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Studies on cultural characteristics of *Fusarium* isolates causing panama wilt of banana under West Bengal

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Abstract

Banana wilt caused by *Fusarium oxysporum* f.sp. *cubense* is a serious disease of the crop & causes economic crop loss in every year particularly in some susceptible commercial cultivars. 14 isolates of the pathogen were collected from different banana cultivars in West Bengal. During invitro culture PDA was recorded to be the best medium for all the isolates. Great variation was recorded not only in the relative growth but also rate of growth and pigmentation in the culture medium. Maximum radial growth (90 mm) was measured in *FOC-3* & *FOC-10* in PDA medium & *FOC-4* in CDA medium after 9 days of inoculation. The slowest grower (75mm) isolates identified as *FOC-5* in PDA medium & *FOC-1* (35mm), *FOC-6* (37mm) & *FOC-7* (26.9mm) in CDA medium & variation among the isolates tested are statistically significant. Isolates were showed specificity with their nutritional requirement. All the tested pathogen prefers asparagin as a source of protein during artificial culture.

Spore population of all the isolates was counted and *FOC-3* showed highest population irrespective of macro-micro conidia. Macro-spore size varies from 14.38x3.13 μ to 28.57x3.39 μ and microspore size varies from 5.65x2.05 μ to 9.49x2.60 μ .

All the fourteen isolates were cultured & measured their relative growth using Czapekdox Agar medium. Different nitrogen sources (KNO₃, NH₄Cl, Asparagin, and NaNO₃) were used in the medium to know the most suitable compound for their utilization during artificial culture.

Keywords: banana, causing panama, characteristics, West Bengal

Introduction

Banana and plantain (*Musa* spp Fam-Musaceae) are among the most important crops in tropical and subtropical countries. The world production during 2011-12 was 30.5 million tons of banana, however, export reduction of 46% from Philippines and India mainly due to this disease and unfavourable weather factor (FAO report, 2015-16). Among the maladies the panama wilt caused by *Fusarium oxysporum* f.sp *cubense* is the most destructive disease of banana. In India the panama wilt is the main constrain for commercial production and in some areas severity may goes up to 80-90% with susceptible varieties (Mustafa & Thangavallu, 2011) [3] disease.

Although the disease was recorded only in the susceptible host with race 1 and race 2 of *Foc*. Great variation among the isolates of *Foc* has been established but the information about the characteristics of isolates of different locality in India is meager. To assess the variation among the isolates having spatial differences the present investigation was done to evaluate cultural characteristics of *Fusarium oxysporum* f sp *cubense* isolates including their sporulating behaviour and nutritional requirement among the tested isolates in artificial culture.

Materials and Method

Infected corms of banana plants were collected from different districts of west Bengal Infected rhizome of the banana plant collected from different location of West Bengal. The infected rhizomes are surface sterilized with 0.1% HgCl₂ solution. Then the rhizomes are aseptically transferred to sterilized petriplates with sterilized blotting paper. After 5-7 days a white cottony growth will come out from the surface of the rhizomes. The cottony mycelia growth of the pathogen was transferred to PDA slant with the help of inoculating needle for further use. Evaluation of growth rate and colony morphology was done by directly inoculating the culture of fungal isolates to the solid medium in Petri plates. The radial diameter of the fungal growth was taken at every 24 hrs. After full growth of the fungi on the Petri plates the colony morphology of different fungal isolates were studied and recorded accordingly. For determination of spore population both macro and micro, spore suspension of each of the fungal isolate was made. Serial dilution of the original spore suspension was prepared to make

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10^3 to 10^7 concentrations to make easier for counting of spores within the microscopic field. By counting the number of spores in 1.0 ml of suspension using the haemocytometer the total spore population per ml of suspension was calculated. All the fourteen isolates were cultured & measured their relative growth using Czapekdox Agar medium of different nitrogen sources (KNO_3 , NH_4Cl , Asparagin, and NaNO_3) were used in the medium to know the most suitable compound for their utilization during artificial culture.

Result and Discussion

A detail symptomatological study on rhizome infecting disease of banana done from different banana cultivars grown under the experimental field of AICRP on tropical Fruits (Mohanpur centre), B.C.K.V., Nadia, West Bengal and also in its vicinity. More than 130 cultivars have been conserved in the banana research centre having different genomic groups (AA, AB, BB, AAA, AAB, ABB, AAAA, AAAB, and AABB). Selected cultivars of these genomic groups were considered under this investigation an infected plant first shows a characteristics yellowing of lower, or outer, leaf-blades. It is in sharp contrast to the normal dark-green leaf colour and, accordingly affected leaves stand out conspicuously. (Table No-1)

Cultural characterization of *Fusarium* isolates grown in different laboratory media

Great morphological variations were recorded among the fourteen *Foc* isolates when grown in two different culture media by changing their nitrogen sources. In most cases isolates favour PDA medium for their growth and development *in vitro*. Most of the isolates produce light to dark, pink or violet pigments in the culture medium. Isolates were also recorded as highly variable in respect of their nature of growth and biomass production.

