Received: 31 May 2020 | Revised: 16 Jul 2021 | Accepted: 20 Jul 2021

Electronic Journal of Plant Breeding

Research Article

Microscopic characterization and performance analysis of chilli (*Capsicum annum*) cultivars

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Abstract
Chilli, also known as hot pepper is an important vegetable and spice crop having immense commercial and export value. The present study was an attempt to select an elite cultivar of Chilli based on their varietal performance, structural, microscopic characterization and elemental analysis. In varietal performance analysis, Kashi Anmol with yield 725.05 g/plant, total soluble solid 6.74° Brix and ascorbic acid 132.53 mg/100 g was found superior to all other cultivars. No major difference was observed in the backbone structures of the green chilli powder of all the cultivars during X-Ray Diffraction analysis. Scanning Electron Microscopy revealed spherical and amorphous in form of the powder. EDAX traces elements like Carbon (58.45-72.63%), Oxygen (26.25-39.47%), Chlorine (0.15-0.27%), Potassium (0.49-1.64%) in all the cultivars except Magnesium (0.26 & 0.22%) and Aluminum (0.15 and 0.22%) which was observed in only two cultivars. The EDAX study also confirmed the absence of any health hazardous elements in the varietal sample. The presented study suggests the cultivation of Kashi Anmol as an elite Chilli variety in the study area as compared to others.

Key words: Chilli, EDAX, Microscopic characterization, SEM, Varietal performance, XRD

INTRODUCTION

Chilli (*Capsicum annum* L.) also known as hot pepper, red pepper, bird pepper, cayenne, paprika belongs to the genus capsicum and family Solanaceae or nightshade family, having a diploid chromosome no. 2n=2x=24 (Rahevar *et al.,* 2021). It is grown in almost every part of the world and used mostly as a vegetable cum spice crop. The fruits are botanically known as a berry and the spiciness of chillies is due to the pungency of a crystalline acrid volatile alkaloid compound "capsaicinoids" (Abhinaya *et al.,* 2016). Chilli fruits are rich sources of vitamins A, C and E, and various secondary metabolites that can be used in the treatment of toothache, ulcers, rheumatism and diabetes. It was introduced by Columbus in Europe in the 15th century and spread to the rest of the globe along the spice trading routes to Africa, India, China and Japan. Portuguese introduced it to India from Southern parts through Brazil in 16th-century (Bosland and Votava, 2000; Kerketta *et al.,* 2018). Chilli cultivation spreads throughout India by the end of the 19th century (Datta and Das, 2013).

India is a leading consumer, producer and exporter of chilli in the world. India, provided with varied agro-climatic condition chilli is grown mostly in every part of the country but principally it is grown in states like Karnataka, Madhya Pradesh, Bihar, Andhra Pradesh, Maharashtra etc. Karnataka is the largest chilli growing state followed by Madhya Pradesh, Bihar and Maharashtra. India exported 44.90 thousand tonnes of Chilli (valuing 22,074.05 Lakh Rupees) in the year 2017-18 (Horticultural Statistics at

a glance, 2018). Area and production of chilli in India account for 309 thousand hectares and 3592 thousand metric tonnes, respectively (NHB database, 2018-19).

To analyze the effect of material and processing conditions on food structure microscopic examination is a useful tool (Wilson, 1991). Scanning electron microscopy (SEM) is an electron beam instrument used widely all over the world (Alsebaeai *et al.,* 2018). It can easily interpret and generates the nature of the micrographs of a given sample. X-ray diffraction (XRD) is a very effective technique for microscopic characterization and rapid analysis of crystalline structures of a given material. Energy dispersive X-ray analysis (E-DAX) provides details about the concentration of different elements present in a given sample. The present study aimed to analyse the performance of different cultivars under given climatic conditions. The structural characterization of green chilli powder was done by X-Ray diffraction, while the SEM was employed for microscopic characterization and elemental concentration was obtained by using E-DAX (Energy Dispersive X-ray Analysis).

MATERIALS AND METHODS

For the varietal performance analysis, the present investigation was carried out from month April to October 2019 at Horticultural Research Centre (HRC), Chauras campus while all the microscopic analysis was carried out at USIC, H.N.B.G.U, Srinagar (Garhwal), Uttarakhand, India. A total of five chilli cultivars *viz.,* Kashi Anmol, Taiwan-1, Srinagar Local, IC-119308 and IC-119290 were grown in a Randomized Block Design (RBD) experimental design with 3 replications each. The plants were grown at a spacing of 45 × 45 cm in squared plots of 1.35 × 1.35 m in size. The data was taken from 5 plants of each replication.

The quality analysis (Ascorbic Acid content, moisture content and total soluble solids) was done as per the procedure of Ranganna (2015) and AOAC (1995). Chlorophyll content was recorded by using a chlorophyll meter/SPAD meter.

The structural characterization of green chilli powder was done by X-Ray Diffraction, microscopic characterization by Scanning Electron Microscopy and elemental study by EDAX. For the structural and microscopic characterization, fresh green chilli was harvested, dried (Tray during at 50^0 C in a Dehydrator) and ground to make a fine powder using a mixer-grinder and mortar-pestle. All samples of green chilli powders were examined by Scanning Electron Microscopy (SEM) (CARL ZEISS, MA 15/ EV 018).

X-Ray Diffraction (XRD) characteristics of green chilli powders were obtained by using an X-ray diffractometer (PAN analytical, X'PERT PRO), with Cu K α_1 radiation of wavelength 1.5405980Å. In a wide range of 2θ (10-90) at a scanning rate of 6 degrees per min. The powder samples (180–250 µg) were placed and slightly pressed in an aluminium holder using a glass slide. The diffractometers were working at the input energy-adjusted at 35 kV and 30 mA.

When the atom in a material is ionized using high energy radiations, they emit out characteristic X-rays. The number of counts in each peak is converted into elemental weight concentration either by comparison with standards or by a simple calculation. With modern detectors, electronics most E-DAX systems can detect X-rays from all the elements in the periodic table above Beryllium (atomic no, Z=4), if present in subsequent quantity.

