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STUDIES ON PREVALENCE AND IDENTIFICATION OF NEW RACES OF *FUSARIUM MONILIFORME* SHELDON INCITANT OF POKKAH BOENG DISEASE FROM UTTAR PRADESH

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Abstract

Field survey conducted during 2011-12 revealed 1.4-30 per cent incidences recorded in six sugarcane varieties *viz.* CoS8432, CoS8436, CoS98259, CoLk8102, CoJ64 and CoSe01424 of pokkah boeng disease. The characteristics of these varieties observed that juice quality was, dark and dusty, while sugar purity and ccs percent were found lesser followed by healthy canes. These varieties were made into twenty four isolates and pure cultures were maintained upto 12 days on different solid and liquid media in which PDA medium followed by Richard's medium was best suited for the growth of all isolates at 28^0C temperature. On the 12 day of incubation, the dry weight of the mycelial mat was recorded in the lower concentration of IBA followed by IAA proved stimulatory to the growth of selected isolates, while higher concentration produced adverse effect on the growth of all the isolates. Out of 24 isolates of *Fusarium moniliforme* only 10 isolates were found pathogenic. On the basis of physiological characters in which 6 isolates exhibited maximum radial growth (70 mm diam) in 8 days, while the same isolates attained maximum radial growth (80 mm diam) in 10 days. Hence, these isolates (Fm111, Fm114, Fm118, Fm1112, Fm1116 and Fm1120) took 12 days to attain (90 mm diam) growth and rest isolates unable and variable radial growth were found same period. On the basis of morphological and pathological characters these were categorized into six groups and pathotypes. On comparison with known pathogens and isolates were tested on 8 pathological differentials of sugarcane their reaction of *Fusarium moniliforme*. Out of 5 pathotypes, 3 and 5 were found similar to races 1(Gorakhpur), 4 with race 3 (Lakhimpur Kheri), 6 with race 2 (Meerut) and 2 with race 6 (Florida) but pathotype 1 was found all together different from all races. Hence, it was concluded that these two isolates belonging to new races are first time reported from Uttar Pradesh.

***Key words*:** Sugarcane, Race, *Fusarium moniliforme.*

Introduction

Sugarcane is one of the most important cash crops and pivotal role in Indian economy. India is the largest consumer of sugar in the world with annual consumption of about 19 million Mt and the second largest producer of sugar next to Brazil, with production in the sugar year 2011-12 crossing 28 million Mt. Uttar Pradesh occupies as premier position of sugarcane cultivation accounting for 42.2 million hectare and 30% of the area and production respectively. Average cane yield of UP (58.2 MT/ha) is very lower than the national average (66.9 MT/ha), and sugar recovery is also lower (9.51%) than national average (10.28%), respectively due to the several diseases by Vishwakarma *et al.,* (2013). Off all the sugarcane diseases such as fungal, bacterial, viral and phytoplasmal diseases, fungal diseases are gaining international importance (Bharti *et al*., 2012). Pokkah boeng of sugarcane caused by *Fusarium moniliformae* and the pathogen was first described by Sheldon (1904) and the perfect stage of pathogen is *Gibberella fujikuroi* (Nirenberg and Donnell, 1998). *Fusarium* is now confirmed by several workers as a causal agent of pokkah boeng in sugarcane in Asia as well established pathogen by many workers (Higgy *et al.*, 1977; Singh *et al*., 2006; Govender *et al*., 2010 and Mohammadi *et al*., 2012). Pokkah Boeng is a Javanese term denoting a malformation or distorted top was originally in Java but in that time no causal agent was established and its incidence was recorded by Patil *et al*., (2007). Pokkah Boeng disease recorded in all over the countries where sugarcane grown and pathogen spreads in wind-blown rain, infected cane cuttings, pupae and adults of sugarcane stem borers (Whittle and Irawan, 2000). This disease was well-known in sugarcane for long time but severity of disease reported in two commercial varieties Co7219 and Co C671 in Maharashtra 1983-1984 (Patil and Hapase, 1987). *Fusarium moniliforme* var. *subglutinans* reduce the quality of the harvested crop mainly among varieties with high sugar yields the sugar production depending upon the variety upto 40.8 - 64.5 per cent (Duttamajumder, 2004). Pokkah boeng disease of sugarcane has associated with several disease of sugarcane such as sett rot, root rot, wilt (Waraitch and Kumar, 1982). Pokkah boeng is now playing a very important role due to its economic threats in UP. He has been recorded during survey years 2007-10 showed increasing trend of disease incidence and most of the commercial cultivars affected by the disease ranged from 1 - 90 per cent (Vishwakarma *et al*., 2013). Although Pokkah boeng comes under minor concern but these days it is going to be major on basis of their rapid epidemiology during last few years. The incidence of pokkah boeng will be recorded 1.4 - 30 percent during our survey (2011-12) from Uttar Pradesh. Although Pokkah boeng comes under minor concern but these days it is going to be major on basis of their rapid epidemiology during last few years. Nowadays, the incidence and severity of pokkah boeng disease has been reported from major sugarcane growing states like Uttar Pradesh, Maharashtra, Punjab, Haryana, Assam, Tamil Nadu and Bihar in India and other sugarcane growing countries. Infection of the disease is caused either by spores or ascospores. The pathogen makes its entry into the host tissues through any sort of injury made by insects or