Similar type of work with *Foc* isolates was done by Groenewald, (2006) [2]. He recorded 3 distinct types of *Foc* isolates on the basis of their nature of growth and pigmentation. Among the three groups some are produced cottony mycelia growth with pink pigmentation with aerial mycelia production, the other few having scanty aerial mycelia with dark pink colouration, while the 3rd group produced purple pigment with scanty aerial mycelia mostly confined in the periphery of the colony. (Table No-2)

Growth estimation of *Foc* isolates cultured in PDA medium

For comparative growth assessment the *Foc* isolates were cultured in the laboratory using PDA medium. Significant variation in cultural growth was recorded among the 14 isolates tested. The maximum radial growth was measured in *Foc*-5 at 9 DAI. Initial growth of the isolates (at 3 DAI). Maximum growth was recorded in *Foc*-3 & minimum was in *Foc*-5. The *Foc*-10 having the maximum radial growth (90.00mm) at 9 DAI but initially the growth was comparatively less (42.50 mm). Thus it indicates, that rate of growth is also variable among the isolates. (Table No-3)

Determination of radial growth variation of *Foc* isolates in different solid media

Maximum growth (90.00 mm) was measured in PDA at 9 DAI in case of *Foc*-3 & *Foc*-10. Whereas the same isolates (*Foc*-3 & *Foc*-10) radial growth was measured only 43.50 mm & 60.40 mm respectively in CDA medium.

Among the isolates much variation on radial growth was recorded, & the result is statistically significant. The highest growth was recorded (at 9 DAI) in case of *Foc*-3, *Foc*-4, & *Foc*-10 irrespective of growing media. The lowest growth was recorded in *Foc*-1 (35.00 mm) in CDA medium. Rate of growth also varied among the isolates. In case of *Foc*-10 the initial growth was 16.00 mm (at 3 DAI) which grows 70.50 mm at 6 DAI & 90 mm at 9 DAI. On the other hand 44.00 mm growth was measured in case of *Foc*-2 (at 3 DAI) but the final growth (at 9 DAI) was only 80.00mm in PDA medium.

The average growth of 14 isolates of *Foc* in PDA, and CDA are 85.14 cm, and 56.57 cm respectively at 9 Days after inoculation. Maximum radial growth was obtained from *Foc*-3 & *Foc*-10 (90.00 cm) in PDA medium at 9 Days after In CDA medium the highest growth was recorded in *Foc*-4 (90.00 cm). The *Foc*-7 only shows poor growth (26.90 cm) in CDA medium as compare to PDA media

From the above findings it can be concluded that pathogen has a choice for its growth and development. Between the two common laboratory media PDA is favoured by the fungus isolate in general *i. e.* synthetic medium does not favour by the pathogen when compared with natural medium or semi-synthetic

Again within the isolates there is a great variation on radial growth indicates specificity of the isolates on their nutritional requirement. The result of this investigation is similar to Groenewald, (2006) [2], who also reported the variation of colony growth among the *Foc* isolates in artificial culture. According to him radial growth in most of the isolates ranges from 75-90 mm in PDA medium. However only seven isolates grow very faster and their average diameter was above 50 mm after 5 days of incubation at 25 °C temperatures and 12 hr photoperiod.

Estimation of radial growth of *Foc* isolates in CDA medium using different nitrogen sources

All the fourteen isolates were cultured & measured their relative growth using Czapekdox Agar medium. Different nitrogen sources (KNO_3 , NH_4Cl , Asparagin, and NaNO_3) were used (fig-2, 3, 4) in the medium to know the most suitable compound for their utilization during artificial culture.

In initial reading at 3 days after inoculation it is recorded that among the 4 nitrogen source KNO_3 supports maximum radial growth (35.63mm) of the isolates, although within the isolates there was a great variation in growth. Minimum radial growth (15.26 mm) was obtained in case of Asparagin at 3 days after inoculation.

Foc-4 was measured 36.12 mm growth which was maximum among the isolates at 3 days after inoculation irrespective of nitrogen sources.

Growth data recorded at 6 days after inoculation indicated that Asparagin was the best supporting nitrogen source among the 4 compound tested, where maximum radial growth obtained was 78.00 mm in *Foc*-14 isolates. NH_4Cl was the least supporting compound among the nitrogen sources where the average radial growth of the isolate is only 27.46 mm.

When the mean growth is consider irrespective of the nitrogen compound *Foc*-4 was measured 63.25 mm & was the highest radial growth among the isolates. *Foc*-11 is still slow growing & was measured 33.05 mm.

