

MOLECULAR CHARACTERIZATION OF *FUSARIUM SOLANI* (MART.) SACC. ISOLATES INCITING DRY ROT OF POTATO TUBERS (*SOLANUM TUBEROSUM* L.)

JANKI PINAKINBHAI PANDYA* AND R. K. PATIL

Department of Plant Pathology, Anand Agricultural University, Anand - 388 011

e-mail: jankipandya00193@gmail.com

KEYWORDS

Fusarium solani
Molecular variation
RAPD-PCR
Fumonisin mycotoxin
Dry rot of potato tubers

Received on :

22.09.2015

Accepted on :

17.04.2016

*Corresponding
author

ABSTRACT

Fusarium dry rot of potato (*Solanum tuberosum* L.) is devastating postharvest disease worldwide and is incited by several species of *Fusarium* (Boyd, 1972). Twenty five isolates of *Fusarium solani*, collected from different places of India were subjected to analysis of genetic variability in terms of DNA polymorphism using RAPD-PCR. The primer OPA-6, OPA-7, OPA-11 and OPA-13 showed the highest percentage (100%) of polymorphism. The results of PCR amplification with fumonisin mycotoxin specific primer FUM1 showed that all isolates have a potential to produce the toxin fumonisin except isolates FS-6, FS-13 and FS-14.

INTRODUCTION

Potato is fourth most important food crop in India after rice, wheat and maize. The global production of potato is about 365 million metric tons covering an area of about 19.13 million hectare (Anon. 2014). The annual production of potato in India for the year 2013-14 was 41,555 MT (Anon. 2014). India contributes about 15 per cent of total potato production of the world. India is the second largest producer of potato (45 million MT) in the world after China (85 million MT).

After harvest, several species of *Fusarium* invade potato tubers, and mostly grew in rotted potato tubers as saprophytes. *Fusarium* dry rot is an important disease which affects tubers in storage as well as seed tuber pieces in the field (Choiseul *et al.*, 2006). Investigations carried on dry rot in the UK showed 70 per cent edible tubers and 100 per cent seed tubers were found infected with *Fusarium solani* (Mart.) Sacc. and more than 1 per cent of the tubers showed the symptoms of dry rot (Bradshaw *et al.*, 2001). Bhardwaj (2012) reported 25 to 60 per cent yield loss in India due to dry rot (*Fusarium solani* (Mart.) Sacc.). Crop losses attributed due to dry rot have been estimated to the tune of 6 to 25 per cent (Chelkowski, 1989). *Fusarium* spp. cause tuber rot, with severe reductions in tuber yield, upto 25 per cent annually (Lui and Kushalappa, 2002). Genetic diversity is commonly measured by genetic distance or genetic similarity, both of which imply that there are either differences or similarities at genetic level. This amplification technique (arbitrarily primed PCR on RAPD) can generate specific DNA fragments useful for genome mapping, identifica-

tion of isolates and application in molecular biology (Hadryz *et al.*, 1992).

Fusarium species are distributed worldwide as saprophytes, soil inhabitants, and parasites of many plants. Some isolates of certain species are capable of producing mycotoxins. During the past few decades, interest in *Fusarium* species has increased because they produce numerous mycotoxins involved in various mycotoxicoses of humans and animals (Marasas *et al.*, 1984).

The objective of the study is to characterization of genetic variability in *F. solani* isolates an incitant of dry rot of potato tubers and to detection of fumonisin toxin using Random Amplification of Polymorphic DNA (RAPD).

MATERIALS AND METHODS

Collection of samples

The diseased potato tuber samples exhibiting typical dry rot symptoms were collected from 25 markets of India (Table 1). The infected samples were brought to the laboratory and were subjected to tissue isolation for further studies.

Fungal isolation and maintenance

The pure cultures obtained were maintained on PDA slant at 4°C temperature for further study.

Study on molecular variability among the isolates of *F. solani* using RAPD primers

Isolation and Purification of Genomic DNA

DNAs of twenty five purified isolates of *F. solani* were ex-

tracted using a modified CTAB method (Leisova *et al.*, 2005) and were purified by ethanol precipitation.