RESULTS AND DISCUSSION

The plant height and number of branches are beneficial traits for any plant as they correspond to the higher number of fruits per plant ultimately increasing the yield of the plant. The highest plant height was observed in Srinagar Local cultivar (102.13 cm) while it was the lowest in Kashi Anmol (66.13). Maximum primary branches were observed in Taiwan-1 (8.80) and minimum in IC-119290 (6.33) (**Table 1**). Similar studies were reported by Sreelathakumary and Rajamony (2004), Manju and Sreelathakumary (2002) Srinivas *et al.* (2017) and Kerketta *et al.* (2018).

Stem girth was found maximum in IC-119308 (1.10 cm) and minimum in Taiwan-1 (0.76 cm). Taiwan-1 was recorded to have maximum leaf area (52.60 cm^2) while that of IC-119290 (30.95cm^2) was recorded with minimum leaf area. The early appearance of flowers determines the time of fruiting which is very crucial for the growers as early flowering and fruiting can fetch a higher market price. Kashi Anmol took the least days for the appearance of the first flower (32.13) while Taiwan-1 (35) took most days for it. Fruit length is also an important plant trait as the higher is the fruit length higher will be the yield of the plant. The fruit length was found maximum in IC-119290 (6.40 cm) and minimum in Taiwan-1 (4.37cm) (**Table 1**). The variations in chilli fruit length were also reported by Sreelathakumary and Rajamony (2004), Chattopadhyay *et. al.* (2011) and Srinivas *et. al.* (2017).

The higher the number of fruits per plant higher will be its yield hence this makes this particular trait very important from the grower's point of view. The highest number of fruits per plant was obtained in Kashi Anmol (391.07) while it was the lowest in IC-119290 (205.93). Lesser days to fruit set leads to early market availability of the harvested produce and hence becomes crucial in gaining better market price. Cultivar Kashi Anmol took the least days to the first fruit set (35.67) while IC-119290 took maximum days for the first fruit set (40.53). Maximum, seeds per fruit were found in IC-119290 (62.13) while it was least in Taiwan-1 (51.07) as shown in **Table 1**. Similar variations were also reported by Chattopadhyay *et al.* (2011), Srinivas *et al.* (2017) and Kerketta *et al.* (2018).

Table 1. Mean performance of chilli genotypes for growth, leaf, flower, fruit, seed, yield and quality traits

Genotypes	Plant height (cm)	Number of primary branches per plant	Stem girth (cm)	Leaf area (cm^2)	Days to first flowering	Fruit length (cm)	Number of fruits per plant	Days to first fruit Set	Number of seeds per fruit	Yield per plant (g)	Moisture %	T.S.S (° Brix)	Ascorbic Acid (mg/100g)	Chloro-phyll content (SPAD)
Kashi Anmol	66.13	8.20	0.84	34.03	32.13	5.49	391.07	35.67	54.82	725.07	89.10	6.74	132.53	62.80
Taiwan-1	78.13	8.80	0.76	52.60	35.00	4.75	264.20	38.53	51.07	611.28	86.07	4.94	109.36	61.50
Srinagar Local	102.13	7.27	0.87	43.40	32.67	5.35	357.73	36.00	57.21	770.03	83.57	5.57	107.08	52.43
IC-119308	90.87	8.27	1.10	52.57	34.27	6.37	277.60	38.40	56.89	733.91	83.30	5.95	85.30	62.25
IC-119290	63.93	6.33	0.87	30.95	37.07	6.40	205.93	40.53	62.13	360.29	79.51	5.71	94.53	59.33

The ultimate aim of a farmer is to obtain maximum yield from the cultivated crops and therefore yield become the most important parameter while selecting and differentiating a particular genotype from others. The highest yield per plant was recorded in Srinagar Local (770.03 g) while it was the lowest in IC-119290 (360.29 g). The highest moisture percentage was recorded in Kashi Anmol (89.10) and the lowest in IC-119290 (79.51). Total soluble solids and ascorbic acid content is an important quality parameter because it determines the quality of the fruits of chilli. Cultivar Kashi Anmol had fruits with the highest total soluble solids (6.74 ^{0}Brix) while the lowest was recorded in fruits of variety Taiwan-1 (4.94° Brix). A similar pattern in chilli was also observed by Kumar and Tata (2009). The maximum amount of ascorbic acid was recorded in Kashi Anmol (132.53 mg/100g) and the minimum in IC-119308 (85.30 mg/100g). The higher chlorophyll content is desirable as it directly influences the yield due to the accumulation of photosynthates. The highest chlorophyll content was shown by the variety Kashi Anmol (62.80 SPAD) and that of Srinagar Local (52.43 SPAD) (**Table 1**). The results were in proximity to the findings of Manju and Sreelathakumary (2002), Kumar *et al.* (2003) and Kerketta *et al.* (2018).

The structural properties of the five green chilli powders were studied by XRD (**Fig. 1, 2, 3, 4 and 5)**. The typical diffraction patterns of green chilli powders showed higher peaks (ranged between 16.674-21.145) at a 2θ value. It was observed that the curve of all green chilli powders was almost similar and having no major difference.

Powders obtained from Taiwan-1, IC-119308 and IC-119290 showed a high peak at 2θ value of about 21.145° that suggests a crystalline structure, a moderate peak through chilli powder of Taiwan-1 and a small peak at a 2θ value were found on a sample obtained from Srinagar Local (16.674°). These results indicated that there is no difference in the backbone structures of the green chilli powders. The present result was in close conformity with the findings of Alsebaeai *et al.* (2017) and Alsebaeai *et al.* (2018).

The surface of powder particles from green chilli cultivars was analyzed in a three-dimensional way by scanning electronic microscopy. The photomicrographs are shown in **fig. 6, 7, 8, 9 and 10**. The typical pulverized particles showed that dried green chillis were crushed into very small particles the edges of the particles were round while it was destroyed in the amorphous region. The morphology of the powder obtained from all the studied cultivars was found to be similar. These observations are synchronous with the earlier observations recorded by Zhu and Zhao (2014), Alsebaeai *et al.* (2017) and Alsebaeai *et al.* (2018).