During final growth observation at 9 days after inoculation it was recorded that Asparagin is the most suitable source of nitrogen in artificial culture for all the *Foc* isolates tested *i.e.*

maximum radial growth was measured for all the fourteen isolates with Asparagin among the 4 compounds.

The average diameter of radial growth of *Fusarium* isolates was maximum in asparagin (86.93 mm). KNO_3 which was best supporting compound initially 3 days after inoculation was also showed good source of nitrogen (68.93 mm). NH_4Cl is not a suitable source of nitrogen for growth & development of *Fusarium* & the average radial diameter was measured is only 34.57 mm.

Foc-14 was observed (76.00 mm) the first growing among the isolates. Foc-6 was observed as slow grower & the growth measured was only 49.69 mm at 9 days after inoculation.

From this experiment it is clear that there is a great variation in growth for using different nitrogen sources. Much variation was also recorded among the isolates irrespective of compounds tested. One interesting point is to be noted that asparagin as a source of nitrogen was not suitable at initial stage & the fungal growth was minimum as compare to all other nitrogenous compound tested. However at 6 days after

inoculation maximum cultural growth was observed with Asparagin irrespective of all the fourteen isolates was highest (67.08 mm) which is much higher than other compound.

The same trend continues up to 9 days after inoculation with asparagin & maximum growth was recorded for all the isolates. It can be seen from the table: 7 the highest growth was measured in Asparagin containing medium for all the isolates & the mean is 86.93 mm which was much higher not only from NH_4Cl (34.57 mm) but also for $NaNO_3$ (56.57 mm) & KNO_3 (68.93 mm).

From the above experiment on the nutrient source of micro-organism it was recorded that there is choice on source of nitrogen for its growth and development in artificial culture. Out of the four chemicals tested against *Foc*, growth of the isolates was maximum on medium having asparagin as a source of nitrogen.

Thus it can be concluded that Asparagin is the most suitable nitrogen source for *Fusarium oxysporum* f sp *cubense* isolates as it supports for best growth & development of the pathogen.

