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Biochemical and molecular characterization of brinjal varieties and promising genotypes of Saurastra region

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Abstract
Brinjal (*Solanum melongena* L.) fruit is known for vegetables of diet food having a high moisture content, low caloritic value, good source of antioxidants as well as some phytonutrients. The present experiment was conducted with an objective to examine molecular diversity among brinjal genotypes and varieties associated with quality characteristics and fruit proximate composition of brinjal fruits. The three year pooled data that the most diverse varieties were found GOB-1 and JBGR-1 compared to the other promising genotypes and varieties based biochemical, nutritional analysis. The clustering pattern on the basis of molecular analysis (SSR) depicting diverse varieties GOB-1 and GJB-3 out grouped from other genotypes with 48% similarity. The diverse GOB-1 contained higher protein, total soluble solids, soluble sugars, phenols, ascorbic acid, PPO activity and flavanoid content and lower in glycoalkaloids and acidity.

Keywords: Antioxidents, Brinjal, Eggplant, Flavanoid, Glycoalkaloid, Molecular Diversity, Phenol, *Solanum melongena* L., SSR

Introduction
Eggplant or brinjal (*Solanum melongena* L.) fruit is known for vegetables of diet food because of high moisture content and low caloritic value. However, it is a good source of antioxidants as well as some phytonutrients (Kandoliya *et al.*, 2015) [4]. The color, size, shape of the eggplant fruit vary significantly with the type of cultivar. Fruits and are ranked amongst the top ten vegetables in terms of antioxidant capacity due to the fruit phenols and flavonoic constituents (Timberlake, 1981; Singh *et al.*, 2009) [30, 27], which have been linked to various health benefits (Ames *et al.*, 1993; Hung *et al.*, 2004) [1, 11]. Eggplant fruits have shown high hydrophilic oxygen radical absorbance capacity (Cao *et al.*, 1996) [5], which has been correlated to phenols compounds presence, including delphinidin as a major component in peel (Wu *et al.*, 2006; Koponen *et al.*, 2007) [34, 18] and chlorogenic acid in flesh (Winter and Hermann, 1986; Whitaker and Stommel, 2003) [33, 32]. Extracts from eggplant are effective for curing a number of diseases, including cancer, high blood pressure, and hepatosis due to content of anthocyanins and strychnine (Magioli and Mansur, 2005; Silva *et al.*, 1999) [21, 26]. Thus, the experiment was conducted with an objective to examine fruit proximate composition, quality characteristic of brinjal and molecular diversity among brinjal genotypes and varieties associated with quality characteristics of brinjal fruits.

Materials and Methods
The experiment was conducted for three years for biochemical parameter in three replications. The fruits of marketable size were collected from each replication were used for biochemical analysis whereas young tender leaves were used for molecular parameters. The mean data of three years were presented under appropriate headings.

Source of Materials
Fruits of brinjal varieties and genotypes used in present experiment were obtained from Vegetable research Centre, Junagadh Agricultural University, Junagadh, for analysis of different parameters as under.

Biochemical Analysis
Nutritional components: Moisture was determined by oven drying at 105°C for 8 hours AOAC (2005) [3]. The amount of total soluble sugar and true protein was estimated by Anthrone reagent (Hedge & Hofreiter, 1962) [14] and Folin-Phenol reagent (Lowry *et al.* 1951) [19]

respectively. Total soluble solids (TSS) were determined using refractometer and expressed as °Brix. Total acidity was determined by titration with a standard solution of NaOH as described by Rangana (1977) [29]. The glycoalkaloid was extracted from appropriate amount of fruit pulp in chloroform: acetic acid: methanol mixture (50:5:45), estimated as per Currier and Kuc (1975) [8] and OD value obtained was directly used for comparison.

Enzyme Activity

Polyphenol oxidase (PPO, EC 1.14.18.1): Appropriate amount of fruit pulp tissue were ground in 5 ml of 100mM sodium phosphate buffer, pH 6.5 The homogenate was centrifuged at 10,000 rpm for 15 min at 4°C and the supernatant was used for enzyme assay. The reaction mixture contained 2.9 ml of catechol (10mM catechol in 10 mM phosphate buffer pH 6.5) and reaction was initiated by the addition of 100 l of enzyme extract. The changes in the colour due to the oxidized catechol were read at 490 nm for one minute at an interval of 15 second. Blank was carried out without substrate. The enzyme activity was expressed as change in OD. min.$^{-1}$ g.$^{-1}$ Fr.Wt. tissues (Malik and Singh, 1980) [20].

Phenylalanine ammonia lyase (EC **4.3.1.24):** The PAL activity was analyzed using three hundred milligram of fruit pulp tissues homogenized with a pre-chilled mortar and pestle in 3 ml of extraction buffer containing 50 mM borate-HCl buffer (pH 8.5) and 0.04% ß-mercaptoethanol. The homogenate was centrifuged at 10,000 rpm for 15 min. The clear supernatant was used as the enzyme source for the assay of PAL (Cheng and Breen, 1991).