RAPD primer amplification and analysis

For the molecular characterization of *F. solani* isolates, the 20 different 10-mer RAPD primers (Eurofins Genomics India Pvt. Ltd.) were screened for the amplification of template DNA of *F. solani* (Table 2). The primers which gave clear and polymorphic amplification patterns were used for further analysis of all the 25 isolates of *Fusarium solani*. The amplified DNA fragments for each accession were scored as present (1) or absent (0). Data generated by 20 RAPD primers were used to compile a binary matrix for cluster analysis. Genetics similarity among accessions was calculated according to Jaccard's coefficient (Jaccard, 1908)

RESULTS

Assay of DNA from the isolates of *Fusarium solani* obtained through Nanodrop technique

The results of spectro photometric DNA analysis showed variability in DNA concentration in mycelia of twenty five isolates of *F. solani* (Table 3). DNA concentration was found ranging from 189.80 to 902.20 ng/ μ l, while the absorbance ratio of DNA at 260/280 wavelength was 1.80 to 2.10. Quality of DNA was further confirmed on agarose gel electrophoresis. DNA thus extracted was of good quality and utilized for molecular markers study through RAPD markers.

Random Amplification of Polymorphic DNA (RAPD) study

In recent years, molecular markers have received arable attention and have been used for genetic diversity, phylogenetic and evolutionary studies and mapping and tagging of agronomically important traits in different plant species. It can also

be used in the study of molecular diversity in microorganism such as fungus, bacteria, viruses, etc. The present investigation was carried out with the twenty five different isolates of *F. solani* to study the molecular variability among these isolates.

The study showed a high level of genetic variability between twenty five isolates of *F. solani*. The highest polymorphism (100%) was recorded by the primer OPA-6, OPA-7, OPA-11 and OPA-13. Whereas, the lowest polymorphism was observed in the primer OPA-4 (77.78%) (Table 4).

These six RAPD primers amplified a total of 55 scorable alleles of which 51 loci were polymorphic, with a range of 6 to 13 alleles per primer. Overall, *Fusarium solani* isolates exhibited high level of genetic diversity. The maximum 13 alleles were generated by OPA-15; whereas, primer, OPA-4, OPA-6, OPA-7 and OPA-13 generated 9, 10, 8 and 9 alleles, respectively. The lowest number of alleles were generated by primer OPA-11 (6 alleles). This result showed the ability of RAPD to discriminate among isolates and suggested their application for species identification.

A total of 20 Primers consisted of di-nucleotide repeat motifs were used for initial screening with two isolates. Out of these, 14 primers gave no amplification at all, while only six primers produced clear banding patterns, and were subsequently used to analyze the entire set of 25 isolates. The fragment size was detected by comparing the amplicons with a 100 bp Ladder.

Primer OPA-4 (AATCGGGCTG):

A maximum of 9 DNA alleles (Fig.1) were observed in OPA-4 primer. This primer showed 7 polymorphic loci out of 9 loci with 77.78 per cent polymorphism and the PIC value of 0.877 was observed (Table 4).

Primer OPA-6 (GGTCCCTGAC):

The RAPD profile of OPA-6 (Fig.1) produced 10 alleles which

Table 1: Statement showing the sources of *Fusarium solani* isolates inciting dry rot of potato tubers

Sr. No.	Name of Isolate	Market	District	State
1.	FS-1	Sardar Patel vegetable market	Anand	Gujarat
2.	FS-2	Goya Talav market	Anand	---
3.	FS-3	Jamalpur market	Ahmedabad	---
4.	FS-4	APMC market	Navsari	---
5.	FS-5	Talav gate vegetable market	Junagadh	---
6.	FS-6	Idar vegetable market	Sabarkantha	---
7.	FS-7	Sardar market	Surat	---
8.	FS-8	Halol local vegetable market	Panchmahals	---
9.	FS-9	Local vegetable market	Bhavnagar	---
10.	FS-10	W.T.P.S	Kheda	---
11.	FS-11	Khanderao market	Vadodara	---
12.	FS-12	Local vegetable market	Rajkot	---
13.	FS-13	APMC market	Banaskantha	---
14.	FS-14	Azadpur mandi	North West	New Delhi
15.	FS-15	Shahu market yard	Kolhapur	Maharashtra
16.	FS-16	Choithram market	Indore	Madhya Pradesh
17.	FS-17	Naini sabji mandi	Varanasi	Uttar Pradesh
18.	FS-18	Sarai road vegetable market	Ludhiana	Punjab
19.	FS-19	Palayam market	Thiruvanantha - puram	Kerala
20.	FS-20	Mukharjee chowk mandi	Udaipur	Rajasthan
21.	FS-21	Bazar samiti	Nalanda	Bihar
22.	FS-22	Ek number market	Khurda	Orissa
23.	FS-23	APMC market	Kullu	Himachal Pradesh
24.	FS-24	LBS market	Bijapur	Karnataka
25.	FS-25	Sheoraphuli bazar	Hoogly	West Bengal