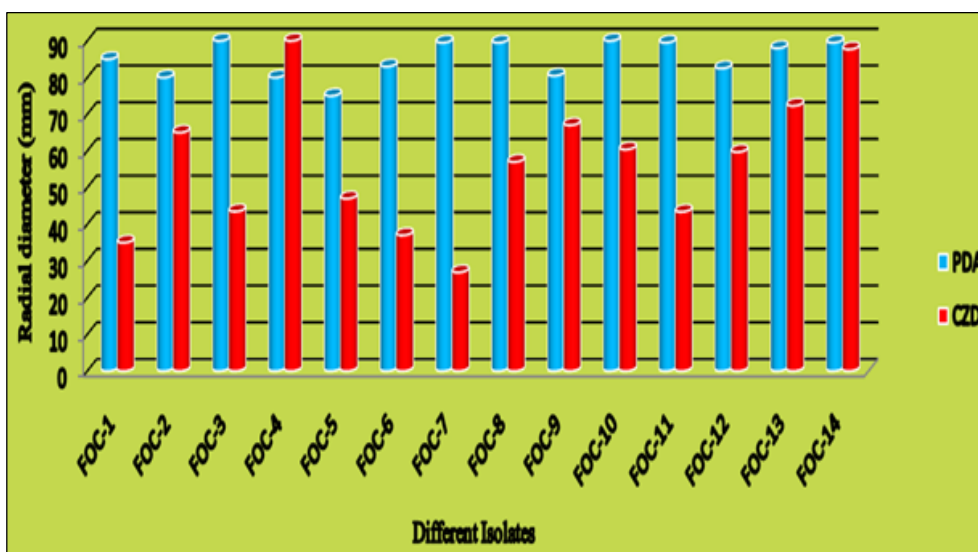


Fig 1: Radial growth variation of *Fusarium* sp at 9 DAI

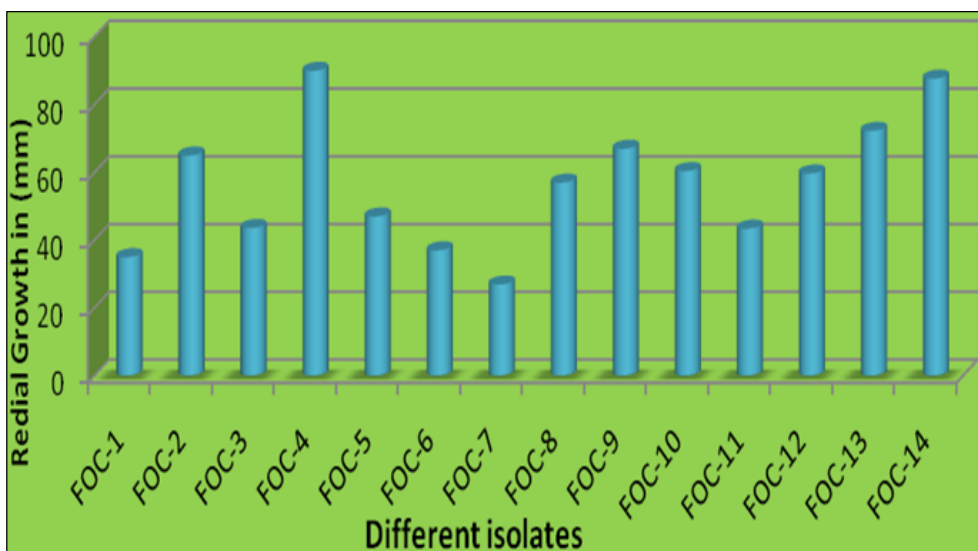


Fig 2: Growth rate of different *Fusarium* isolates in Czapekdox Agar medium containing KNO_3 instead of $NaNO_3$ at 9 DAI