Antioxidant related components: The phenol content in was determined by method of Malik and Singh (1980) [20] using methanolic extract. Total Ascorbic acid was quantified according to the colorimetric method described by Omaye *et al.*, (1979). Total flavonoid was estimated as described by the Chanda and Dave (2009) [6]. Total anthocyanins were analyzed by differential pH method (Cheng and Breen, 1991) [7] and expressed as OD per g fresh weight.

Molecular Parameters

DNA extraction fifteen brinjal genotypes constituted the experimental material. The DNA was isolated from the leaves. It was found that DNA isolated by modified Dellaporta method was of high purity and the yield was also substantial (Dellaporta *et al.*, 1983) [9]. Hence the DNA isolated for all the samples by this method was used for further analysis.

Polymerase chain reaction (PCR) Purified DNA was subjected to amplification for SSR in a 25 µl reaction mixture containing 50 ng DNA, 1 U Taq DNA polymerase (MBI Fermentas), 1x Taq buffer (MBI Fermentas), 3 mM $MgCl_2$, 200 µM dNTP, and 0.4 µM of each primer (forward and reverse). Amplification was carried out in 96 well thermocycler (Perkin Elmer, Model 9600) programmed for an initial denaturation at 94°C for 4 min., followed by 35 cycles of 94°C for 1 min., 65°C for 1 min., 72 °C for 2 min. and finally a 5 min., extension at 72°C. The amplified products were resolved on a 3% metaphor agarose gel containing ethidium bromide at a constant voltage of 60 V for 3 h using a horizontal gel electrophoresis system (BioRad). The amplified fragments were visualized and photographed under UV light using a gel documentation system. Polymorphic information content (PIC) as a measure of allele diversity at a locus was determined for each SSR primer pair (Anderson *et al.* 1992). The amplified fragments were scored manually for their presence (denoted as '1') or absence (denoted as '0') for each primer for SSR marker systems. The binary matrix was used to estimate Jaccard's genetic similarity coefficients (Jaccard) for SSR. The similarity matrices were subjected to unweighted pair group method of arithmetic averages (UPGMA; Sneath and Sokal,) clustering method in order to construct the two dendrograms. The NTSYS-PC software, Version 2.1 (Exter Software, Setauket, NY, USA) was employed to carry out these analyses (Rohlf,).

Results and Discussion

a) Biochemical analysis

The three years pooled analysis mean data on various nutritional parameters like protein content, soluble sugar content and antioxidant contributing factors like total phenol, flavonoids, total anthocyanins, ascorbic acid content in fruits of brinjal varieties and molecular markers are given in Table 1 to 7 and Figure 1 & 2. The results are reported as mean values of three replications on fresh weight basis.

Brinjal is known for vegetables of diet food because of high moisture per cent and low calorific value. The mean data of the moisture content in fruits of brinjal varieties and genotypes for three years varied from 90.65 to 91.91% which were not differed significantly during the individual year as well as for the pooled analysis. The variety wise moisture value for individual year also remains inconsistent (Table 1).

The total soluble solid (TSS Brix) data showed statistically significant differences for the pooled data and for the individual year 2014-15 (Table. 1). The mean highest TSS value over the year was observed for the variety GOB-1 (9.33%). The lowest value (8.17%) was observed for the GBL-1, GJB-3, Pant Ruturaj and Surati ravaiya. Dendrogram prepared from SSR primers also showed that theGOB-1 and JBL-1 falls under different group/clusters. The variety with low TSS value i.e. Pant Ruturaj and Surati ravaiya also falls in a same cluster. However, the same was not true for the other varieties.

The data for the total soluble sugar content showed significant differences for the pooled, year 2014-15 and year 2016-17 data. However the data for the year 2015-16 showed nonsignificant results (Table. 2). The highest mean total soluble sugar percent was observed for the variety JBGR-1 (4.49%) which remained at par with GOB-1 (4.36%), JBL-08-8 (4.22%), GBL-1(4.18%) and JBL-12-06-4-1 (4.17%). The high amount of sugar components confers its significant roles to human health because, apart from the supply of energy, they are also needed in numerous biochemical reactions not directly concerned with energy metabolism. The lowest value was observed for variety GJB-3 (3.31%). The variety showed the highest value (i.e. JBGR-1) and the lowest value (i.e. GJB-3) falls under different clusters as analyzed by SSR data (Fig. 2).

The higher phenol content is associated with higher antioxidant capacity as well as higher resistance against disease and pest in various crops (Kandoliya and Vakharia, 2013; Mori *et al.*, 2017; Patel *et al.*, 2015;) [14, 22, 24]. Various studies have also reported a good correlation between the total phenol content of plant extracts and antioxidant activity (Kandolia *et al.* 2015b; Kandolia *et al.* 2016) [16, 17]. In present study, the mean phenol content was varied significantly from 18.49 mg.100g^{-1} in AB-08-14 to 44.86 mg.100g^{-1} in JBGR-06-8 (Table.2).