Table 2: Statement showing the list of primers used for RAPD analysis and toxin detection of *Fusarium solani*

Primer	Sequence(5'-3')	Temp. Value (°C)	GC Content(%)
OPA1	CAGGCCCTTC	34	70
OPA2	TGCCGAGCTG	34	70
OPA3	AGTCAGCCAC	32	60
OPA4	AATCGGGCTG	32	60
OPA5	AGGGGTCTTG	32	60
OPA6	GGTCCCTGAC	34	70
OPA7	GAAACGGGTG	32	60
OPA8	GTGACGTAGG	32	60
OPA9	GGGTAACGCC	34	70
OPA10	GTGATCGCAG	32	60
OPA11	CAATCGCCGT	32	60
OPA12	TCGGCGATAG	32	60
OPA13	CAGCACCCAC	34	70
OPA14	TCTGTGCTGGC	32	60
OPA15	TTCCGAACCC	32	60
OPA16	AGCCAGCGAA	32	60
OPA17	GACCGTTGT	32	60
OPA18	AGGTGACCGT	32	60
OPA19	CAAACGTCGG	32	60
OPA20	GTTGCGATCC	32	60
FUM1 For.	CCATCACAGTGGGACACAGT	53.8	55
FUM1 Rev.	CGTATCGTCAGCATGATTAGC	52.4	47.6
FUM2 For.	GAGGCCCGAGCGAGCACTGG	62	75
FUM2 Rev.	CCAGCCGCGGAAATTAGGGATGTG	60.8	58.3

Table 3: DNA assay of the *Fusarium solani* isolates obtained through "Nanodrop technique"

Sr. No.	Isolate	(260/280)Wavelength Ratio	Concentration (ng/μl)
1	FS-1	1.98	902.20
2	FS-2	2.05	644.36
3	FS-3	1.80	423.21
4	FS-4	1.92	854.50
5	FS-5	2.01	326.12
6	FS-6	2.04	569.38
7	FS-7	1.97	535.29
8	FS-8	2.08	224.56
9	FS-9	1.95	468.20
10	FS-10	1.91	820.21
11	FS-11	1.85	189.80
12	FS-12	2.10	830.80
13	FS-13	1.89	278.16
14	FS-14	1.95	486.14
15	FS-15	2.03	671.92
16	FS-16	1.84	347.89
17	FS-17	1.92	425.50
18	FS-18	2.08	321.06
19	FS-19	2.02	762.20
20	FS-20	1.82	508.50
21	FS-21	1.96	281.23
22	FS-22	2.06	365.20
23	FS-23	1.82	195.90
24	FS-24	1.96	254.30
25	FS-25	2.10	424.20

were found to be polymorphic showing 100 per cent polymorphism. The PIC value of 0.856 was observed (Table 4).

Primer OPA-7 (GAAACGGGTG)

Primer OPA-7 (Fig.1) amplified upto 8 alleles and all of them were polymorphic and hence showed 100 per cent polymorphism. The PIC value of 0.873 was obtained (Table 4).

Primer OPA-11 (CAATCGCCGT)

The PCR amplification with primer OPA-11 (Fig.2) generated 6 alleles out of which all were polymorphic with 100 per cent polymorphism. The PIC value for the primer was found to be 0.772.

Primer OPA-13 (CAGCACCCAC)

The RAPD profile of OPA-13 (Fig. 2) produced 9 alleles which were found to be polymorphic showing 100 per cent poly-

Table 4: Details of amplification obtained with different RAPD primers

Sr. No	Name of primer	Sequence (5'-3')	Annealing temperature	No. of total	No. of polymorphic	No. of mono	Polymorphism percent (%)	Total No. of loci amplified	PIC* value
1	OPA-4	AATCGGGCTG	32	9	7	2	77.78	173	0.877
2	OPA-6	GGTCCCTGAC	34	10	10	0	100	134	0.856
3	OPA-7	GAAACGGGTG	32	8	8	0	100	163	0.873
4	OPA-11	CAATCGCCGT	32	6	6	0	100	82	0.772
5	OPA-13	CAGCACCCAC	34	9	9	0	100	126	0.839
6	OPA-15	TTCCGAACCC	32	13	11	2	84.61	240	0.911
Total				55	51	4	562.39	918	5.128
Average			9.16	8.5	0.66	93.73	153	0.854	

*PIC-Polymorphism Information Content

morphism. The PIC value of 0.839 was observed (Table 4).