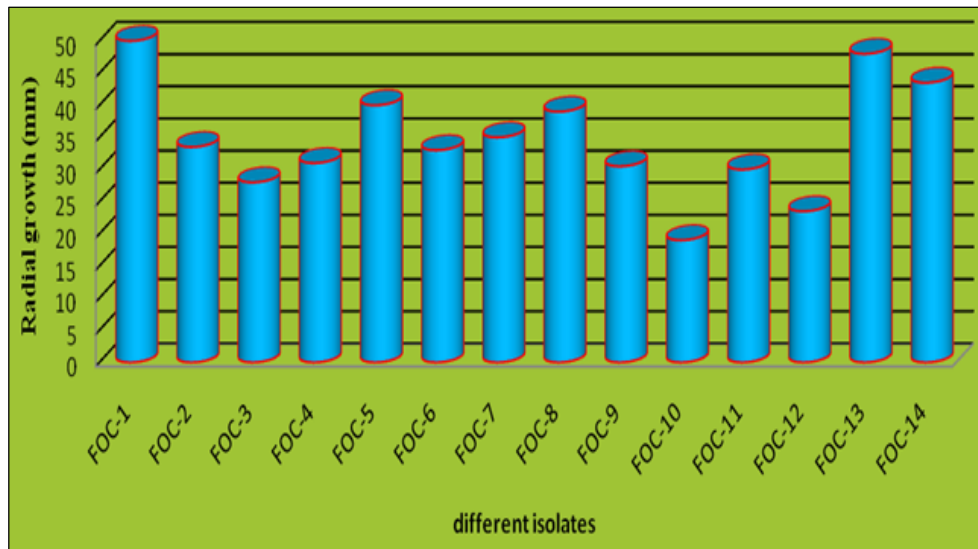


Fig 3: Growth rate of different *Fusarium* isolates in Czapexdiox Agar medium containing NH₄Cl instead of NaNO₃ at 9 DAI

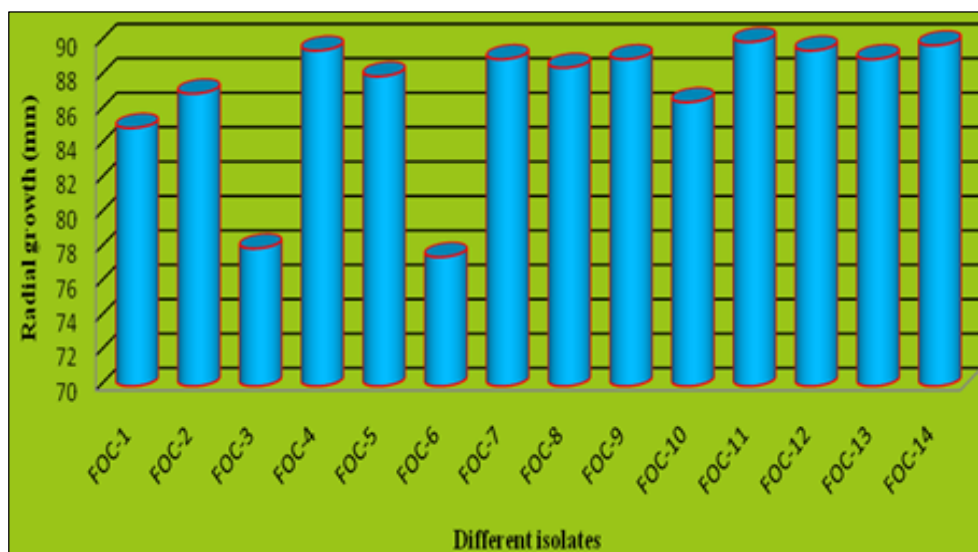
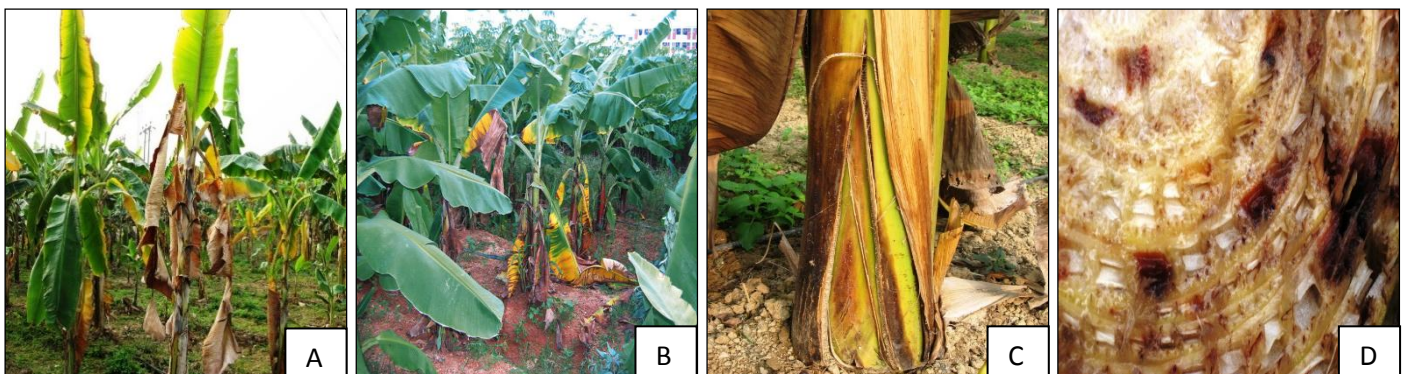