Protein content of the brinjal fruit is responsible for its nutritional value. The data on protein content showed significant differences for the pooled as well as individual year (Table. 3). The variety GOB-1 showed highest value of total protein in all the three year tested as well as pooled data over the year (1.30%) on fresh weight basis. Whereas the mean lower value was recorded for the genotype AB-08-14 (0.72%). Both the variety falls under the different cluster as per the molecular analysis (Fig. 2).

The ascorbic acid content, the major antioxidant and neutraceutically important compound (Vyas *et al.* 2015; Bajaniya *et al.*, 2015) [31, 4]. Ascorbic acid content was analyzed from different varieties and genotypes of eggplant showed significant variation. Ascorbic acid (mg/100 g) content found lowest value for the individual year cosistantly as well as pooled data over the year was recorded for the genotype AB-08-14 (Table. 3). The mean lowest value for the genotype AB-08-14 was 11.00 mg/100 g fresh pulp weight of brinjal. The highest value was recorded for the genotype AB-07-2-15 (23.97 mg/100 g) which remain at par with GOB-1 (23.80 mg/100 g) shared on cluster as per the dendrogram prepared based on SSR data. AB-08-14 falls in a diverse group as compared to above variety.

A high anthocyanin content and a low glycoalkaloid content are considered essential, regardless of how the fruit is to be used. Bitterness in eggplant is due to the presence of glycoalkaloids which are of wide occurrence in plants of Solanaceae family. The Glycoalkaloid content found significant variation over the year among the variety. The lowest value was recorded in AB-07-2-15 (0.27 $OD.g^{-1}$) which remained statistically at par with JBGR-1 (0.27 $OD.g^{-1}$). The mean highest value (0.53 $OD.g^{-1}$) was recorded for the genotype JBL 12-06-4-1 and the same was found highest (0.62 $OD.g^{-1}$) for the year 2015-16. The Anthocyanin varied significantly in the brinjal fruits with the highest value 2.53 $OD.g^{-1}$ in the JB-10-208 while, JBGR-1 exhibited minimum content i.e. 0.74 $OD.g^{-1}$ (Table.4).

Browning reaction is important for fruit quality of brinjal. Polyphenol oxidase (PPO) is the enzyme responsible for the same. The enzyme catalyses the *o*-hydroxylation of monophenols (phenol molecules in which the benzene ring contains a single hydroxyl substituent) to *o*-diphenols (phenol molecules containing two hydroxyl substituents). They can also further catalyzed the oxidation of *o*-diphenols to produce *o*-quinines. It is the rapid polymerization of *o*-quinones to produce black, brown or red pigments (polyphenols) that is the cause of fruit browning. The PPO activity for brinjal fruits of different variety and genotype studied and found significant variation from 1.07 min/gm fresh weight in JBGR-06-8 to 1.84 OD. min/gm fresh weight in JBGR-1. The phynylalanine ammonia lyase (PAL) activity varied significantly in the brinjal fruits with maximum 1.35 OD. min/gm. fresh weight in JBL-08-8 & JBGR-06-08 and minimum 0.91 OD. min/gm. fresh weight in JB-10-208 (Table 5). The flavanoid content also varied significantly in the brinjal fruits and was recorded minimum (6.85 mg/100 g) in pant ruturaj which remais at par with JBL-08-8 (6.83 mg/100g). The highest value was measured 15.16 mg/100g in JBGR-1 (Table. 6).

The significant differences were observed for the titratable acidity and the genotype JBGR-06-8 contained maximum value while, the minimum with the brinjal variety Swarna Mani (Table 6) and both were fall in different sub-cluster as per the dendrogarm prepared from theSSR analysis (Fig. 2).

b) Molecular analysis

Total 47 SSR primers were used to amplify genomic DNA of 15 brinjal genotypes (Table 7). The 47 SSR markers produced total 165 alleles of which 158 were polymorphic with an average of 3.36 alleles per primer and 7 alleles were monomorphic. Among the 158 polymorphic alleles, 107 alleles were shared polymorphic within two or more genotypes, while 51 alleles were unique-polymorphic. The SSR markers EEMS07 and Xgdm-62-3D produced maximum number of 10 alleles, while EEMS14, EEMS17, EEMS23, EEMS29, EEMS39 and Xgdm-93-4B produced minimum number of 1 allele. The percent polymorphism obtained for SSR primers were ranged from 0.00 to 100% with an average value of 92.6% per primer. The Polymorphism Information Content (PIC) values for SSR marker were ranged from 0.000 (EEMS14, EEMS23, EEMS29 and Xgdm-93-4B) to 0.94 (EEMS49) with an average value of 0.46 per primer and SSR primer index (SPI) differed from 0.00 (EEMS14, EEMS23, EEMS29 and Xgdm-93-4B) to 14.27 (EEMS07).

Similarity index and cluster analysis of 15 brinjal genotypes were carried out by Jaccard's similarity coefficient and UPGMA using NTSYSpc-2.02i software, respectively. The similarity coefficient ranged from 25 to 74% (Fig. 2). The fifteen brinjal genotypes were grouped into two main clusters: cluster A and cluster B shared 25% similarity. The cluster A comprised of two clusters, cluster A1 and cluster A2. Cluster A1 consisted of GOB-1 and GJB-3 genotypes and shared 48% similarity. While, cluster-A2 comprised of single genotype AB-07-2-15. The cluster B consisted of two clusters, cluster B1 and cluster B2 with minimum similarity 49%. The subcluster B1 grouped out maximum genotypes with 53% similarity and while subcluster B2 comprised of AB-08-14 and Doly-5 having 53% similarity.