E-DAX Analysis can be very helpful in tracing the presence of any harmful element in the studied sample showed the presence of various elements in all chilli powders such as Carbon, Oxygen, Chlorine, Potassium, Magnesium and Aluminium which is presented in the form of graphs in **fig. 11, 12, 13, 14, 15 and 16.** The concentration of Carbon varied from 58.45 (Taiwan-1) to 72.63 per cent (IC-119290), while the concentration of oxygen was found minimum in IC-119290 (26.25%) and maximum in Taiwan-1 (39.47%). The amount of Chlorine ranged from 0.27 (Srinagar Local) to 0.15 per cent (Kashi Anmol), while the amount of potassium was maximum in Taiwan-1 (1.64%) and minimum in IC-119290 (0.49%). Element Magnesium was found in only two varieties Taiwan-1 (0.26%) and Srinagar Local (0.22%), while Aluminium was also found only in two varieties Srinagar Local (0.22%) and IC-119290 (0.15%) There was an absence of any health hazardous element in the powders. A similar study was also conducted by Goutam *et al.* (2017) in coriander and found similar results as there was no adulteration in the Zinc oxide nanoparticles.

For, the quality, yield and its attributing traits, Kashi Anmol was found superior among all the cultivars. The diffraction pattern obtained from different chilli cultivars ranged between 16.674-21.145°. It was observed that the curve obtained from chilli powder of Srinagar local cultivar (16.674°) was different from other cultivars. The scanning electron micrographs showed that the surface morphology of all chilli powders obtained from different cultivars was almost similar. Results of E-DAX analysis showed the presence of various elements (C, O, Cl, K, Mg, Al) in