Fig 4: Growth rate of different *Fusarium* isolates in Czapexdiox Agar medium containing Asparagin instead of NaNO₃ at 9 DAI

Estimation of spore population and measurement of spore size of different *Fusarium* isolates

Sporulation studies of *Foc* isolates were done in the laboratory after growing in PDA medium. Significant variations were recorded among the isolates both in respect of spore production and size of individual spore. Ratios of macro and micro conidia are also variable among the isolates tested. (Table-8 &). Most of the isolates prefer PDA medium for their spore production in artificial culture. However, only *Foc-2* favours CDA medium for its sporulation. Maximum sporulation was counted from *Foc-3* (103.33 lakh/ml) in PDA

medium. Both macro and micro conidia of each isolate was measured after slide culture using PDA medium. No. of macro conidia as well as their size (Plate-6 & 7) were also highly variable among the *Foc* isolates. Maximum average size of macro conidia (28.57 μ x 3.39 μ) was measured in *Foc-5* which is almost similar to macro conidia of *Foc-13* (28.27 μ x 3.48 μ) while the *Foc-8* showed minimum length of macro conidia. Again the widest macro conidia was measured in *Foc-4* (3.72 μ) & minimum was recorded in case of *Foc-6*.



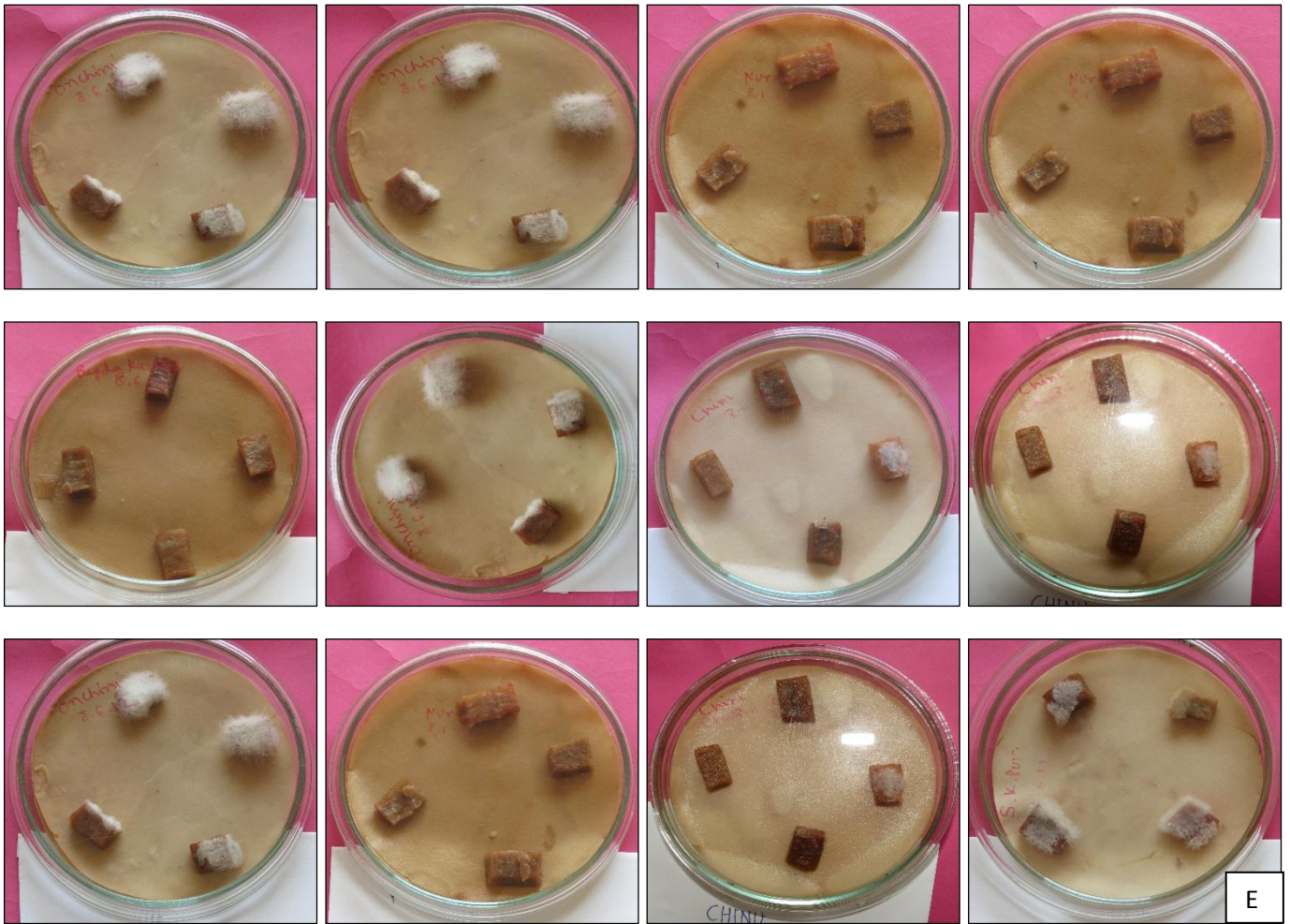


Plate 1: (A). Panama wilt symptom of banana, (B) Yellowing of leaves with petiole bucking, (C). Splitting of pseudostem, (D). Discolouration of vascular bundle in peduncle (E). Isolation of rhizome infecting fungi.