borers or natural growth cracks etc. After the entry, the infection thread develops normal hyphae which grows within the host tissues for some time and then emerge out through the cells to the outer surface and develops acervuli. Rains and heavy dews usually wash the acervuli developed on nodes and internodes and the spores get lodged around the nodes behind the leaf sheath. The spores germinate and the mycelium gets established in bud scales, root primordial or leaf scars and later within the plant tissues. Pokkah boeng has become a major problem not only due to cane yield reduction but also by affecting juice quality.

The details of the symptoms as four phases as per the Chlorotic phase I, Chlorotic phase II, Acute phase and Knife Cut phase are suffering from complete under symptoms (Patil, 2002; Burgess, 1981; Kamal and Singh, 1979)). No information regarding morphological physiological and pathological variation among the isolates is available for the existing isolates in different state as such Uttar Pradesh. In Uttar Pradesh its incidence has also been reported from 1-30 per cent literature revealed that only four races of *Fusarium moniliforme* have been indentified and reported across the world, of which 2 are from India no work has been done in Uttar Pradesh with regard to the existence of races. Therefore, a study was conducted in Indian Institute of Sugarcane Research Lucknow, Uttar Pradesh and results are embodied herein.

Materials and Methods

***Collection of diseased samples and isolation of Fusarium moniliforeme Sheld*:**

A survey was undertaken during, (2011-12) of Uttar Pradish in sugarcane growing areas (Gorakhpur, Deoria, Kushinagar, Maharajganj, Lakhimpur Kheri, Hardi, Shahjahanpur, Bareilly, Bagpat, Meerut, Muzaffarnagar and Saharanpur). Six sugarcane varieties whose completed four phases of pokkah boeng symptoms were collected and observed as cane juice and cane weight quality parameter compare to healthy and infected sugarcane varieties *viz.* CoS8436, CoS98259, CoS8432, CoLk8102, CoJ64 and CoSe01424 whose rind color varied from light to dark (Singh and Singh, 2006). These infected cane samples made 24 isolates and collected for the purpose of the possible biotypes of the pathogen. Pokkah boeng were collected for 24 isolates of *Fusarium moniliforeme* strains were isolated from lesions on infected stem pieces (Table 1). Three 5-5 mm pieces of tissue were taken from the margin of infected tissues, surface sterilized by dipping in 1% sodium hypochlorite for 1 minute, immersed in 70% ethanol for 1 min and rinsed three times with sterilized water and finally dried in sterilized tissue paper (Agrwal and Singh, 1974). Cultures were placed on water agar and incubated at room temperature (26 - 28^{0}C). The growing edges of any fungal hyphae developing from the tissues were then transferred aseptically to potato dextrose agar medium (PDA) and fungi were identified following sporulation. Subcultures were obtained for each isolate using the procedure described (Agrawal and Sharabhoy, 1978; Arshad *et al.*, 1992). When the fungus showed sporulation, spore masses were pieced off with a sterilized weir loop and streaked on the surface of water agar. After inoculating overnight at 26±2^{0}C on Biological Oxygen Demand (BOD), single germinated spores were picked up with a sterilized needle and transferred to Austhan and agar medium.

In-vitro growth effects on different solid and liquid media, temperature and growth regulators hormone were selected for diametric growth and mycelium production with different aspects of *Fusarium moniliforme*. Pathogencity and cross inoculation studies proved that the same organism was responsible for causing pokkah boeng and last phase under controlled conditions. The isolation and pathogencity test was made following usual mycological techniques employing for the pathogen.

The reaction was recorded as resistant, moderately resistance, moderately susceptible and susceptible on the basis of 0 - 4 scale as suggested by (Patil, 2002) and final score was analyzed. The cultures of different isolates were maintained on PDA slants at 4^{0}C for further studies.