From using all the biochemical data, denrogram prepared by online software (http://genomes.urv.cat/UPGMA/UPGMAboot_v12.cgi) was depicted in Fig. 1.

As per the dendrogram, the varieties divided in to two different main group. The most diverse genotype each other on the basis of biochemical-nutritional parameters were GOB-1 with AB-08-14. The variety GOB-1 and AB-08-14 also found in two different diverse group as dendrogram prepared from the molecular analysis done using SSR primers. The diverse GOB-01 recorded highest protein (1.30%), highest TSS (9.33 brix%), higher total soluble sugar (4.36%), higher total phenol (33.48 mg/100g), higher Ascorbic acid (23.80 mg/100 g) as well as higher PPO activity, flavanoid content, lower glycoalkaloid (0.32 $OD.g^{-1}$) and lower acidity (0.20%). Whereas, genotype AB-08-14 found most away from GOB-1 and showed comparatively inferior quality having lower protein (0.72%), lower TSS (8.33 brix%), lower total soluble sugar (3.47%), lowest total phenol (18.49 mg/100g), lowest ascorbic acid (11.00 mg/100 g) as well as lower PPO activity.

Table 1: Per cent moisture and TSS % (Brix) content of brinjal fruits from different varieties and genotypes

Sr. No.	Name of Variety/ Genotype	Moisture %			Pooled over the year	TSS % (Brix)			Pooled over the year
		2014-15	2015-16	2016-17		2014-15	2015-16	2016-17	
1	AB-08-14	92.18	91.98	90.95	91.70	8.0	8.5	8.50	8.33
2	AB-07-2-15	91.90	92.04	91.78	91.91	9.0	8.0	8.00	8.33
3	JB-10-208	90.98	91.12	90.78	90.96	9.0	8.5	8.50	8.67
4	JBGR-06-8	91.43	91.62	90.41	91.15	9.0	9.0	8.50	8.83
5	JBL-08-8	91.33	90.87	91.25	91.15	9.0	8.5	8.00	8.50
6	JBL 12-06-4-1	91.69	92.11	90.95	91.58	8.5	8.0	8.50	8.33
7	JBGR-1	91.62	91.96	91.12	91.57	8.5	8.5	8.00	8.33
8	GOB-1	90.03	91.12	90.79	90.65	10.0	9.5	8.50	9.33
9	Swarna Mani	91.47	91.98	90.99	91.48	9.0	8.5	8.50	8.67
10	GBL-1	91.87	92.67	90.89	91.81	8.5	8.0	8.00	8.17
11	GJB-2	91.55	91.06	91.00	91.20	9.0	8.5	8.50	8.67
12	GJB-3	92.77	91.99	90.83	91.86	8.0	8.5	8.00	8.17
13	Pant Ruturaj	92.50	92.15	90.95	91.87	8.0	8.0	8.50	8.17
14	Surati Ravaiya	92.10	91.78	91.61	91.83	8.5	8.5	7.50	8.17
15	Doly-5	92.50	91.63	90.36	91.50	9.0	8.5	9.00	8.83
	S.Em+	1.27	1.40	0.61	0.66	0.27	0.30	0.31	0.17
	CD. @ 5%	NS	NS	NS	NS	0.79	NS	NS	0.48
	CV%	2.39	2.64	1.16	2.17	5.42	6.10	6.48	6.00
	Year								
	S.Em+				0.30				0.08
	CD. @ 5%				NS				NS
	Y x T								
	S.Em+				1.15				0.29
	CD. @ 5%				NS				NS

Table 2: Total Soluble Sugar and Total Phenol content of brinjal fruits from different varieties and genotypes