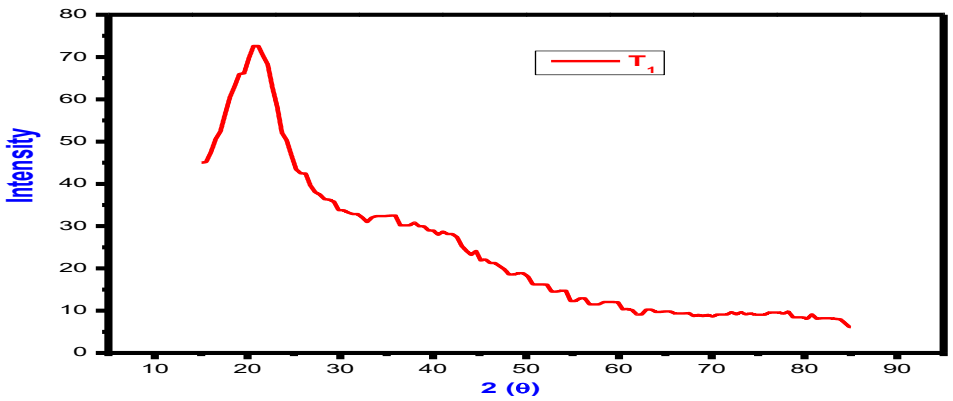


Fig. 1. XRD Pattern obtained from green chilli powder of variety Kashi Anmol

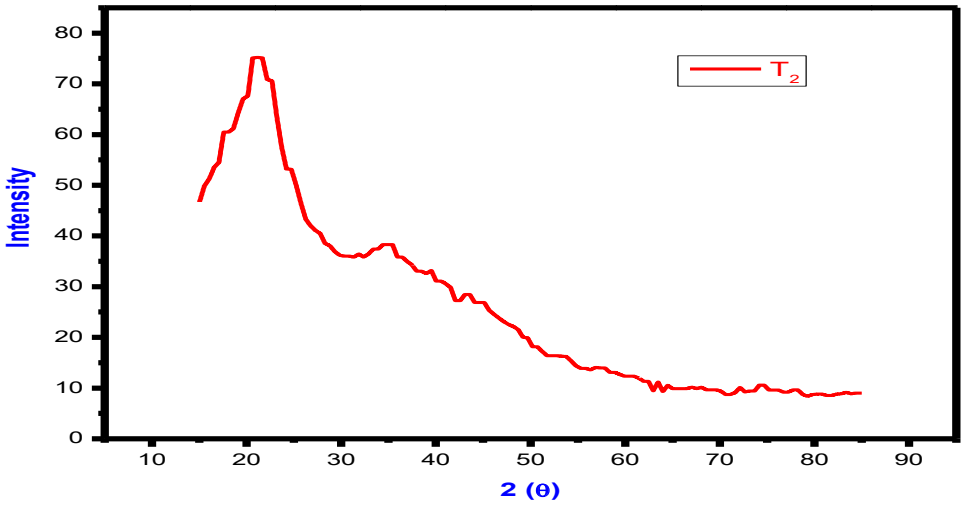


Fig. 2. XRD Pattern obtained from green chilli powder of variety Taiwan-1

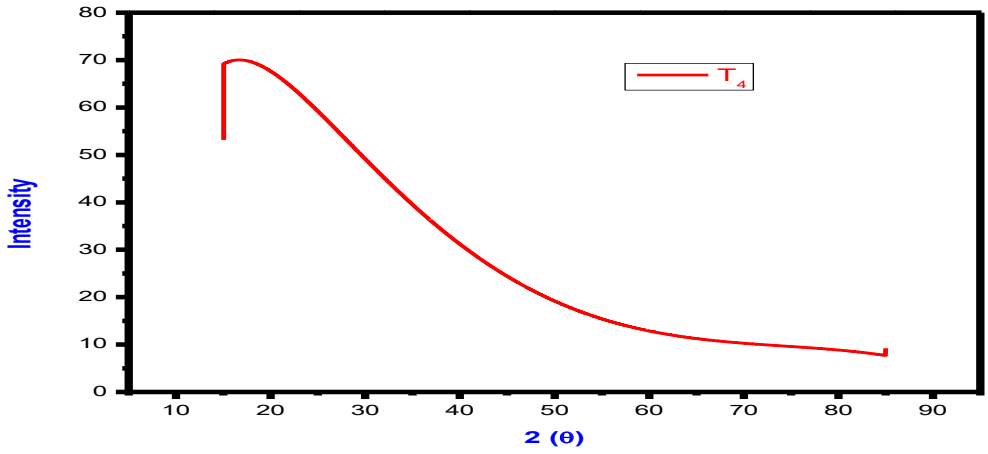


Fig. 3. XRD Pattern obtained from green chilli powder of variety Srinagar Local

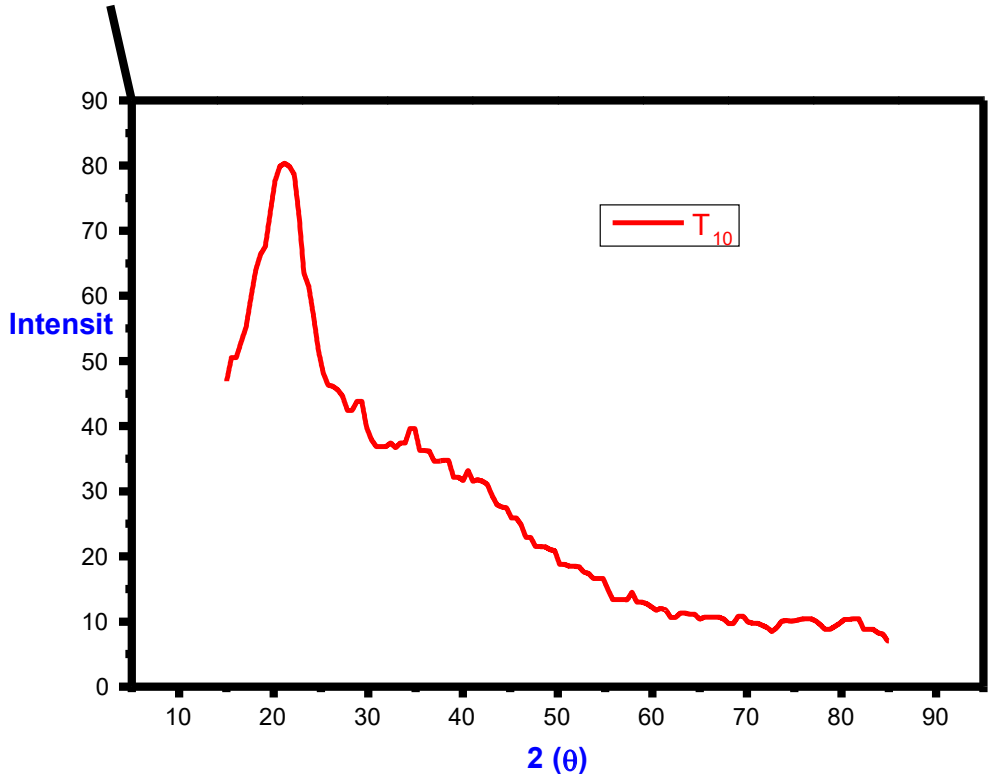


Fig. 4. XRD Pattern obtained from green chilli powder of variety IC- 119308

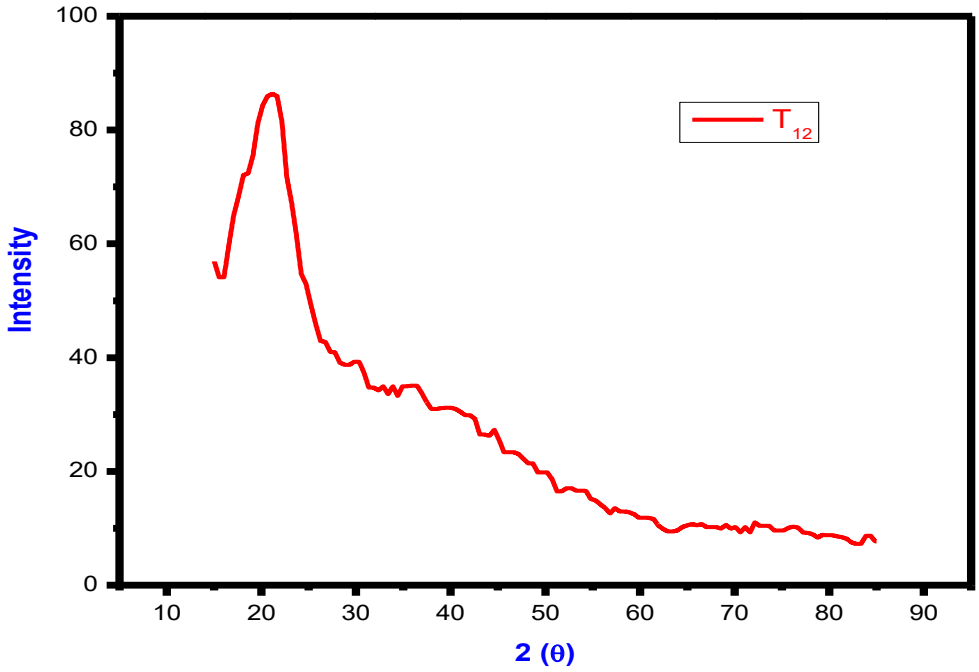


Fig. 5. XRD Pattern obtained from green chilli powder of variety IC- 119290

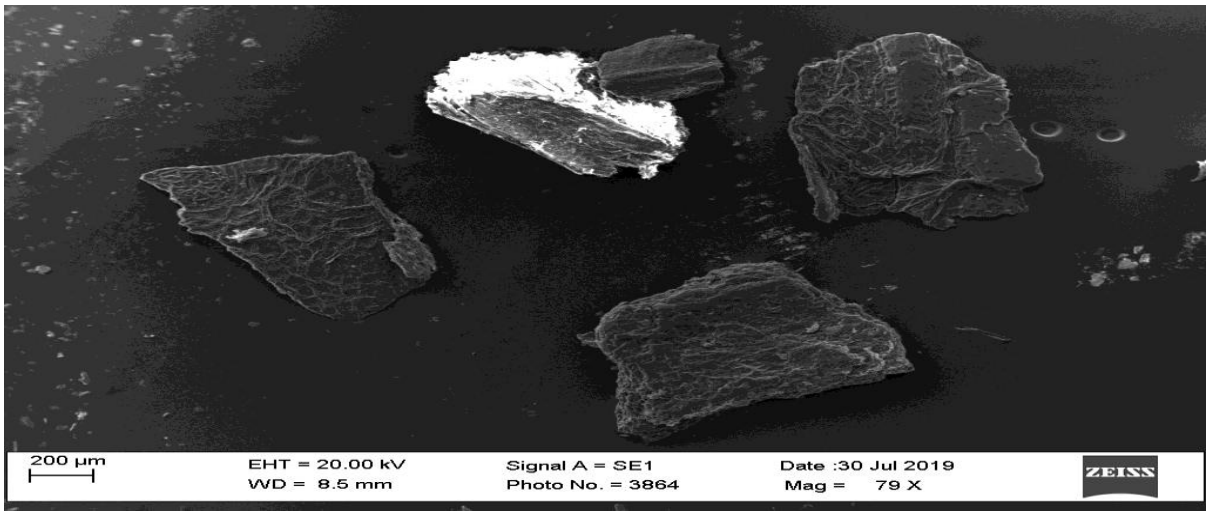


Fig. 6. Scanning electron micrograph obtained from green chilli powder of variety Kashi Anmol