Radial growth of Fusarium moniliforme in different solid media

The effect of different solid media *viz.* Asthana and Hawkers, Brown's agar, Capek's agar, Czapek's Dox, Elliots agar, Oatmeal agar, Potato dextrose agar, Richard's agar and Sucrose ammonium nitrate prepared as described on the growth of the selected isolates. A mycelia disc of 5 mm diameter cut from 7 days old culture of test isolate was separately inoculated. Each treatment contained 5 replicates and the average calculated.

Growth on liquid media of Fusarium moniliforme

The effect of different liquid media *viz.* Elliot's medium, Dextrose yeast extract, Richard's medium, Asthana and Hawker's medium, Sucrose Ammonium Nitrate, Czapek's Dox, Brown's medium, Czapek's II medium, Dextrose Ammonium nitrate and Pfeiffer's medium prepared as above was poured and production of mycelium growth was calculated.

Growth effect on different temperatures of Fusarium moniliforme

Oat meal agar medium was prepared and 15 ml. of the molten medium was aseptically poured in presterilized petriplates. 5 mm discs of each isolates cut from 7 days old cultures were inoculated at different temperatures (10,15, 20, 25, 30, 35 and 40^{0}C separately and average calculated .

Growth effect on growth regulator of Fusarium moniliforme

Richards's broth medium proved in presterilized conical flasks containing 0.5 ml absolute alcohol and requisite quantities of indole-3 acetic acid (IAA) and Indole-3 butyric acid (IBA), separately so as to procure 1, 5, 10, 25 and 50 ppm concentrations of the growth regulators with respect to the volume of the medium. A mycelial disc of 5mm in diameter cut from 7 days old cultures of test isolate was aseptically inoculated in each assay flasks. On 12th day of incubation, the dry weight of the mycelial mat was recorded and average calculated.

Morphological Characteristics of Fusarium moniliforme

Cultural growth of morphological characters were studied on Austhan and Hawkers' of which only pathogenic isolates were categorized into six groups on the basis of morphological and pathological characters (Table 2). The mycelia disc (5 mm diam.) were taken from actively sporulating areas near the growing edge of 7 day old cultures and transferred to (PDA) medium and incubated at 26±2°C temperature. The replicate cultures of each isolate were investigated and culture were purified by single conidia, hyphae tip and identified on the basis of their morphological characters. Morphological characters like shape and size of mycelium, conidia and conidiophores of *F. moniliforme* were examined. Colony characters, colour and growth diameter of all the isolates were observed.

For studying the degree of sporulation and the spores of each isolates were harvested in presterilized conical flask containing 100 ml of distilled water. The suspension was thoroughly mixed in warring blender for 3 minute with the help of haemocytometer the amount of conidia/ ml of distilled water was calculated. At least 3 replicates were maintained for each isolate and each observation was repeated twice and averages were calculated. The shape of conidia was observed under high power of microscope and the length and width of the conidia was measured after calibrating the microscope with oculars and stage micrometer.

***Pathogen city assay*:**

The experiment was carryout during 2012-13 (Table 3) in the pathogenic variability of *F. moniliforme*, six isolates namely Fm111, Fm114, Fm118, Fm1112, Fm1116 and Fm1120 were tested on eight national pathological standard *viz.* Co997, Co1148, Co7717, CoC671, CoJ64, Kakhai, SES594 and CoS8436 pathotypes. Artificial inoculation was done on healthy standing canes of 6 months old crop in the month of July using IISR inoculators developed (Kumar *et al.*, 2011). Spore suspensions of all isolates were prepared in distilled water as described earlier for conidial germination. The concentration of spores was maintained to be 10^6spores/ml using a haemocytometer. The spore suspension was placed on the 3rd exposed internodes from bottom for keeping away from oxidation and contamination. (Zai-ul-Hussnain *et al.*, 2007). Parameters for assessing the pathogenesis on the basis of the international scale (0 to 4) were adopted as suggested by Higgy *et al*., (1977). For calculating the disease index, observations recorded on the nature with condition of top, Chlorotic phase I, Chlorotic phase II, Acute phase and Knife Cut phase by splitting open the canes 30 days after inoculation.

Results and Discussion

Sample collection and fungi isolation

During our survey infected cane varieties namely *viz.* CoS8436, CoS98259, CoLk8102, CoJ64, CoS8432 and CoSe01424 selected for morphological, physiological and pathological studies. It is a common experience that certain varieties give good quality of healthy cane while infected cane give poor quality, sugar purity and commercial cane sugar per cent were found less under the same conditions.

