyed Purple group (185 – A)	Cremish white	Kidney	Creamish White	Yellow	Absent
RL-9	Black group (202 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-10	Greyed Yellow group (162 – B)	Cremish white	Oval	Creamish White	Yellow	Absent
RL-11	Greyed Orange group (163 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-12	Greyed Purple group (183 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-13	a) Greyed Orange group (172 – A) b) Greyed Red group (178 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-14	Greyed Purple group (187 – B)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-15	Greyed Purple group (187 – A)	Cremish white	Oval	Creamish White	Yellow	Absent
RL-16	a) Greyed Orange group (165 – C) b) Greyed Orange group (175 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-17	Greyed Purple group (187 – B)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-18	a) Greyed Orange group (168 – D) b) Greyed Red group (178 – A)	Cremish white	Kidney	Creamish White	Yellow	Absent
RL-19	Greyed Orange group (164 – A)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-20	Greyed Yellow group (161 – A)	Cremish white	Cuboid	Creamish White	Yellow with red tinge	Absent
RL-21	Greyed Purple group (187 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-22	a) Greyed Purple group (187 – A) b) Greyed Red group (180 – B)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-23	Greyed Orange group (164 – B)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-24	Greyed Purple group (183 – B)	Cremish white	Kidney	Creamish White	Yellow	Absent
RL-25	a) Greyed Purple group (183 – B) b) Greyed Orange group (163 – C)	Cremish white	Kidney	Creamish White	Yellow	Absent
RL-26	Red Purple group (59 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-27	Greyed Purple group (187 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-28	Greyed White group (156 – C)	Cremish white	Oval	Creamish White	Yellow	Absent
RL-29	Red Purple group (59 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-30	a) Purple group (79 – A) b) Greyed Orange group (164 – D)	Cremish white	Kidney	Creamish White	Yellow	Absent
RL-31	a) Orange White group (159 – A) b) Greyed Purple group (187 – A)	Cremish white	Cuboid	Creamish White	Purple	Absent
RL-32	a) Red Purple group (59 – A) b) Orange group (27 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-33	a) Greyed Orange group (165 – C) b) Greyed Orange group (175 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-34	a) Orange White group (159 – A) b) Greyed Purple group (185 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL- 35	White group (155 – D)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-36	White group (155 – D)	Cremish white	Oval	Creamish White	Yellow	Absent
RL-37	a) Greyed Orange group (165 – D) b) Greyed Purple group (185 – A)	Cremish white	Oval	Creamish White	Yellow	Absent
RL-38	Greyed Orange group (165 – A)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-39	Greyed Purple group (187 – A)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-40	Greyed Purple group (183 – B)	Cremish white	Truncate	Creamish White	Yellow	Absent

continue Table 3.....

continue Table 3.....

RL-41	a) Greyed Orange group (164 – B) b) Greyed Orange group (166 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-42	a) Greyed Orange group (165 – C) b) Greyed Orange group (173 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-43	a) Orange White group (159 – A) b) Greyed Purple group (187 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-44	Black group (202 – A)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-45	a) Greyed Orange group (164 – A) b) Greyed Purple group (183 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-46	Greyed Yellow group (161 – A)	Cremish white	Truncate	Creamish White	Yellow with purplish tinge	Absent
RL-47	a) Greyed Purple group (184 – A) b) Greyed Orange group (165 – C)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-48	Greyed Orange group (167 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-49	Greyed Orange group (163 – A)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-50	a) Greyed Orange group (165 – B) b) Greyed Orange group (165 – A)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-51	Greyed Yellow group (161 – A)	Cremish white	Oval	Creamish White	Yellow	Absent
RL-52	Greyed Orange group (163 – B)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-53	a) Greyed Purple group (185 – A) b) Greyed Orange group (164 – C)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-54	Greyed Purple group (185 – A)	Cremish white	Oval	Creamish White	Yellow	Absent
RL-55	Orange White group (159 – A)	Cremish white	Kidney	Creamish White	Yellow	Absent
RL-56	White group (155 – D)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-57	a) Greyed Orange group (165 – C) b) Black group (202 – A)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-58	a) Greyed Red group (182 – A) b) Greyed Orange group (165 – C)	Cremish white	Truncate	Creamish White	Yellow	Absent
RL-59	a) Greyed Orange group (165 – C) b) Greyed Red group (180 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent
RL-60	a) Orange White group (159 – A) b) Black group (202 – A)	Cremish white	Cuboid	Creamish White	Yellow	Absent

grouped as medium (15) and the seeds that measured more than 15.5 mm were considered as large (11).

Based on variation in seed width (Table 4), 2 French bean genotypes were classified as medium (5.0 to 6.0 mm) and 58 genotypes as large (>6.5 mm).