Sr. No.	Name of Variety/ Genotype	Total Soluble Sugar %			Pooled over the year	Phenol mg/100g			Pooled over the year
		2014-15	2015-16	2016-17		2014-15	2015-16	2016-17	
1	AB-08-14	3.75	3.71	2.95	3.47	17.35	15.48	22.64	18.49
2	AB-07-2-15	4.01	4.13	3.13	3.76	25.65	24.36	21.32	23.78
3	JB-10-208	4.24	4.05	3.95	4.08	18.75	17.39	23.57	19.90
4	JBGR-06-8	4.23	4.32	3.38	3.98	50.36	47.33	36.89	44.86
5	JBL-08-8	4.32	4.39	3.96	4.22	38.42	42.36	42.35	41.04
6	JBL 12-06-4-1	4.46	4.41	3.64	4.17	27.62	25.08	33.12	28.61
7	JBGR-1	4.96	4.76	3.74	4.49	39.42	42.56	38.17	40.05
8	GOB-1	4.89	4.71	3.48	4.36	36.47	34.33	29.65	33.48
9	Swarna Mani	4.29	4.11	3.19	3.86	31.42	29.16	30.17	30.25
10	GBL-1	4.44	4.23	3.87	4.18	40.65	43.22	36.28	40.05
11	GJB-2	4.10	4.06	3.12	3.76	33.24	31.98	28.16	31.13
12	GJB-3	3.58	3.38	2.96	3.31	31.14	28.63	24.35	28.04
13	Pant Ruturaj	3.80	3.69	3.12	3.54	28.29	26.82	31.12	28.74
14	Surati Ravaiya	4.21	4.32	3.65	4.06	36.24	34.87	29.17	33.43
15	Doly-5	3.63	3.48	3.12	3.41	28.35	25.85	30.46	28.22
	S.Em+	0.06	0.28	0.15	0.11	0.40	0.96	0.63	1.98
	CD. @ 5%	0.18	0.81	0.43	0.30	1.17	2.77	1.83	5.72
	CV%	2.50	11.71	7.56	8.23	2.17	5.29	3.59	3.88
	Year								
	S.Em+				0.048				0.883
	CD. @ 5%				NS				NS
	Y x T								
	S.Em+				0.186				0.701
	CD. @ 5%				NS				NS

Table 3: Per cent protein and Ascorbic acid content (mg/100 g) of brinjal fruits from different varieties and genotypes

Sr. No.	Name of Variety/ Genotype	Protein %			Pooled over the year	Ascorbic Acid mg/100g			Pooled over the year
		2014-15	2015-16	2016-17		2014-15	2015-16	2016-17	
1	AB-08-14	0.64	0.69	0.84	0.72	9.6	11.3	12.10	11.00
2	AB-07-2-15	0.81	0.83	0.79	0.81	20.3	22.7	28.90	23.97
3	JB-10-208	0.87	0.91	0.73	0.84	15.4	14.8	12.40	14.20
4	JBGR-06-8	0.79	0.76	0.76	0.77	16.9	16.8	14.30	16.00
5	JBL-08-8	1.04	1.12	0.98	1.05	17.4	18.8	15.40	17.20
6	JBL 12-06-4-1	1.36	1.29	1.05	1.23	19.4	20.8	17.50	19.23
7	JBGR-1	1.29	1.32	1.03	1.21	21.5	22.6	18.60	20.90
8	GOB-1	1.43	1.36	1.12	1.30	21.8	22.9	26.70	23.80
9	Swarna Mani	1.20	1.24	1.03	1.16	13.7	14.5	12.30	13.50
10	GBL-1	0.80	0.96	0.85	0.87	19.3	20.6	22.50	20.80
11	GJB-2	0.63	0.82	0.75	0.73	20.1	21.3	17.30	19.57
12	GJB-3	0.77	0.89	0.68	0.78	14.1	15.5	21.50	17.03
13	Pant Ruturaj	1.07	1.11	0.98	1.05	17.8	18.6	15.60	17.33
14	Surati Ravaiya	0.67	0.74	0.84	0.75	10.8	13.4	12.60	12.27
15	Doly-5	0.84	0.81	0.73	0.79	14.0	13.9	13.30	13.73
	S.Em+	0.03	0.03	0.04	0.05	0.28	0.64	0.44	1.25
	CD. @ 5%	0.10	0.09	0.10	0.15	0.82	1.86	1.27	3.63
	CV%	6.34	5.36	7.12	6.25	2.90	6.22	4.38	4.77
	Year								
	S.Em+				0.023				0.560
	CD. @ 5%				0.066				NS
	Y x T								
	S.Em+				0.034				0.478
	CD. @ 5%				0.095				1.347

Table 4: Glycoalkaloid and Anthocyanine content of brinjal fruits from different varieties and genotypes

Sr. No.	Name of Variety/ Genotype	Glycoalkaloid ($OD.g^{-1}$)			Pooled over the year	Anthocyanine ($OD.g^{-1}$)			Pooled over the year
		2014-15	2015-16	2016-17		2014-15	2015-16	2016-17	
1	AB-08-14	0.42	0.39	0.54	0.45	2.49	2.56	2.38	2.48
2	AB-07-2-15	0.29	0.31	0.21	0.27	2.06	2.23	2.21	2.17
3	JB-10-208	0.44	0.39	0.32	0.38	2.52	2.62	2.44	2.53
4	JBGR-06-8	0.38	0.36	0.46	0.40	1.86	2.01	1.95	1.94
5	JBL-08-8	0.43	0.45	0.38	0.42	1.59	1.63	1.76	1.66
6	JBL 12-06-4-1	0.56	0.62	0.42	0.53	0.47	0.98	0.82	0.76
7	JBGR-1	0.26	0.24	0.36	0.29	0.62	0.85	0.75	0.74
8	GOB-1	0.34	0.36	0.26	0.32	1.03	0.96	0.91	0.97
9	Swarna Mani	0.52	0.49	0.42	0.48	2.13	2.23	2.29	2.22
10	GBL-1	0.42	0.39	0.33	0.38	1.98	2.03	2.11	2.04
11	GJB-2	0.36	0.32	0.28	0.32	1.86	1.98	1.19	1.68
12	GJB-3	0.36	0.37	0.27	0.33	1.59	1.76	1.82	1.72
13	Pant Ruturaj	0.33	0.34	0.39	0.35	1.62	1.52	1.59	1.58
14	Surati Ravaiya	0.34	0.32	0.38	0.35	1.88	1.98	1.81	1.89
15	Doly-5	0.29	0.31	0.41	0.34	1.46	1.36	1.46	1.43
	S.Em+	0.01	0.03	0.02	0.03	0.05	0.08	0.07	0.08
	CD. @ 5%	0.03	0.09	0.05	0.10	0.14	0.23	0.20	0.24
	CV%	4.92	13.92	7.82	9.66	5.04	7.62	7.01	6.70
	Year								
	S.Em+				0.015				0.038
	CD. @ 5%				NS				NS
	Y x T								
	S.Em+				0.021				0.066
	CD. @ 5%				0.059				0.187