Wide spread incidence of pokkah boeng was recorded on naturally infected those sugarcane varieties. The maximum varietal incidence was recorded (CoS98259-30±6%) followed by (CoS8436-24±2%) and (CoSe01424-16.2±4%) in sugarcane growing areas of Uttar Pradesh (Table 1). Samples of pokkah boeng infected cane collected for the purpose of isolation of the possible biotypes of the pathogen. Pure cultures of all 24 isolates maintained and slants were allowed to grow in Petri plates (90 mm) containing 15 ml Potato Dextrose Agar (PDA) medium and cultures were incubated at 26±2°C. The growth of the isolate was measured at 48 h interval in mutually perpendicular directions up to 12 days and averages calculated (Table 1).The observation were recorded in revealed that there was considerable variation in the growth rate of different isolates. Maximum growth were found isolates *viz.* Fm111, Fm114, Fm118, Fm1112, Fm1116 and Fm1120 followed by 11, Fm113, Fm115, Fm1117, Fm1121 and Fm1123 and minimum growth were found only two isolates namely Fm119 and Fm1124.

Radial growth of Fusarium moniliforme in different solid media

The effect of different solid media *viz.* Asthana and Hawkers, Brown's agar, Capek's agar, Czapek's Dox, Elliots agar, Oatmeal agar, Potato dextrose agar, Richard's agar and Sucrose ammonium nitrate prepared as described on the growth of the selected isolates was calculated. The averages of the diametric growth calculated from 5 replicates are given in (Fig.1). The results reveal that (PDA) medium followed by Asthana and Hawkers medium was best suited for the growth of all isolates of *F. moniliforme* under study.

Growth on liquid media of F. moniliforme

The effect of different liquid media viz. Elliot's medium, Dextrose yeast extract, Richard's, Asthana and Hawker's, Sucrose Ammonium Nitrate, Czapek's Dox, Brown's, Czapek's II medium, Dextrose ammonium nitrate and Pfeffer's medium prepared as described in materials and methods were assessed and the average of 5 replicates are given in (Fig. 4). Results recorded for Richard's followed by Czapek's dox liquid media was found to be the best medium for the growth of all the isolates of *F. moniliforme*.

Effect of different temperatures on the growth of F. moniliforme

Potato dextrose agar medium was prepared and 15 ml of the molten medium was aseptically poured in presterilized petriplates and 5 mm discs of each isolates, cut from 7 days old cultures on PDA were inoculated in assay plates. To evaluate the effect of temperature, the assay plates were inoculated at different temperatures *viz.* 10, 15, 20, 25, 30, 35 and 40°C in separate and incubated. The assay plats used as controls, were incubated at room temperature 24±2°C. The growth of the fungal was recorded in mutually perpendicular directions of intervals of 2, 4, 6, 8, 10 and 12 days and averages recorded in (Fig.2). It is apparent from that the growths of all isolates were maximum at 28°C temperature and interestingly, very minimum growth was observed at 40°C temperature in any test isolates of *F. moniliforme*.

Effect of growth regulator hormones on the growth of F. moniliforme

Richards's broth medium proved in presterilized conical flasks containing 0.5 ml absolute alcohol and requisite quantities of indole-3 acetic acid (IAA) and indole-3 butyric acid (IBA), separately so as to procure 2, 4, 8, 16 and 32 ppm of the growth regulators with respect to the volume of the medium. A mycelial disc of 5 mm in diameter cut from 7 days old cultures of test isolate was aseptically inoculated in each assay flasks. Control sets contained Richard's medium and 0.5 ml absolute alcohol but were without any growth regulator. The control and treatment assay flasks were incubated at 26±2°C temperature. On the 12th day of incubation, the dry weight of the mycelial mat was recorded and averages of 5 replicates are given. The results recorded in (Fig. 3) revealed that lower concentration of IAA and IBA proved stimulatory to the growth of selected isolates, while higher concentration produced adverse effect on the growth of all the isolates.

Cultural growth and physiological characters of Fusarium moniliforme

The six isolates exhibited maximum radial growth (70 mm diam) in 8 days, while the same isolates attained maximum radial growth (80 mm diam) in 10 days. Remaining isolates (Fm111, Fm114, Fm118, Fm1112, Fm1116 and Fm1120) took 12 days to attain (90 mm diam) and rest isolates discemilar radial growth were found same period. However, on 6th day the maximum radial growth of (40 mm diam) were obtained the same isolates, while minimum growth of (24 mm diam) was obtained isolate (Fm113) in the same period in the (Table 1).