Primer OPA-15 (TTCCGAACCC):

The RAPD banding pattern generated through OPA-15 (Fig.2) showed maximum of 13 alleles out of which 11 were polymorphic and recorded 84.61 per cent polymorphism. The PIC value was 0.911 (Table 4).

Cophenetic Correlation Test

Cophenetic correlation value for RAPD was found to be 0.44 to 0.89. The R value obtained was 0.86. These cophenetic correlation values indicated that there was high level of diversity detected by RAPD despite the number of RAPD markers used in study was less.

Pooled RAPD

Dendro gram was constructed according to Jaccard's coefficient (Jaccard, 1908) among the isolates of *F.solani* using the Un weighted Pair Group Method of Arithmetical averages (UPGMA) algorithm. The similarity coefficient ranged from 0.35 to 0.86 with 6 primers (Table 5). Highest similarity was observed between FS-10 and FS-13 (0.86), while lowest similarity was observed between FS-6 and FS-1 (0.35).

The dendro gram clearly revealed that one big cluster of twenty four isolates, while FS-6 isolate was separately situated in similarity matrix of 0.349 with FS-1. All isolates were sub divided in three clusters A, B and C. The cluster A contained twenty two isolates i.e. FS-1, FS-2, FS-19, FS-3, FS-5, FS-4, FS-7, FS-10, FS-13, FS-14, FS-11, FS-17, FS-23, FS-24 FS-12, FS-25, FS-20, FS-21, FS-18, FS-22, FS-8 and FS-9, while cluster B contained two isolates i.e. FS-15 and FS-16. Cluster C contained only one isolate i.e. FS-6 which was diverged from all twenty four isolates. The sub-cluster A₁ consisted FS-1, FS-2 and FS-19 isolates, while sub-cluster A₂ contained nineteen isolates i.e. FS-3, FS-5, FS-4, FS-7, FS-10, FS-13, FS-14, FS-11, FS-17, FS-23, FS-24 FS-12, FS-25, FS-20, FS-21, FS-18, FS-22, FS-8 and FS-9. Among all isolates, FS-3 and FS-5 showed high genetic similarity, while FS-7, FS-10 and FS-13 were found closely related.

In present study total 20 primers of OPA series were screened against isolates of *Fusarium solani*. Out of these 6 primers were found useful for amplification of DNA of *F. solani*. Among 6 primers the primer viz., OPA-6, OPA-7, OPA-11 and OPA-13 showed 100 per cent polymorphism, followed by OPA-15 (84.61%) and OPA-4 (77.78%) which helped to ascertain variability among the isolates. Therefore, these primers could be very useful for ascertaining variability among the population of other species of *Fusarium*.

Fumonisin is a new class of toxic secondary metabolites produced by *Fusarium* species. Two set of specific primer FUM1 For./FUM1 Rev. and FUM2 For./FUM2 Rev. were used to determine the Fumonisins mycotoxin. The results of PCR amplification with primer FUM1 showed that all isolates have a potential to produce the toxin fumonisin except FS-6, FS-13 and FS-14, while results of PCR amplification with primer FUM2 showed that isolate FS-6 and FS-13 were not able to produce fumonisin (Fig.3). An amplification product with this primer was detected yielding 186 bp amplicon with FUM1 primer, while 230 bp amplicon with FUM2 primer

DISCUSSION

Results of the study revealed that PCR-based technique could be used not only to differentiate the *Fusarium* species from other genera of fungi but also to identify the *Fusarium* species producing fumonisin.

Gupta *et al.* (2009) studied the genetic polymorphism of the isolates of *Fusarium solani* and found that 10 randomly amplified polymorphic DNA markers (OPA1-OPA10) tested in the genome of *Fusarium solani* and grouped on the basis of allelic data. This pattern of genetic variability in the isolate was also supported by the analysis of the similarity indices and UPGMA based dendro gram.

Younes *et al.* (2013) studied the genetic variation among the isolates of *Fusarium solani* based on RAPD-PCR analysis and results revealed that high levels of variation in banding patterns of all isolates were noted unique banding profiles were obtained with two isolates of *F. solani* (I4 & I6).