Table 5: Enzymatic activity of PPO and PAL (OD. min /gm) of Brijal fruits from different varieties and genotypes

Sr. No.	Name of Variety/ Genotype	PPO (OD. min /gm)			Pooled over the year	PAL (OD. min /gm)			Pooled over the year
		2014-15	2015-16	2016-17		2014-15	2015-16	2016-17	
1	AB-08-14	1.11	0.98	1.24	1.11	1.23	1.36	1.43	1.34
2	AB-07-2-15	1.42	1.36	1.03	1.27	1.11	1.19	0.89	1.06
3	JB-10-208	1.35	1.25	1.95	1.52	0.98	0.91	1.12	1.00
4	JBGR-06-8	1.03	0.96	1.23	1.07	1.29	1.35	1.42	1.35
5	JBL-08-8	1.22	1.19	1.36	1.26	1.29	1.39	1.36	1.35
6	JBL 12-06-4-1	1.55	1.65	1.79	1.66	1.15	1.26	1.34	1.25
7	JBGR-1	1.76	1.85	1.92	1.84	1.36	1.29	1.22	1.29
8	GOB-1	1.59	1.47	1.15	1.40	1.33	1.37	1.08	1.26
9	Swarna Mani	1.26	1.32	1.46	1.35	1.18	1.28	1.36	1.27
10	GBL-1	1.51	1.62	1.12	1.42	1.24	1.36	1.42	1.34
11	GJB-2	1.24	1.19	1.23	1.22	1.23	1.11	1.24	1.19
12	GJB-3	1.26	1.15	1.05	1.15	1.22	1.19	0.96	1.12
13	Pant Ruturaj	1.48	1.55	1.69	1.57	1.15	1.19	1.21	1.18
14	Surati Ravaiya	1.39	1.42	1.23	1.35	1.24	1.26	1.31	1.27
15	Doly-5	1.55	1.62	1.96	1.71	1.19	1.03	1.13	1.12
	S.Em+	0.03	0.09	0.05	0.10	0.04	0.07	0.04	0.06
	CD. @ 5%	0.08	0.27	0.13	0.30	0.11	0.19	0.12	0.17
	CV%	3.65	11.66	5.58	7.69	5.55	9.27	6.02	7.16
	Year								
	S.Em+				0.046				0.03
	CD. @ 5%				NS				NS
	Y x T								
	S.Em+				0.062				0.05
	CD. @ 5%				0.174				0.14

Table 6: Flavanoid (mg/100 g) content and per cent acidity of brinjal fruits from different varieties and genotypes

Sr. No.	Name of Variety/ Genotype	Flavanoid mg/100g			Pooled over the year	Acidity %			Pooled over the year
		2014-15	2015-16	2016-17		2014-15	2015-16	2016-17	
1	AB-08-14	10.65	11.65	12.57	11.62	0.32	0.28	0.33	0.31
2	AB-07-2-15	9.25	9.19	10.11	9.52	0.18	0.22	0.18	0.19
3	JB-10-208	8.67	8.54	9.12	8.78	0.29	0.26	0.31	0.29
4	JBGR-06-8	7.48	8.41	9.56	8.48	0.38	0.41	0.38	0.39
5	JBL-08-8	6.68	5.98	7.83	6.83	0.24	0.26	0.22	0.24
6	JBL 12-06-4-1	9.72	10.12	10.53	10.12	0.22	0.23	0.24	0.23
7	JBGR-1	13.25	15.36	16.86	15.16	0.31	0.29	0.32	0.31
8	GOB-1	12.14	14.68	13.45	13.42	0.19	0.23	0.18	0.20
9	Swarna Mani	8.23	9.12	8.68	8.68	0.12	0.14	0.14	0.13
10	GBL-1	8.85	9.09	11.12	9.69	0.18	0.22	0.19	0.20
11	GJB-2	8.76	8.34	8.63	8.58	0.21	0.24	0.21	0.22
12	GJB-3	7.47	7.88	8.11	7.82	0.23	0.21	0.16	0.20
13	Pant Ruturaj	6.89	6.54	7.13	6.85	0.17	0.19	0.22	0.19
14	Surati Ravaiya	7.32	7.89	8.79	8.00	0.23	0.25	0.28	0.25
15	Doly-5	8.24	8.65	9.52	8.80	0.14	0.17	0.21	0.17
	S.Em+	0.20	0.43	0.23	0.36	0.01	0.02	0.01	0.01
	CD. @ 5%	0.58	1.25	0.66	1.06	0.03	0.06	0.03	0.04
	CV%	3.90	7.96	3.89	5.57	8.62	16.20	8.25	11.73
	Year								
	S.Em+				0.16				0.02
	CD. @ 5%				0.47				NS
	Y x T								
	S.Em+				0.31				0.04
	CD. @ 5%				0.86				NS