Morphological characters of Fusarium moniliforme

Out of 24 isolates of *Fusarium Moniliforme* only 6 isolates were preserved and further morphologically studies. These isolates were categorized into six groups on the basis of similarity and dissimilarity of different parameters as colony characters, growth of micro and macro conidia, septation and pathogenic behavior. The data of duplicated that fluffy to partially submerged colony was recorded in all groups and colour also varied from group to group. Maximum growth 20.98 - 46.64 × 3.36 - 6.66 mμ was recorded in group 6 whereas minimum 16.65 - 36.64 × 3.36 - 4.98mμ) in five group. Similarly number of septa also varied from 3 - 6 septa were found in groups 2, 3, 5 and 6 whereas 2 - 5 in septa groups 1 and 4 size of micro conidia also varied from 4.95 × 1.66 - 4.99 mμ was observed in group 4 and 5 whereas other group 1, 2 and 3 exhibited 4.66 - 4.98 × 1.66 - 499 mμ (Table 2). It is reported that 220 cultures of *Fusarium* on various culture media and found that in *Fusarium moniliforme*, there were sub species and the spores production was also markedly affected by 2 - 3% tartaric acid in the media (Patil, 2002). Prasad and Padwick 1939; Singh, 1958; Burgess, 1981) examined the morphology of the fungus *Fusarium* with the help of 21 days old cultures and observed that spores with one septation, two septation and three septation measured in the range of 6 – 14 ×5 mμ, 14 – 21 × 2 -4 mμ and 19 – 28 × 2 - 5 mμ respectively. Siti Nordahliawate *et al.*, (2008) described the morphology of the fungus *Fusarium oxysporum* which showed aerial mycelum white purple to violet in colour. Morphology of *Fusarium* species from 20 days old cultures and indicated that when grown on PDA it measured 9.3 to 29.7μm in the length and 2.7 to 6.0 μm in breath (Chattopadhyay and Gupta, 1967). Bourne (1953) discussed the identity of white and purple strains of *Fusarium* occurring in association with cane stalk rot a pokkah boeng disease in Florida. Khanna and Rafay (1953) have been shown that low and high proportion of glucose affects the length of the conidia of *Fusarium moniliforme* appreciably.

Pathogencity assay

Six isolates (Fm111, Fm114, Fm118, Fm1112, Fm1116 and Fm1120) were tested on 8 sugarcane pathological differentials *viz.* Co1148, Co7717, Co997, SES594, Khakai, CoC671, CoJ64 and CoS8436 and they showed distinct differences in their reaction to all the differentials. The maximum incidence was found isolates *viz.* Fm111 and Fm1120 followed by Fm1112 and rest isolates variable reaction on different genotypes (Table 3). These isolates were compared further tested for their reaction on same differentials of sugarcane to confirm the different pathotypes. There isolates were again tested and compared with identified races of this pathogen in the same eight differentials (Table 4). The results revealed that all the tested cultivars of sugarcane except CoS8436 exhibited susceptible to moderately susceptible reaction to all differentials, of sugarcane. It conforms that *Fusarium moniliforme* was composed of 6 different pathtypes or races (Fm111, Fm114, Fm118, Fm1112, Fm1116 and Fm1120). It is clear from the data presented in (Table 4) that as for as seven recess have already been reported across the world of which 1, 2, 3 and 4 races were identified in Uttar Pradesh (Patil, 2002; Ali and Sonar, 1984). Haware and Nene (1979; 1982) remaining 0 and 5 from Florida (1) and 6 from Florida (Bourne (1961)). The reaction of these recess were compared with presently identified pathotypes on eight differentials, pathotypes 1 was found to be different from other identified recess of *Fusarium moniliforme* in their reaction on differentials host. Pathotypes 3 and 5 posses similar reaction and coincided with race 1 Gorakhpur) and 5, whereas pathotypes 4 was similar to race 3 (Lakhimpur Kheri). Similarly pathotypes 2 was found to be similar in reaction with race 6 of Florida (Bourne (1961) while pathotypes 6 showed very much similar reaction to race 2 (Meerut) on various differentials.

Therefore, it is apparent from the present study that 6 different recess are prevalent in Uttar Pradesh of which pathotype 2 was similar to race 6 of Florida (Bourne (1961)) and is reported for first time from Uttar Pradesh in India, which existing Gorakhpur and Lakhimpur Kheri district. It is interesting to note that none of the pathotypes exhibition similar reaction to race 4 of Lucknow and Gorakhpur. Similarly pathotypes 1 was identified for the first time from Uttar Pradesh and this pathotype seems to be more virulent and potential.