Brigitte (2006) studied the seed morphological characteristics of 18 different germplasm accessions of hyacinth bean (*Lablab purpureus*) and observed that seed sizes ranged from 5.7 to 14.3 mm in length and 4.0–8.6 mm in width. Similarly, Rania *et al.* (2010) studied the seed length

Table 4: Grouping of genotypes based on seed quantitative morpho-metric characteristics of indigenous French bean (*Phaseolus vulgaris* L.) landraces

Seed Characteristics	Number of genotypes	Genotypes
A. 100 seed weight (g)		
Light (<32.5 g), and	22	RL- 2, 4, 9, 15, 17, 19, 21, 23, 28, 29, 36, 38, 39, 40, 44, 46, 50, 52, 55, 56, 57, 60
Medium (32.5 to 42.5 g)	11	RL- 5, 14, 18, 20, 24, 33, 42, 45, 49, 58, 59
Heavy (>42.5 g)	27	RL- 1, 3, 6, 7, 8, 10, 11, 12, 13, 16, 22, 25, 26, 27, 30, 31, 32, 34, 35, 37, 41, 43, 47, 48, 51, 53, 54
B. Seed Length (mm)		
Small (<14.0 mm)	34	RL- 2, 4, 9, 10, 14, 15, 17, 18, 19, 21, 23, 28, 29, 30, 31, 33, 34, 35, 36, 37, 38, 39, 40, 44, 46, 49, 50, 51, 52, 53, 55, 57, 58, 60
Medium (14.0 to 15.5 mm)	15	RL- 5, 6, 11, 16, 20, 22, 24, 26, 41, 42, 43, 45, 47, 56, 59
Large (>15.5 mm)	11	RL- 1, 3, 7, 8, 12, 13, 25, 27, 32, 48, 54
C. Seed Width (mm)		
Small (<5 mm)	-	None
Medium (5.0 to 6.0 mm)	2	RL-2, 57
Large (>6.5 mm)	58	RL- 1, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 58, 59, 60