Table 7: Polymorphism of SSR primers across 15 brinjal genotypes

No.	SSR Primer Name	Allele/Band size (bp)	Total no. of Bands	Monomorp. Bands	Polymorphic bands			Polymor Phism (%)	PIC	SPI
					S	U	T			
1	EEMS06	129-383	8	0	3	5	8	100	0.86	6.88
2	EEMS07	115-1173	16	0	6	10	16	100	0.89	14.27
3	EEMS10	122-265	3	0	1	2	3	100	0.27	0.82
4	EEMS12	89-289	4	0	2	2	4	100	0.57	2.27
5	EEMS13	110-372	4	0	1	3	4	100	0.39	1.56
6	EEMS14	239	1	0	1	0	1	100	0.00	0.00
7	EEMS15	269-279	2	0	2	0	2	100	0.30	0.60
8	EEMS16	215-715	3	0	0	3	3	100	0.67	2.00
9	EEMS17	190	1	0	1	0	1	100	0.00	0.00
10	EEMS18	175-410	3	0	2	1	3	100	0.49	1.46
11	EEMS19	145-285	5	0	5	0	5	100	0.75	3.75
12	EEMS20	180-208	3	0	3	0	3	100	0.58	1.74
13	EEMS21	130-150	3	1	2	0	2	66.7	0.56	1.12
14	EEMS22	145-151	2	0	2	0	2	100	0.32	0.64
15	EEMS23	145	1	1	0	0	0	0	0.00	0.00
16	EEMS24	200-210	2	0	2	0	2	100	0.39	0.78
17	EEMS25	210-218	2	0	2	0	2	100	0.50	1.00
18	EEMS26	298-310	2	0	2	0	2	100	0.44	0.89
19	EEMS28	130-510	8	0	8	0	8	100	0.83	6.67
20	EEMS29	105	1	0	1	0	1	100	0.00	0.00
21	EEMS30	180-1200	3	1	0	2	2	66.7	0.21	0.43
22	EEMS31	155-395	4	0	2	2	4	100	0.44	1.74
23	EEMS32	135-150	2	0	2	0	2	100	0.41	0.82
24	EEMS33	191-198	2	0	2	0	2	100	0.00	0.00
25	EEMS34	241-250	2	0	2	0	2	100	0.23	0.46
26	EEMS35	202-211	2	0	2	0	2	100	0.23	0.46
27	EEMS36	98-120	3	1	2	0	2	66.7	0.51	1.02
28	EEMS37	109-202	3	0	3	0	3	100	0.62	1.86
29	EEMS38	140-155	2	0	2	0	2	100	0.24	0.49
30	EEMS39	245	1	1	0	0	0	0	0.00	0.00
31	EEMS42	140-510	3	0	3	0	3	100	0.57	1.70
32	EEMS44	190-200	2	0	2	0	2	100	0.26	0.52
33	EEMS46	145-175	3	0	3	0	3	100	0.59	1.76
34	EEMS47	210-235	2	0	1	1	2	100	0.13	0.27
35	EEMS48	135-210	3	0	3	0	3	100	0.66	1.98
36	EEMS49	130-350	7	0	3	4	7	100	0.94	6.61
37	EEMS50	200-220	2	0	2	0	2	100	0.48	0.96
38	Xgwm-617-6A	250-410	2	0	2	0	2	100	0.43	0.86
39	Xgwm-577-7B	120-145	3	0	1	2	3	100	0.46	1.38
40	Xgwm-428-7D	230-850	4	0	3	1	4	100	0.66	2.65
41	Xgdm-3-5D	145-500	4	2	2	0	2	50	0.69	1.37
42	Xgdm-19-1D	175-1100	5	0	5	0	5	100	0.79	3.96
43	Xgdm-61-4D	195-1485	8	0	7	1	8	100	0.81	6.47
44	Xgdm-33-A	320-950	5	0	3	2	5	100	0.73	3.67
45	Xgdm-62-3D	195-1090	10	0	0	10	10	100	0.90	9.00
46	Xgdm-93-4B	850	1	0	1	0	1	100	0.00	0.00
47	Xgdm-109-5A	130-1490	3	0	3	0	3	100	0.63	1.89
Total			165	7	107	51	158	-	-	-
Average			-	-	-	-	3.36	92.6	0.46	2.10