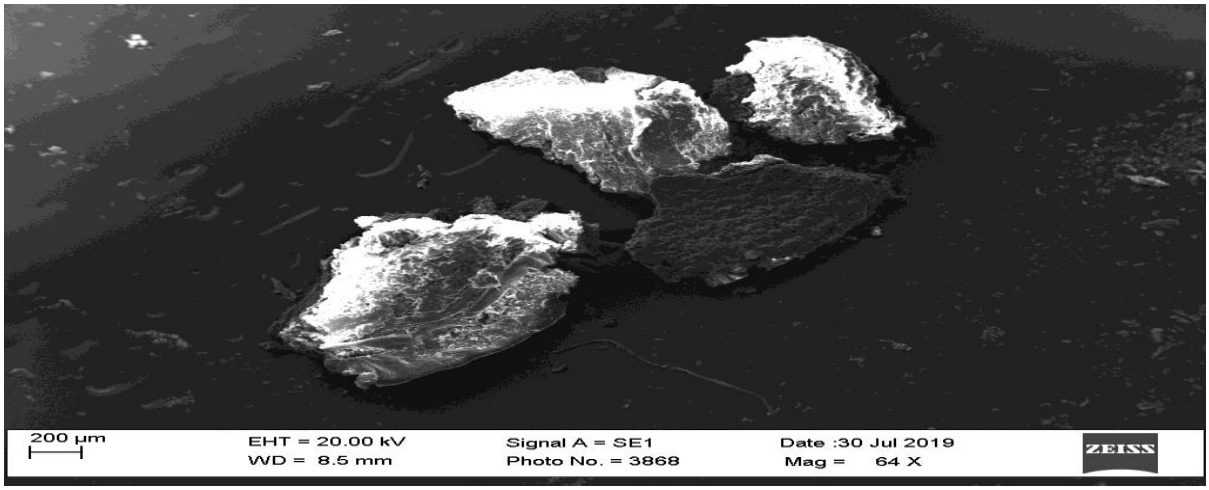


Fig. 7. Scanning electron micrograph obtained from green chilli powder of variety Taiwan-1

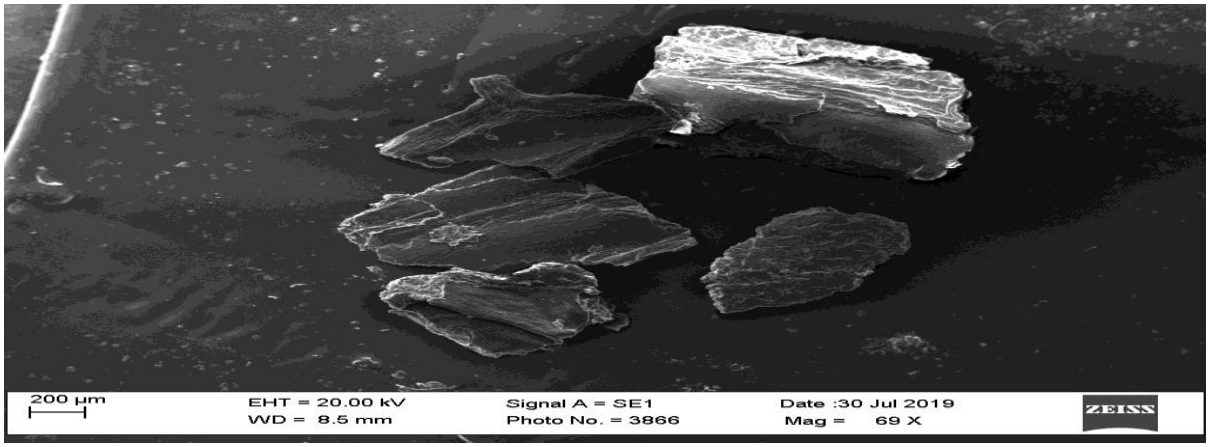


Fig. 8. Scanning electron micrograph obtained from green chilli powder of variety Srinagar Local