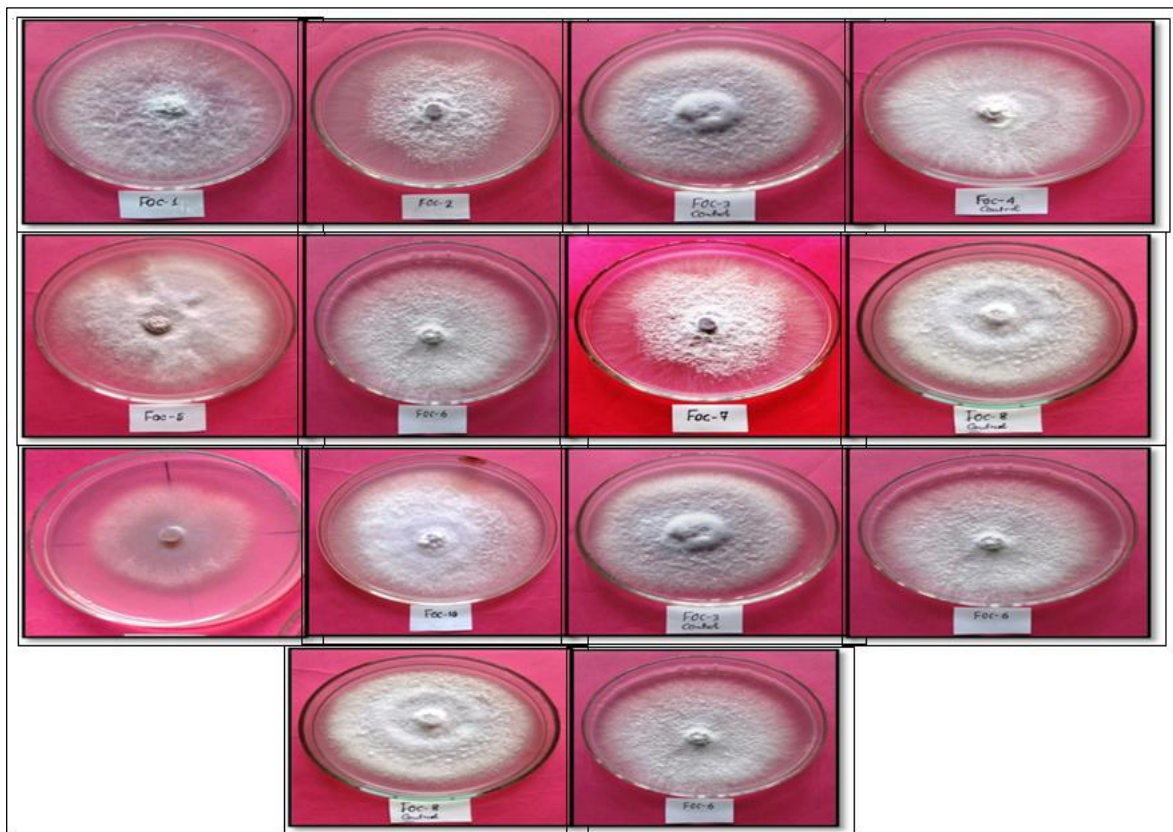
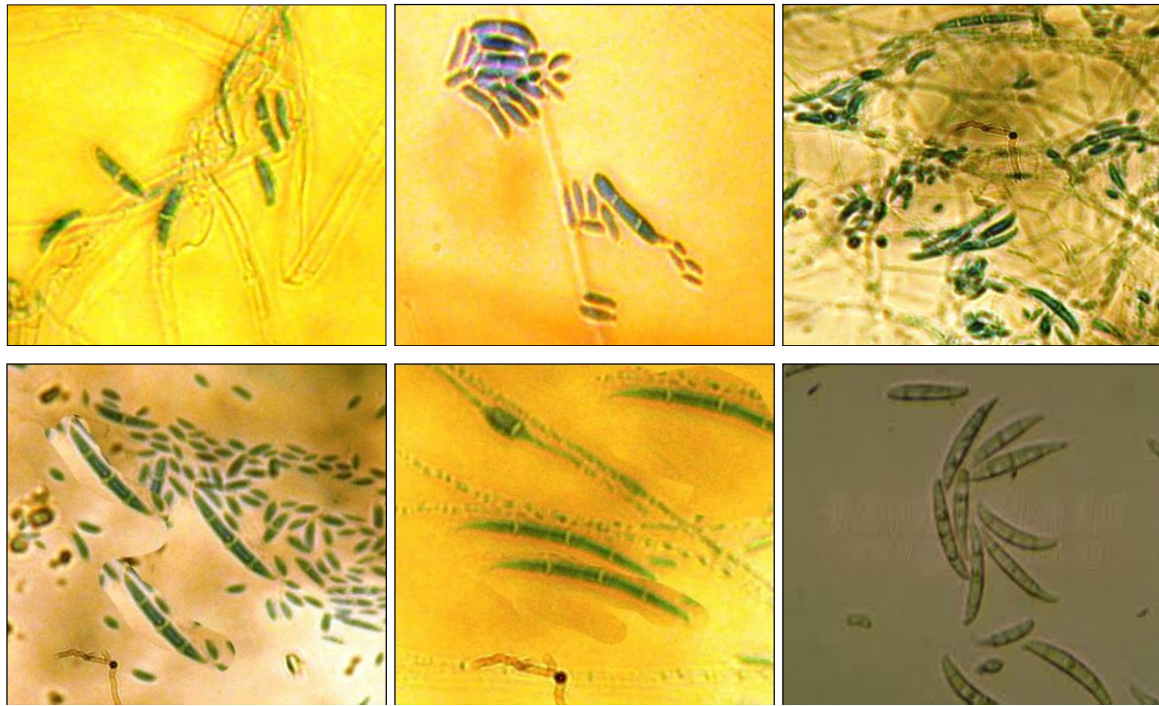


Plate 2: Fusarium isolate growing in PDA medium

Microscopic photograph of macro & micro conidial of different *Fusarium* isolatesTable 1: Collection of *Foc* isolates from different areas of West Bengal

| Serial no. | Isolates | Date of Collection | Place of collection | Banana varieties (genomic group) |
|------------|----------|--------------------|---|----------------------------------|
| 1. | FOC-1 | 10.03.12 | University farm (AICRP on tropical fruits), Nadia | Kanthali (ABB) |
| 2. | FOC-2 | 20.03.12 | Hooghly | Kanthali (ABB) |
| 3. | FOC-3 | 30.03.12 | Jaguli, Nadia | Cooking (ABB) |
| 4. | FOC-4 | 30.04.12 | Santipur, Nadia | Hanumanjata (ABB) |
| 5. | FOC-5 | 11.05.12 | Ranaghat, Nadia | Kanthali (ABB) |
| 6. | FOC-6 | 11.06.12 | Kampa, North-24-pgs | Kanthali (ABB) |
| 7. | FOC-7 | 11.06.12 | Dubrajpur, BIRbhum | Kanthali (ABB) |
| 8. | FOC-8 | 16.06.12 | University farm for AICRP on tropical fruits | Chinua (ABB) |
| 9. | FOC-9 | 16.06.12 | University farm for AICRP on tropical fruits | Rasthali (AAB) |
| 10. | FOC-10 | 04.07.12 | University farm for AICRP on tropical fruits | Martaman (ABB) |
| 11. | FOC-11 | 12.07.12 | Govindapur, Nadia | Champa-IV (AAB) |
| 12. | FOC-12 | 12.07.12 | Srikrishnapur, North 24 pgs. | Monohar-I (AAB) |
| 13. | FOC-13 | 12.08.12 | Nurpur, Nadia | Karpuravelli (ABB) |
| 14. | FOC-14 | 12.08.12 | Govindapur, Nadia | Chinua (ABB) |