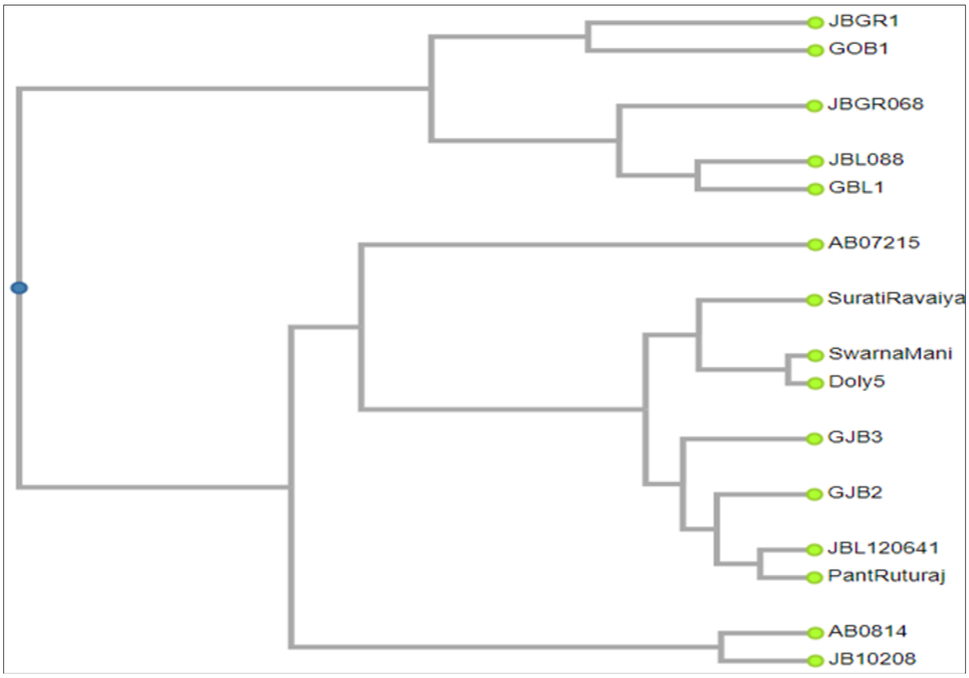

Fig 1: Dedrogram prepared using Biochemical Data (http://genomes.urv.cat/ UPGMA/ UPGMA boot_v12.cgi)

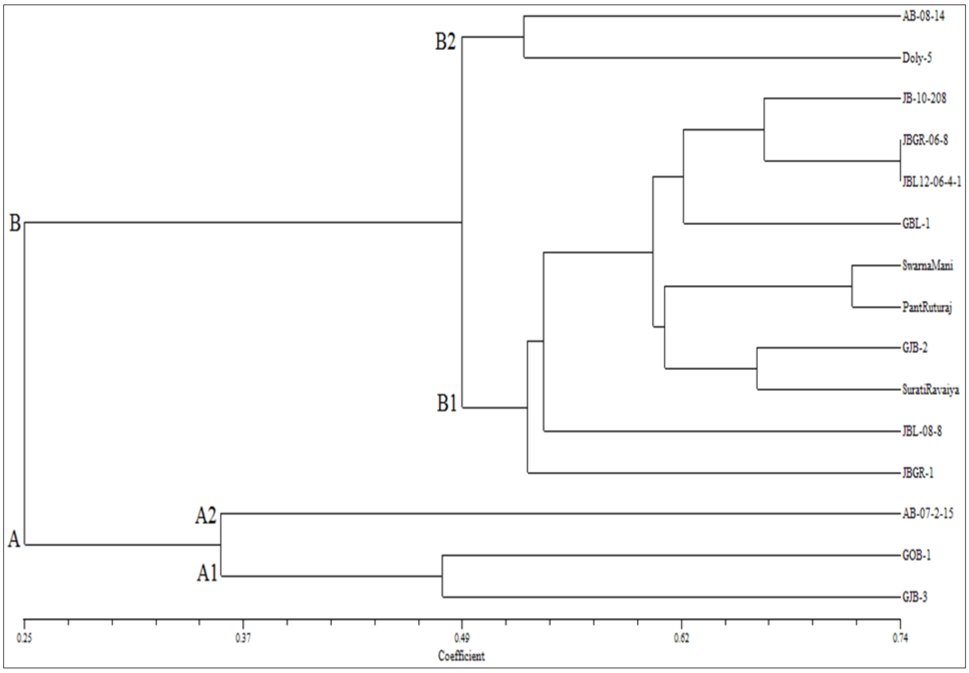

Fig 2: Dendrogram depicting genetic relationship between brinjal genoptypes

Conclusion

From the above results, the most diverse varieties were found to be GOB-1 and JBGR-1 compared to the other promising genotypes and varieties based biochemical, nutritional analysis. The diverse GOB-1 contained higher protein, total soluble solids, soluble sugars, phenols, ascorbic acid, PPO activity and flavanoid content and lower in glycoalkaloids and acidity. The clustering pattern on the basis of molecular analysis (SSR) depicting diverse varieties GOB-1 and GJB-3 out grouped from other genotypes with 48% similarity.

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