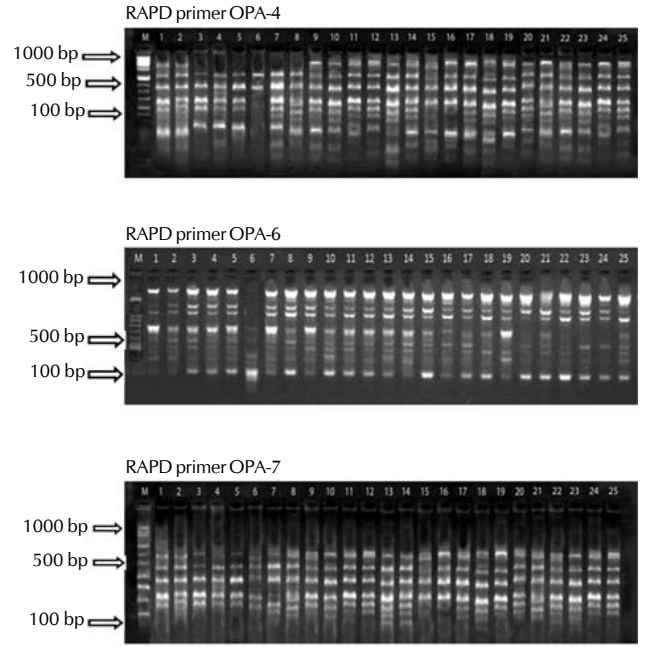
Sutthisa *et al.* (2010) studied genetic variability among 30 isolates of *Fusarium* spp. using random amplified polymorphic DNA (RAPD) markers. Four primers A02, A03, A07 and A09 produced fingerprint profiles which clearly distinguished between the different species of *Fusarium*.

Sankar *et al.* (2014) studied molecular characterization of *Fusarium solani* isolates inciting dry root rot of sweet orange using random amplified polymorphic DNA (RAPD) markers. Twelve representative isolates were characterized using 20 random primers of OPM series, out of which, 17 primers produced 174 amplified products, indicating 90.22 per cent polymorphism.

Sreenivasa *et al.* (2006) reported that out of 32 isolates of *Fusarium* species, 8 isolates showed a positive result with FUM1 gene set of primers at 183 bp, while no bands were seen in all other isolates of *Fusarium* and genera of *Aspergillus* and *Alternaria*. All the tested samples of *F. verticillioides*, *F. proliferatum*

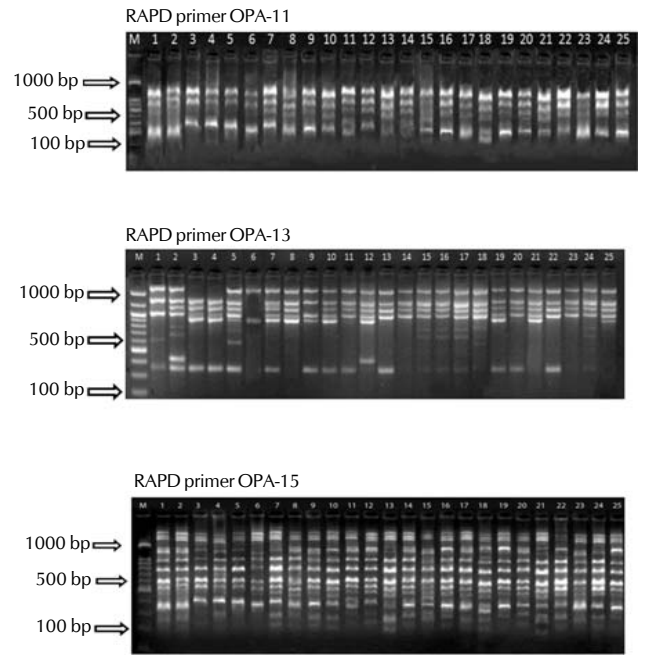
Table 5: Jaccard's similarity coefficient between twenty five isolates of *F.solani* based on RAPD data