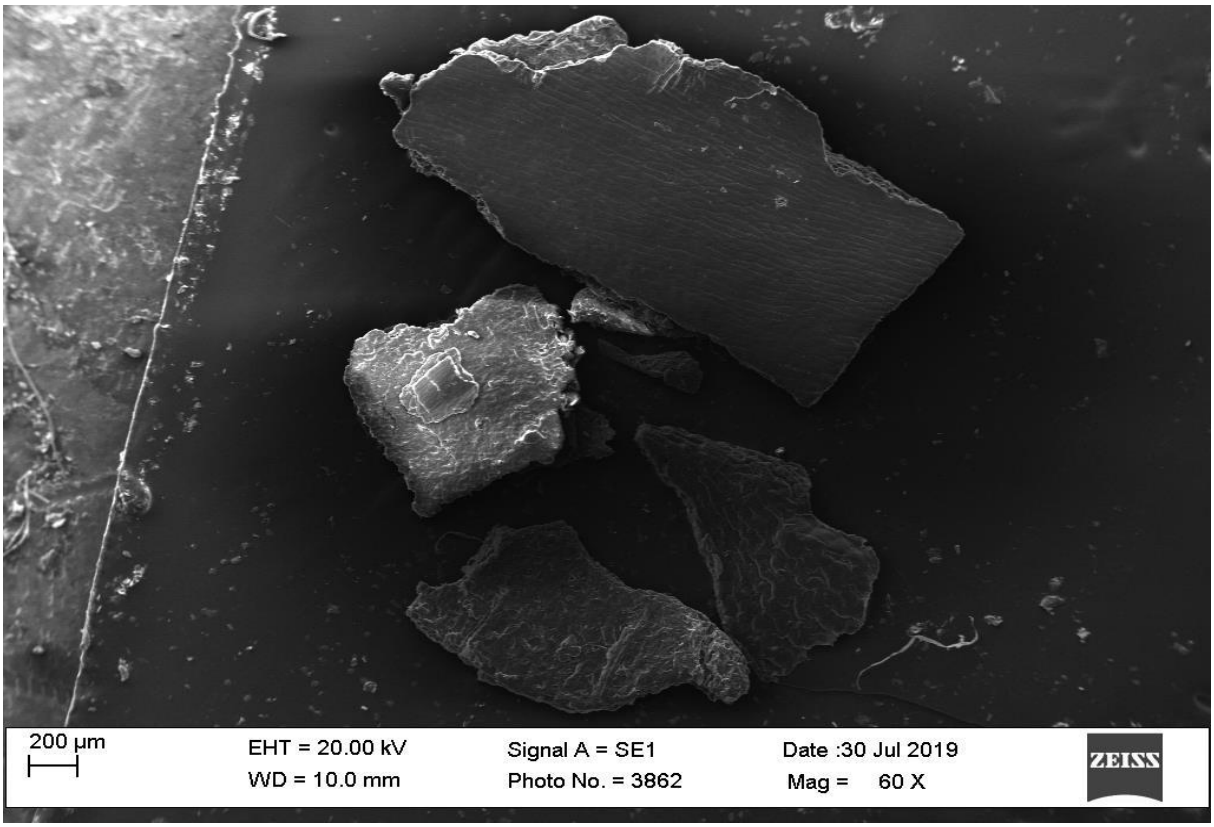


Fig. 9. Scanning electron micrograph obtained from green chilli powder of variety IC- 119308

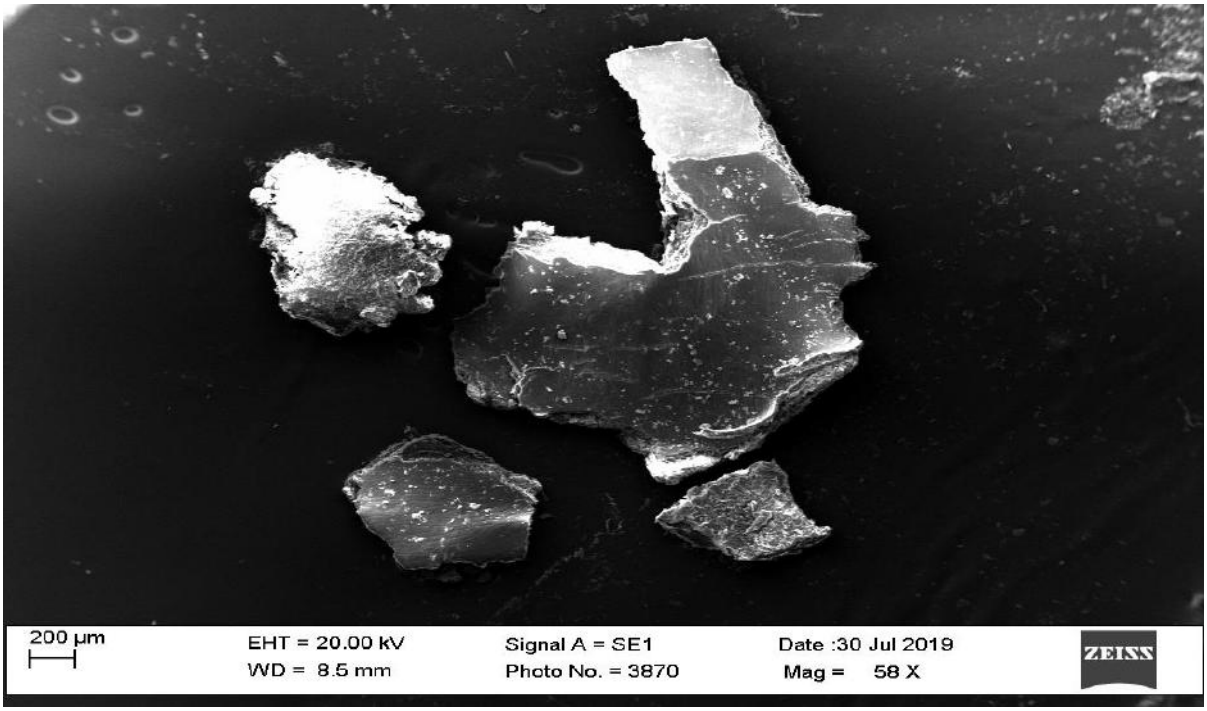


Fig. 10. Scanning electron micrograph obtained from green chilli powder of variety IC- 119290