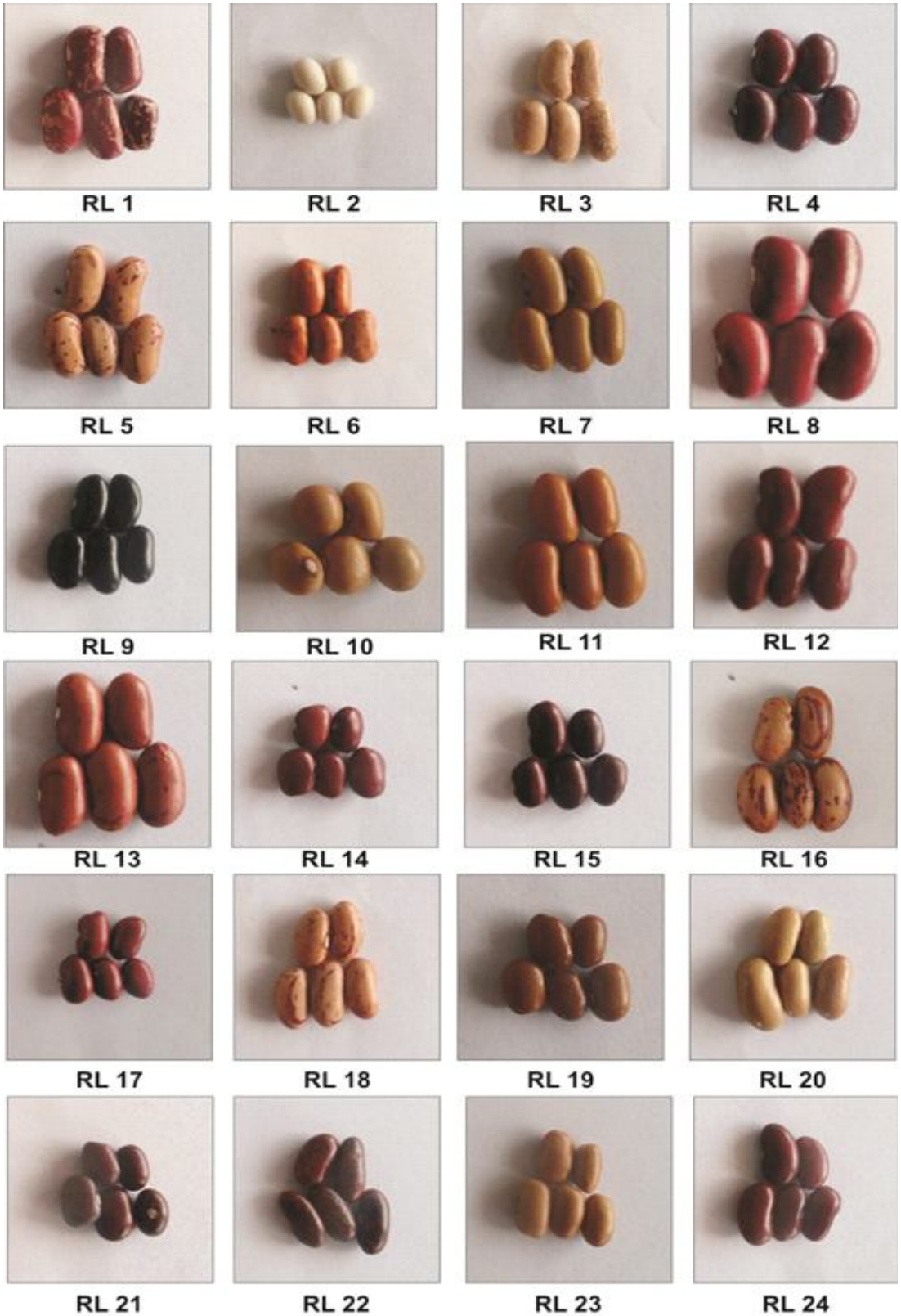

Plate 1: French bean germpiasm of Himachal Pradesh

Plate 2: French bean germpiasm of Himachal Pradesh

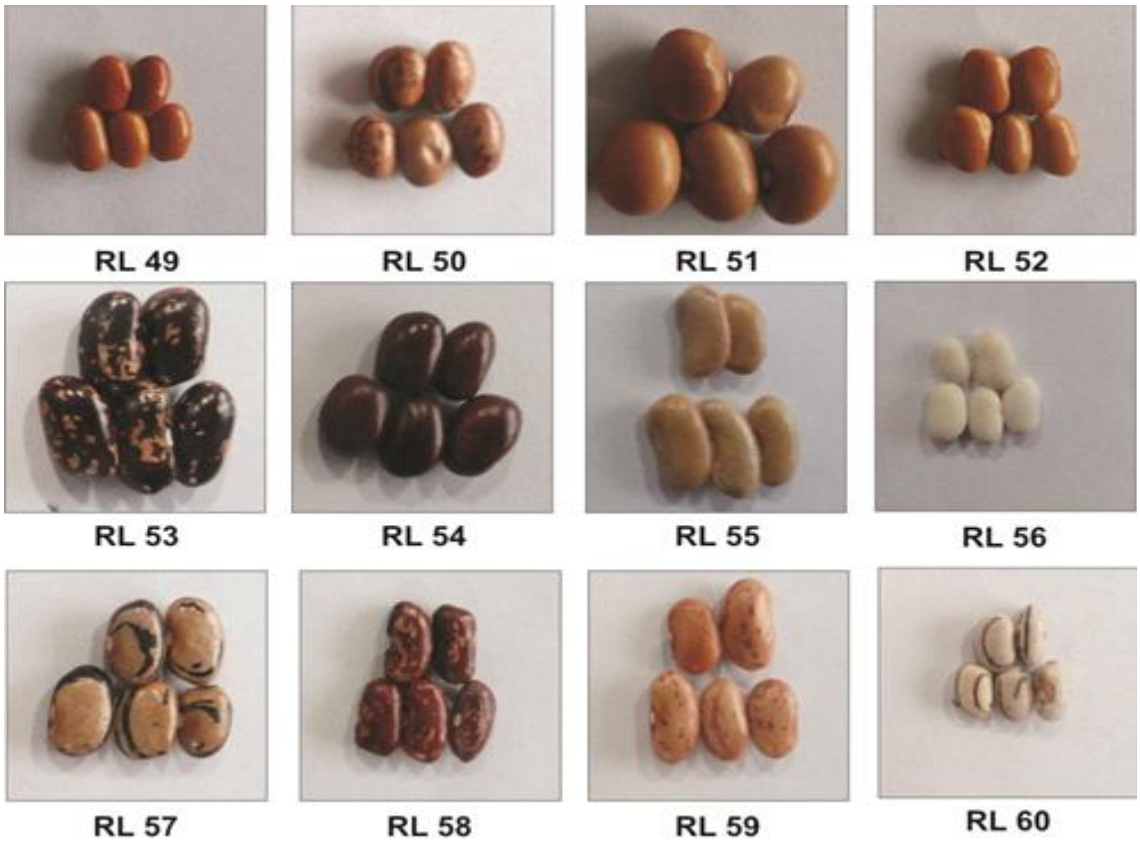

Plate 3: French bean germpiasm of Himachal Pradesh

and width of *Phaseolus vulgaris*. The authors observed that the seed length was about 1.6 cm and breadth about 0.7 cm.

The report provides a brief acquaintance of various indigenous French bean genotypes located at different geographical regions of Himachal Pradesh. There is revelation of population distribution and species diversity. Knowledge about the seed variation could prove useful for the breeders indulged in crop improvement programme to select distinguishing lines for breeding purpose. Further, conservationist can target their conservation strategy towards the desired region to safeguard the genetic material for future use.

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