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	
FS-1	1																									
FS-2	0.83	1																								
FS-3	0.71	0.67	1																							
FS-4	0.6	0.64	0.86	1																						
FS-5	0.67	0.64	0.89	0.82	1																					
FS-6	0.35	0.4	0.45	0.49	0.73	1																				
FS-7	0.65	0.65	0.73	0.74	0.73	0.44	1																			
FS-8	0.6	0.6	0.67	0.67	0.67	0.45	0.77	1																		
FS-9	0.6	0.6	0.6	0.6	0.6	0.49	0.76	0.63	1																	
FS-10	0.62	0.62	0.72	0.73	0.72	0.46	0.85	0.76	0.79	1																
FS-11	0.71	0.71	0.79	0.76	0.79	0.47	0.72	0.71	0.67	0.79	1															
FS-12	0.7	0.73	0.7	0.7	0.73	0.49	0.78	0.77	0.72	0.81	0.77	1														
FS-13	0.61	0.61	0.64	0.61	0.64	0.39	0.79	0.67	0.7	0.86	0.74	0.69	1													
FS-14	0.67	0.7	0.67	0.6	0.67	0.43	0.75	0.7	0.73	0.82	0.81	0.79	0.8	1												
FS-15	0.59	0.63	0.51	0.55	0.51	0.46	0.47	0.51	0.48	0.51	0.63	0.54	0.5	0.55	1											
FS-16	0.6	0.71	0.57	0.64	0.6	0.41	0.62	0.64	0.53	0.69	0.67	0.7	0.64	0.7	0.67	1										
FS-17	0.72	0.75	0.72	0.65	0.72	0.44	0.69	0.61	0.67	0.76	0.83	0.73	0.75	0.78	0.64	0.65	1									
FS-18	0.63	0.6	0.7	0.6	0.7	0.45	0.65	0.63	0.63	0.75	0.78	0.69	0.7	0.77	0.58	0.63	0.74	1								
FS-19	0.79	0.78	0.71	0.61	0.67	0.34	0.69	0.57	0.67	0.72	0.74	0.66	0.71	0.77	0.52	0.64	0.71	0.78	1							
FS-20	0.69	0.69	0.65	0.62	0.69	0.48	0.7	0.69	0.61	0.77	0.8	0.78	0.72	0.79	0.53	0.73	0.8	0.76	0.72	1						
FS-21	0.7	0.7	0.74	0.71	0.74	0.49	0.76	0.7	0.66	0.75	0.7	0.8	0.7	0.73	0.55	0.7	0.74	0.73	0.7	0.8	1					
FS-22	0.59	0.56	0.7	0.63	0.74	0.44	0.71	0.66	0.62	0.78	0.73	0.76	0.69	0.72	0.47	0.62	0.7	0.77	0.66	0.7	0.8	1				
FS-23	0.62	0.62	0.65	0.59	0.65	0.44	0.63	0.62	0.68	0.73	0.8	0.74	0.72	0.75	0.57	0.58	0.8	0.76	0.69	0.74	0.75	0.71	1			
FS-24	0.61	0.61	0.72	0.65	0.72	0.46	0.77	0.68	0.67	0.77	0.8	0.78	0.75	0.78	0.56	0.64	0.8	0.75	0.65	0.73	0.75	0.74	0.77	1		
FS-25	0.72	0.76	0.76	0.73	0.76	0.46	0.73	0.68	0.67	0.73	0.76	0.82	0.65	0.71	0.52	0.68	0.69	0.67	0.76	0.7	0.79	0.67	0.7	0.73	1	



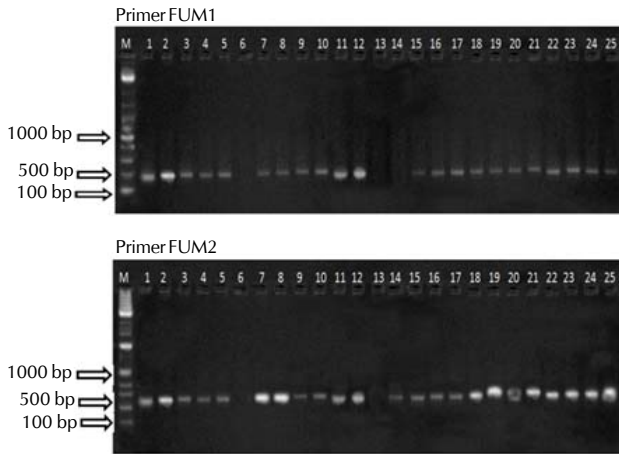
M - 100 + bp
1 - FS1, 2 - FS2, 3 - FS3, 4 - FS4, 5 - FS5, 6 - FS6, 7 - FS7, 8 - FS8, 9 - FS9, 10 - FS10, 11 - FS11, 12 - FS12, 13 - FS13, 14 - FS14, 15 - FS15, 16 - FS16, 17 - FS17, 18 - FS18, 19 - FS19, 20 - FS20, 21 - FS21, 22 - FS22, 23 - FS23, 24 - FS24, 25 - FS25

Figure 1: RAPD amplification pattern of 25 isolates of *F.solani* using primers OPA-4, OPA-6 and OPA-7.