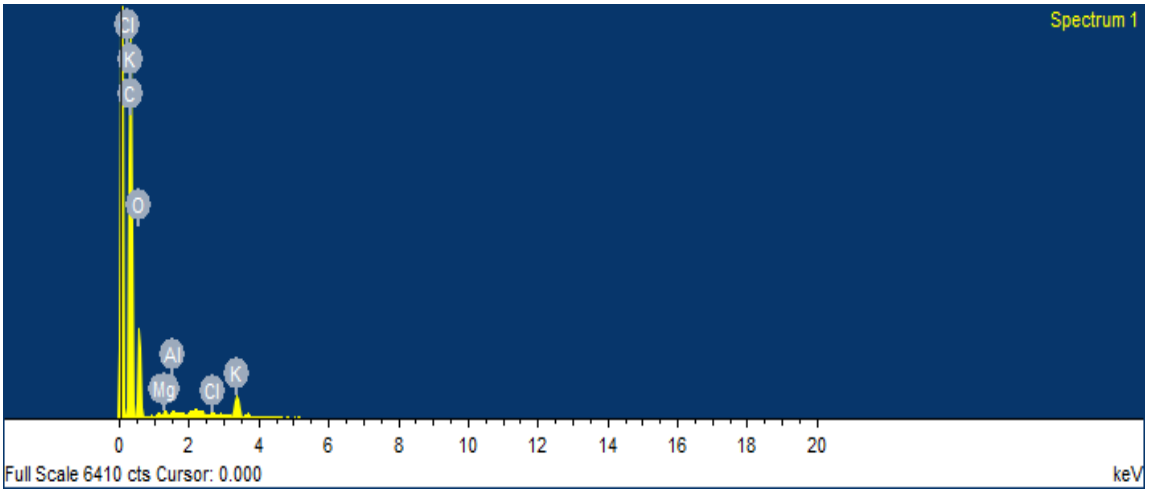


Fig. 11. E-DAX data obtained from green chilli powder of variety Kashi Anmol

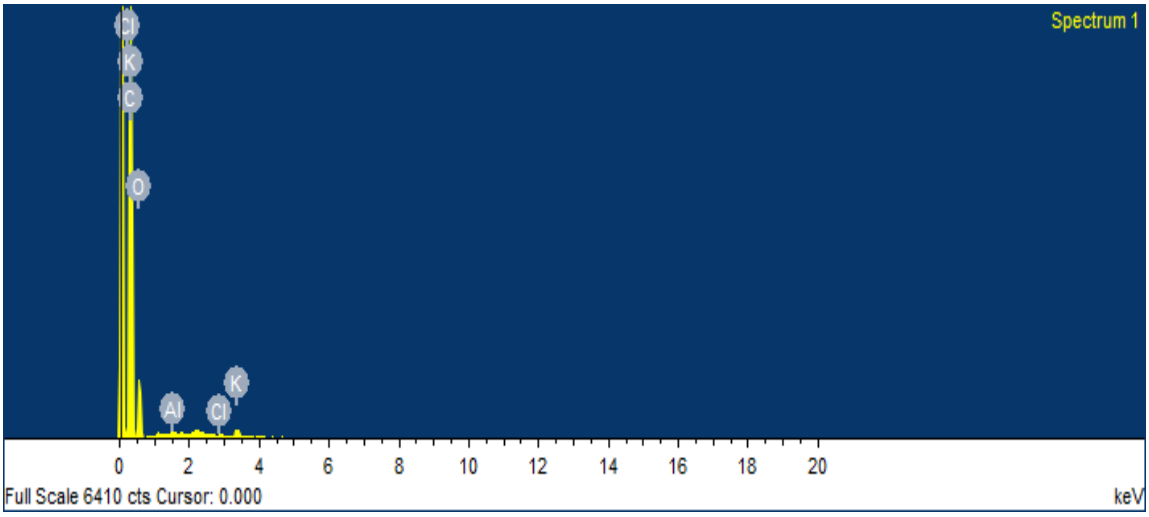


Fig. 12. E-DAX data obtained from green chilli powder of variety Taiwan-1

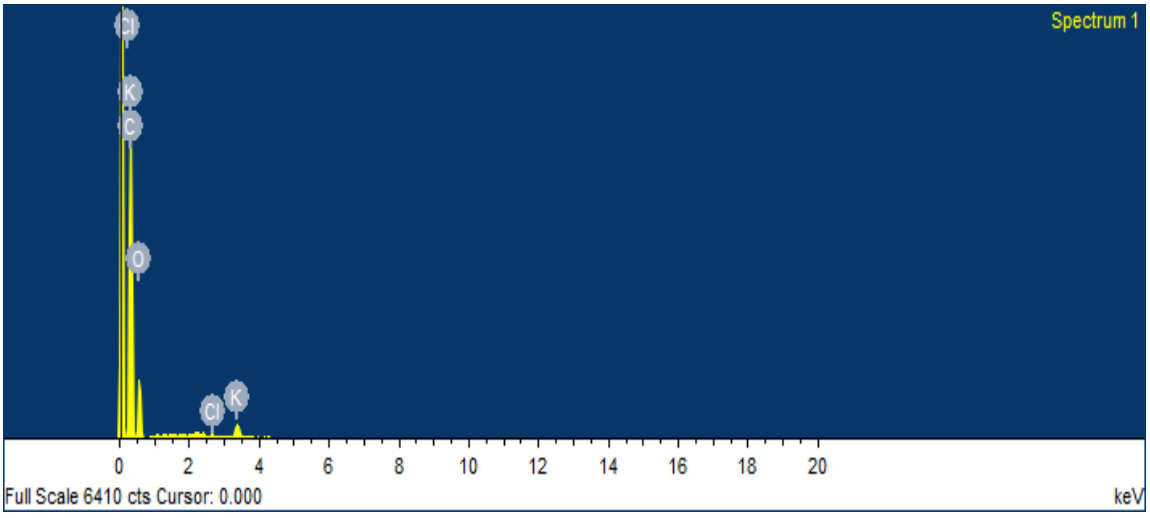


Fig. 13. E-DAX data obtained from green chilli powder of variety Srinagar Local

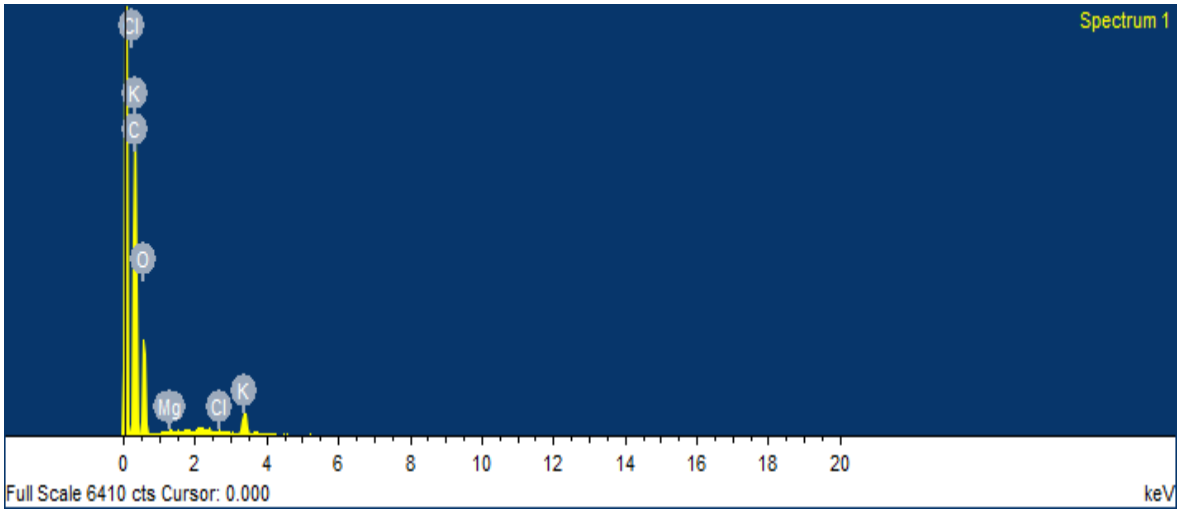


Fig. 14. E-DAX data obtained from green chilli powder of variety IC-119308

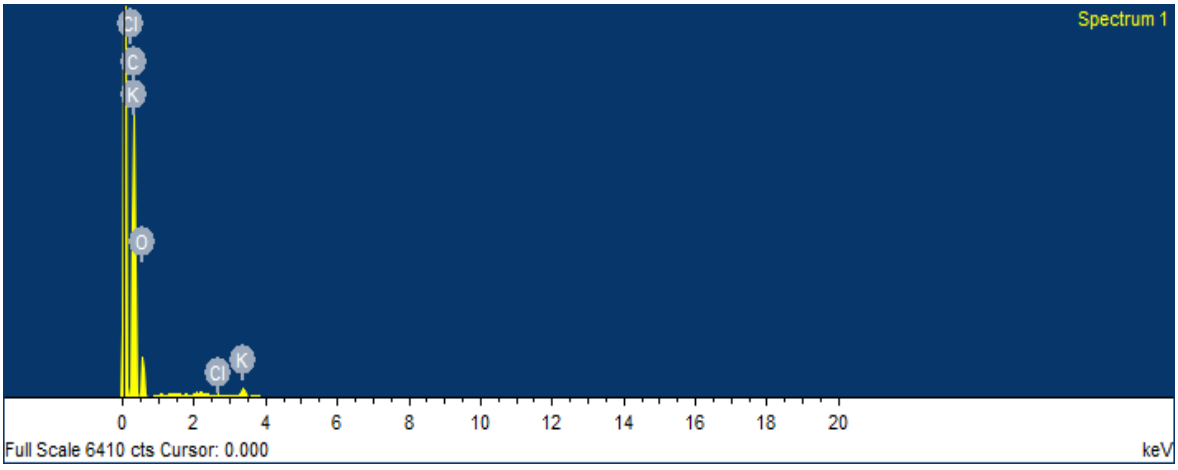
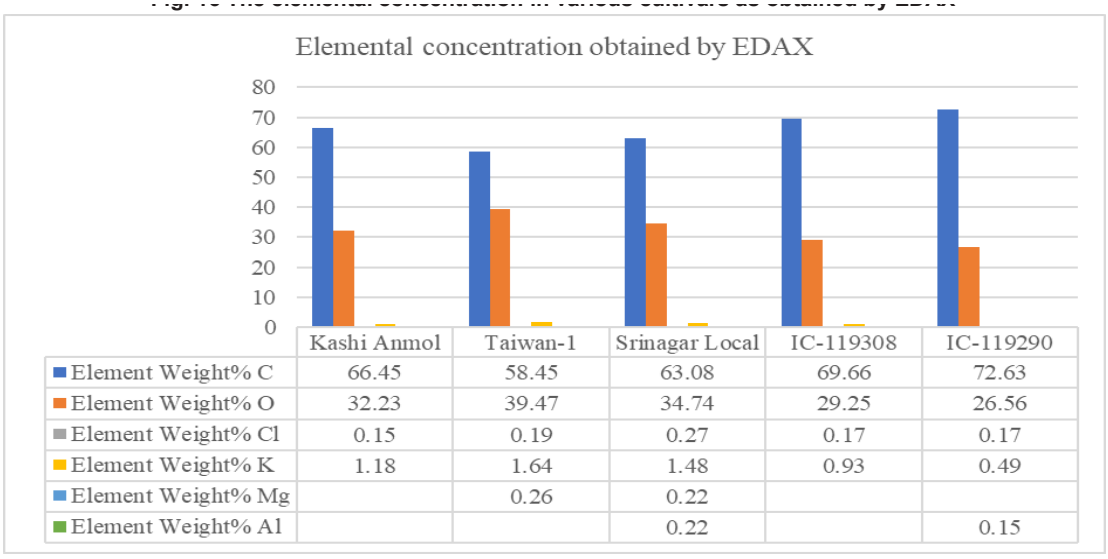


Fig. 15. E-DAX data obtained from green chilli powder of variety IC-119290



	Kashi Anmol	Taiwan-1	Srinagar Local	IC-119308	IC-119290
Element Weight% C	66.45	58.45	63.08	69.66	72.63
Element Weight% O	32.23	39.47	34.74	29.25	26.56
Element Weight% Cl	0.15	0.19	0.27	0.17	0.17
Element Weight% K	1.18	1.64	1.48	0.93	0.49
Element Weight% Mg		0.26	0.22		
Element Weight% Al			0.22		0.15

Fig. 16 The elemental concentration in various cultivars as obtained by EDAX

the studied chilli cultivars in different concentrations. IC-119290 showed the highest amount of carbon percentage (72.63%) which along with the presence of low moisture content can be correlated to the high dry weight of this cultivar. Any health-hazardous elements was absent. This study suggests the cultivation of Kashi Anmol when looking for commercialization of chilli in the study area, though the local variety comes very close to it in terms of yield the presence of quality traits like total soluble solids, ascorbic acid and chlorophyll content in very low amount in the local variety made us emphasize on Kashi Anmol. The present study can be further utilized in the evaluation of other cultivars and comparing them with the present best cultivar (Kashi Anmol) in the study area as well as in different parts of the country. The study can also be utilized for the detection of harmful elements resulting due to pesticidal residues in comparing cultivars grown under chemical methods and in an organic set of cultivation practices.

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