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Appendix

Table 1: Growth rate of twenty four isolates of *Fusarium moniliforme* on PDA

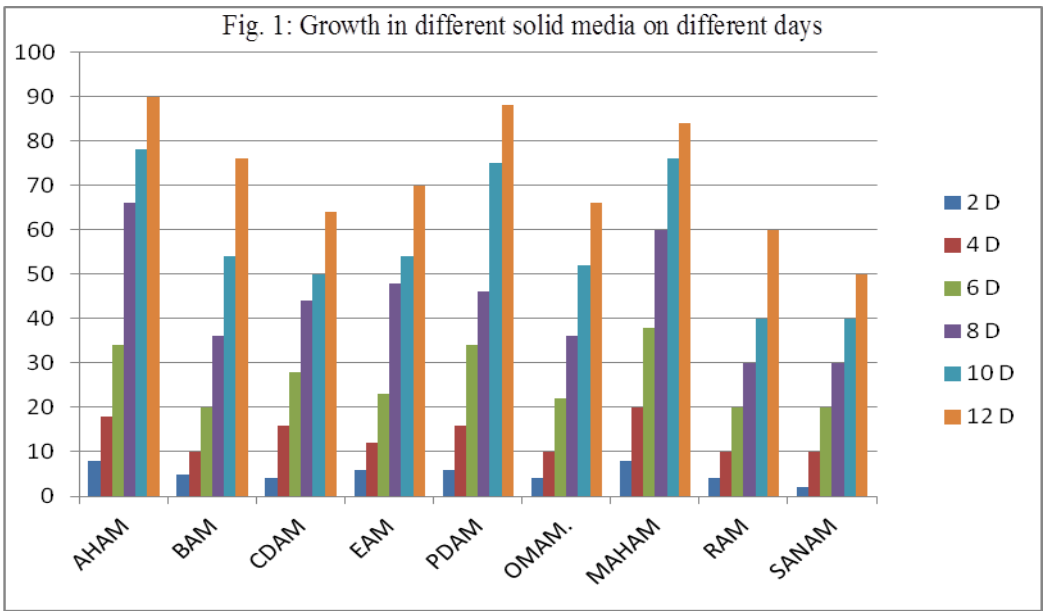
Source of isolates	Incidence %	Isolates No.	Radial growth in mm.					
			2 days	4 days	6 days	8 days	10 days	12 days
CoSe01424	16	Fm111	6	20	40	70	80	90
CoS8436	24	Fm112	2	10	36	56	76	82
CoS98259	22.2	Fm113	5	16	24	45	70	86
CoLk8102	8.8	Fm114	4	20	40	70	80	90
CoS8432	3.8	Fm115	2	14	26	36	55	86
CoSe01424	1.8	Fm116	4	16	30	50	66	80
CoJ64	14	Fm117	2	16	26	36	65	86
CoS8436	10	Fm118	8	18	40	70	80	90
CoS98259	30	Fmf119	5	12	30	42	56	76
CoS8432	26	Fm1110	2	14	32	40	70	80
CoSe01424	20.4	Fm1111	4	16	30	46	68	82
CoLk8102	22	Fm1112	6	20	40	70	80	90
CoS8436	25	Fm1113	5	13	30	50	72	85
CoJ64	14	Fm1114	2	10	28	70	80	-
CoSe01424	18	Fm1115	4	14	29	36	76	80
CoS8432	21	Fm1116	6	21	40	70	80	90
CoLk8102	2	Fm1117	2	18	34	39	76	-
CoS8436	5.6	Fm1118	6	14	38	56	67	80
CoS98259	2.8	Fm1119	4	15	32	56	69	84
CoSe01424	16.2	Fm1120	8	20	40	70	80	90
CoS8432	10	Fm1121	6	16	30	48	68	86
CoLk8102	8	Fm1122	4	12	34	54	76	84
CoJ64	6.4	Fm1123	4	18	34	50	60	86
CoS98259	1.4	Fm1124	2	10	26	38	58	76

Table 2: Morphological observations of six isolates of *Fusarium moniliforme* Sholden