M - 100 + bp
1 - FS1, 2 - FS2, 3 - FS3, 4 - FS4, 5 - FS5, 6 - FS6, 7 - FS7, 8 - FS8, 9 - FS9, 10 - FS10, 11 - FS11, 12 - FS12, 13 - FS13, 14 - FS14, 15 - FS15, 16 - FS16, 17 - FS17, 18 - FS18, 19 - FS19, 20 - FS20, 21 - FS21, 22 - FS22, 23 - FS23, 24 - FS24, 25 - FS25

Figure 2: RAPD amplification pattern of 25 isolates of *E solani* using primers OPA-11, OPA-13 and OPA-15.



M - 100 + bp
 1 - FS1, 2 - FS2, 3 - FS3, 4 - FS4, 5 - FS5, 6 - FS6, 7 - FS7, 8 - FS8, 9 - FS9, 10 - FS10, 11 - FS11, 12 - FS12, 13 - FS13, 14 - FS14, 15 - FS15, 16 - FS16, 17 - FS17, 18 - FS18, 19 - FS19, 20 - FS20, 21 - FS21, 22 - FS22, 23 - FS23, 24 - FS24, 25 - FS25

Figure 3: PCR amplification pattern of 25 isolates of *F. solani* using funonisin specific primers FUM1 and FUM2

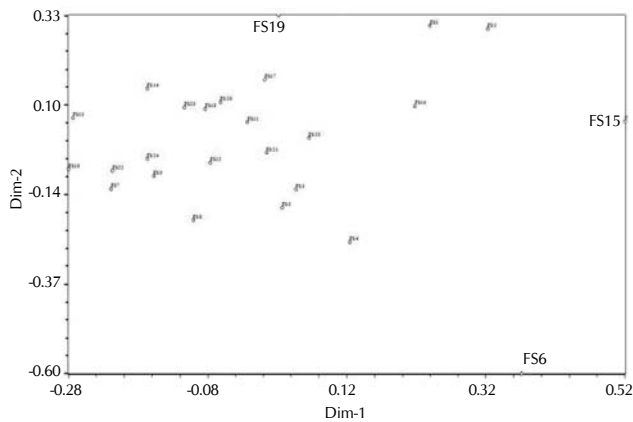


Figure 5: 2D plating of 25 isolates of *Fusarium solani* as the clustering method for RAPD

and *F. anthophilum* exhibited the FUM1 gene of Fumonisin production and amplified fragments with the molecular size of 183 bp (Yazeed *et al.*, 2011).

ACKNOWLEDGEMENT

Authors are grateful to the Professor and Head, Department of Plant Pathology and Principal, B. A. College of Agriculture, Anand Agricultural University, Anand (Gujarat) for providing necessary facilities.

REFERENCES

Anonymous 2014. "National Horticulture Board" <http://nhb.gov.in/area-pro>.
Anonymous 2014. "FAOSTAT database" <http://faostat.fao.org/site/297/default.aspx>.
Bhardwaj, S. K. 2012. Evaluation of Plant Extracts as Antifungal Agents Against *Fusarium solani* (Mart.) Sacc., *World J. Agril. Sci.* **8(4):**

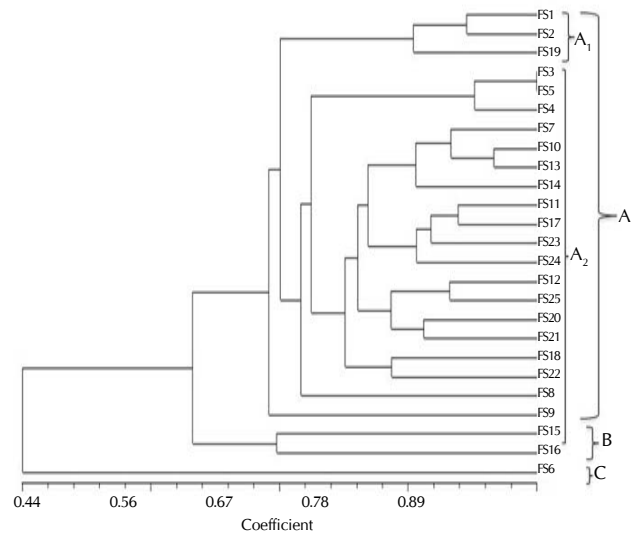


Figure 4(a): Dendrogram of 25 isolates of *Fusarium solani* on jaccard's similarity coefficient using UPGMA as the clustering method for REPD.