S.N.	Colony characters	Macro conidia		Micro conidia	Isolates included
		Size µ	Septation	Size µ	
1	Mycelium fluffy to partially submerged, fast growing, light buff to pale cream, reverse light to yellowish brown macro conidia abundant, micro conidia moderate	16.64-46.66 × 3.36-6.66	2-5	4.66-9.98 × 2.49-6.66	Fm111
2	Aerial mycelium, pinkish white in colour, branched, hyaline to greenish septate, sometimes very distinct septa.	16.64-36.68 × 3.36-6.66	3-6	4.66-9.99 × 1.66-4.99	Fm114
3	Mycelium fluffy to partially submerged light buff to pale cream, reverse light to yellowish brown macro conidia produced more than micro conidia	16.65-36.68 × 3.36-6.66	3-6	4.66-9.99 × 2.49-4.99	Fm118
4	Aerial mycelium, rosy pink pigmentation, hyphae hyaline, septate branched, septa not very clear, macro conidia less than micro conidia	16.64-39.26 × 3.36-6.66	2-5	4.99-9.99 × 1.66-4.98	Fm 1112
5	Submerged slightly pale, whitish pigmentation, hyaline hyphae branched septate, septa not very distinct, brown macro conidia produce micro conidia Aerial mycelium micro conidia in the form of the lang chains greenish septate, macro conidia produce micro conidia	16.64-36.68 × 3.36-4.99	3-6	4.99-9.99 × 2.49-4.99	Fm 1116
6		20.98-46.64 × 3.36-6.66	3-6	4.66-9.99 × 1.66-4.99	Fm 1120

Table 3: Reactions of six isolates of *Fusarium moniliforme* on standard differentials of sugarcane

Group of isolates	CO1148	CO7717	COJ64	CO997	COC671	SES 594	Khakai	COS8436
Fm111	S	S	S	S	M	R	M	S
Fm114	S	R	R	R	R	R	R	R
Fm118	M	R	R	S	R	R	M	S
Fm1112	S	R	S	M	S	R	S	M
Fm1116	M	R	R	M	R	R	M	M
Fm1120	S	M	S	S	R	R	S	S