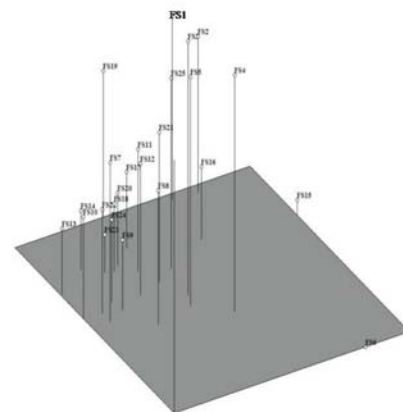


Figure 6: 3D plotting isolates of *Fusarium solani* as the clustering method for RAPD

385-388.
Bradshaw, N. J., Turner, J. A. and Elcock, S. J. 2001. Potatoes: A survey of disease 2000/01. Ministry of Agriculture, Fisheries and Food Report.
Boyd, A. E. W. 1972. Potato storage diseases. *Rev. Plant Pathol.* **51:** 297-321.
Chelkowski, J. 1989. Toxinogenicity of *Fusarium* species causing dry rot of potato tubers. In: *Fusarium Mycotoxins, Taxonomy and Pathogenicity* (J. Chelkowski, ed.). Elsevier Publishing Co., New York. pp. 435-440.
Choiseul, J., Allen, L. and Carnegie, S. 2006. Fungi causing dry tuber rots of seed potatoes in storage in Scotland. *Potato Res.* **49:** 241-253.
Gupta, V. K., Misra, A. K., Gaur, R., Pandey, R. and Chauhan, U. K. 2009. Studies of genetic polymorphism in the isolates of *Fusarium solani*. *Australian J. Crop Sci.* **3(2):** 101-106.
Hadrys, H., Balack, M. and Scherwater, B. 1992. Applications of random amplified polymorphic DNA (RAPD) in molecular ecology. *Mol. Ecol.* **1:** 55-63.
Jaccard, P. 1908. Nouvelles recherches sur la distribution florale.

Bulletins of Sco. Vaud. Sci. Nat. **44**: 223-270.

Leisova, L., Kucera, L., Minarikova, V. and Ovesna, J. 2005. AFLP-based PCR markers that differentiate spot and net form of *Pyrenophora teres*. *Plant Pathology*. **54**: 66-74.

Lui, L. H. and Kushalappa, A. C. 2002. Response Surface Models to Predict Potato Tuber Infection by *F. sambucinum* from Duration of Wetness and Temperature, and Dry Rot Lesion Expansion from Storage Time and Temperature. *Int. J. Food Microbiol.* **76**: 19-25.

Marasas, W. F. O., Nelson, P. E. and Tousson, T. A. 1984. Toxigenic *Fusarium* species, identity and mycotoxicology. Pennsylvania State University Press, University Park.

Sankar, T. G., Gopal, K., Gopi, V. and Sreenivasulu, Y. 2014. Molecular characterization of *Fusarium solani* isolates causing dry root rot of sweet orange (*Citrus sinensis* osbeck). *Int. J. Curr. Microbiol. App. Sci.* **3(3)**: 105-114.

Sreenivasa, M. Y., Dass, R. S., Charith Raj, A. P. and Janardhana, G. R. 2006. Molecular detection of fumonisin producing *Fusarium* species of freshly harvested maize kernels using polymerase chain reaction (PCR). *Taiwania*. **51(4)**: 251-257.

Sutthisa, W., Sanoamuang, N. and Chuprayoon, S. 2010. Morphological and molecular characterization of *Fusarium* spp., the fungi associated with mulberry root rot disease in north-eastern Thailand. *Thai J. Bot.* **2(1)**: 25-39.

Yazeed, H. A. E., Hassan, A. Reda, E.A., Hamed, M. and Refai, M. 2011. Molecular Detection of Fumonisin-producing *Fusarium* Species in Animal Feeds Using Polymerase Chain Reaction (PCR). *J. Appl. Sci. Res.* **7(4)**: 420-427.

Younes, H. M., Elamri, N. A. and Farag, I. S. 2013. Genetic variation of *Fusarium solani* isolates based on RAPD-PCR analysis. *Persian Gulf Crop Prot.* **2(3)**: 10-17.