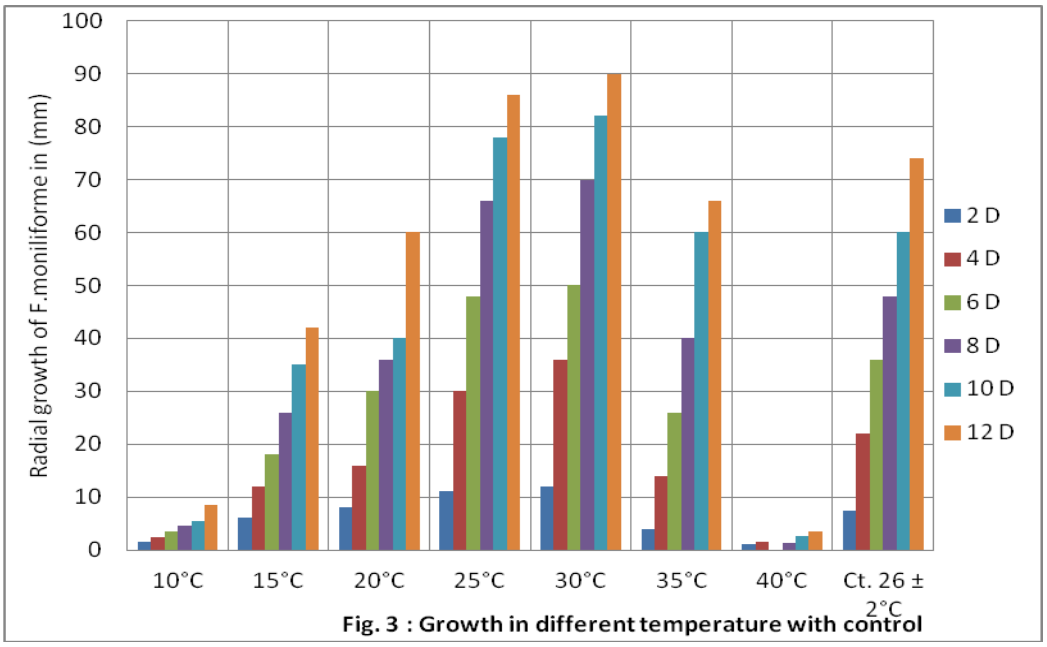


Fig. 3 : Growth in different temperature with control

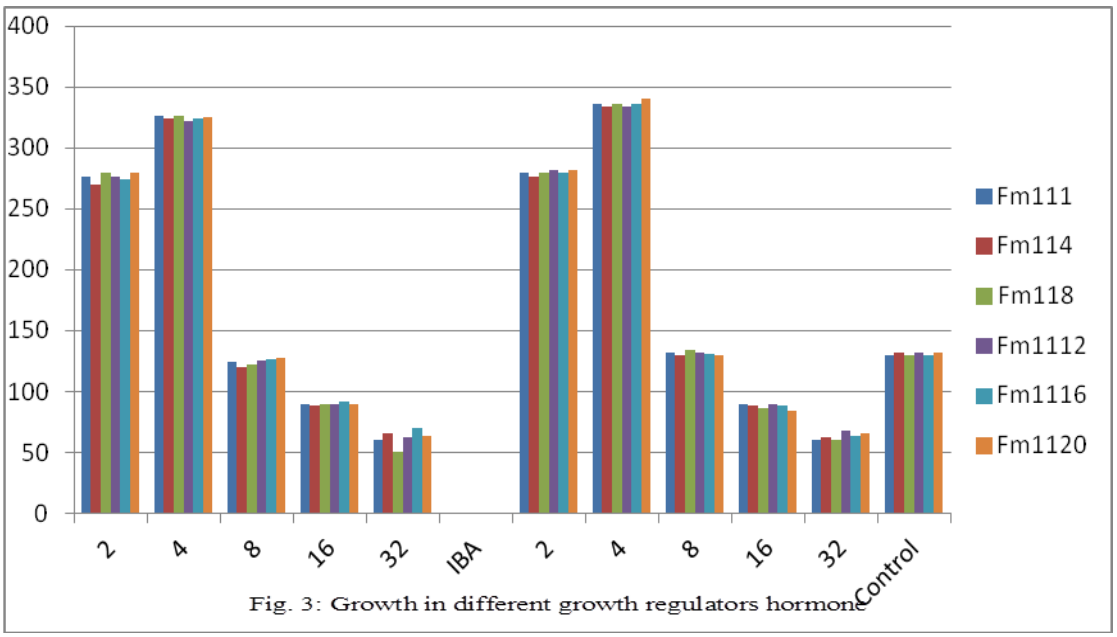
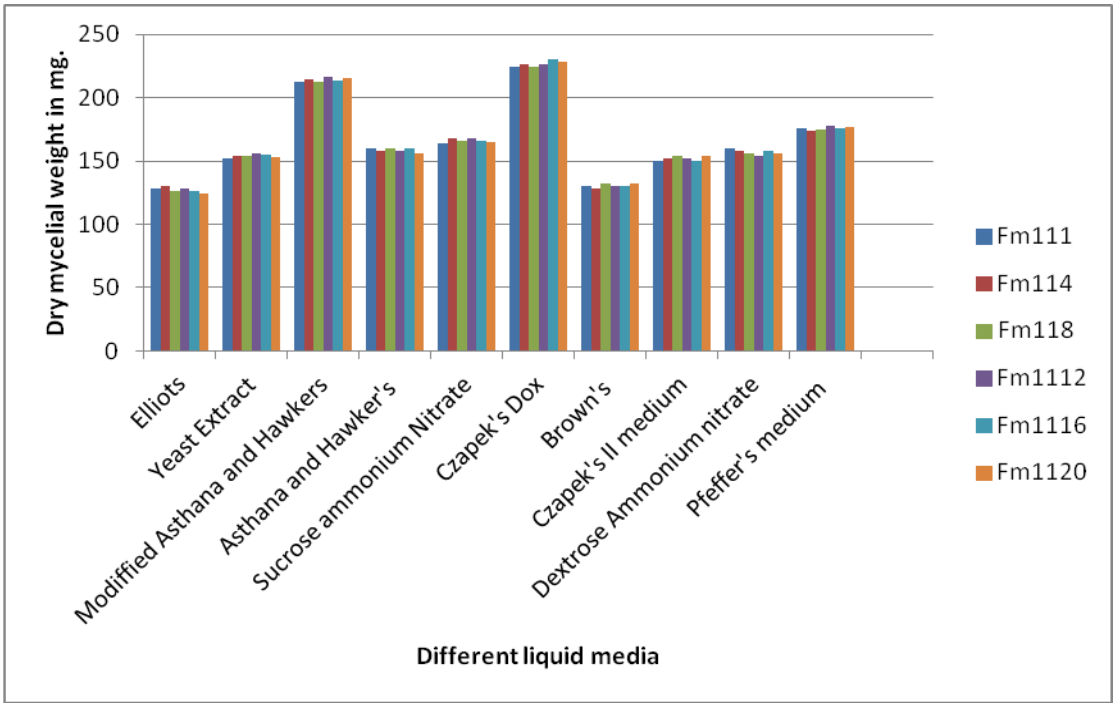


Fig. 3: Growth in different growth regulators hormone


Dry mycelial weight in mg.
250
200
150
100
50
0
Elliots
Yeast Extract
Modiffied Asthana and Hawkers
Asthana and Hawker's
Sucrose ammonium Nitrate
Czapek's Dox
Brown's
Czapek's II medium
Dextrose Ammonium nitrate
Pfeffer's medium
Different liquid media
Fm111
Fm114
Fm118
Fm1112
Fm1116
Fm1120