Table 2: Cultural characterization of *Foc* isolates grown in different laboratory media

| Isolates | Colony characteristics (PDA) | Colony characteristics (CDA) |
|----------|--|---|
| Foc-1 | Very vigorous growth, very fluppy, huge mass, violet colour pigmentation | Thin growth, no fluppiness, less mycelia growth |
| Foc-2 | Thin but vigorous growth, light pink pigmentation | Not so fluppy but growth is moderate |
| Foc-3 | Highly vigorous, more fluppy, dark pink pigmentation. | Vigorous but thin growth, little mass |
| Foc-4 | Whitish, mat like, little biomass | Very thin, little mass |
| Foc-5 | Thin growth, less mass, no fluppiness, violet pigmentation | Thin growth, no fluppiness, growth faster than PDA |
| Foc-6 | No fluppiness, thin growth | Very thin growth |
| Foc-7 | Thin but vigorous growth, light pink pigmentation | Not so fluppy but the colony growth is comparatively parsed |
| Foc-8 | Fluppy vigorous growth, violet pigmentation | Fluppy and vigorous growth |
| Foc-9 | No fluppiness, little mass, dark pinkish on backside of plate | Little bit fluppy, no pigmentation |
| Foc-10 | Huge mass, extremely fluppy, violet pigmentation is prominent | Very thin, little mass no pigmentation |
| FOC-11 | Very vigorous, more fluppy, dark pink pigmentation | Thin growth, less mycelia growth |
| FOC-12 | Thin but vigorous growth, light pink pigmentation | Thin growth, fluppiness, growth faster than PDA |
| FOC-13 | Fluppy vigorous growth, | Thin growth, growth faster than PDA |
| FOC-14 | Whitish mat like, thin growth | Thin growth, no fluppiness, growth faster than PDA |

PDA: Potato Dextrose Agar Medium; CDA: Czapek's Dox Agar Medium;

Table 3: Variation on radial growth (mm) of *Foc* isolates in PDA medium

| Isolates | Day-3 | Day-6 | Day-9 |
|----------|-------|-------|-------|
| Foc-1 | 44 | 70 | 85 |
| Foc-2 | 44 | 67 | 80 |
| Foc-3 | 48.5 | 76.5 | 90 |

| | | | |
|--------|------|------|------|
| Foc-4 | 39.5 | 66.5 | 80 |
| Foc-5 | 35 | 55 | 75 |
| Foc-6 | 35.5 | 62.5 | 83 |
| Foc-7 | 45 | 71 | 89.5 |
| Foc-8 | 40.5 | 68.5 | 89.5 |
| Foc-9 | 39.5 | 63.5 | 80.5 |
| Foc-10 | 42.0 | 70.5 | 90 |
| Foc-11 | 43 | 70 | 89.5 |
| Foc-12 | 37 | 56.5 | 82.5 |
| Foc-13 | 35 | 50 | 88 |
| Foc-14 | 46.5 | 70.5 | 89.5 |

Table 4: Different Nitrogen Source of Czapek Dox Agar Medium (At 3rd, 6th & 9th days after inoculation)

| Isolates | CZD (NaNO ₃) | | | CZD (KNO ₃) | | | CZD (NH ₄ Cl) | | | CZD (asparagin) | | |
|----------|--------------------------|------|------|-------------------------|------|------|--------------------------|------|------|-----------------|------|------|
| | 3rd | 6th | 9th | 3rd | 6th | 9th | 3rd | 6th | 9th | 3rd | 6th | 9th |
| FOC-1 | 10 | 21.0 | 35.0 | 20.0 | 43.5 | 79.0 | 17.5 | 36.5 | 50.0 | 20.0 | 75.0 | 85.0 |
| FOC-2 | 30.5 | 46.0 | 65.0 | 22.3 | 40.1 | 79.0 | 18.5 | 27.0 | 33.5 | 13.0 | 69.0 | 87.0 |
| FOC-3 | 17.5 | 26.5 | 43.6 | 14.0 | 31.0 | 60.0 | 13.5 | 25.5 | 28.0 | 15.0 | 60.0 | 78.0 |
| FOC-4 | 62.5 | 89.5 | 90.0 | 51.0 | 60.0 | 83.0 | 19.0 | 28.5 | 31.0 | 12.0 | 75.0 | 89.5 |
| FOC-5 | 18.0 | 25.0 | 47.0 | 56.0 | 65.0 | 73.0 | 21.0 | 35.5 | 40.0 | 16.0 | 71.0 | 88.0 |
| FOC-6 | 17.0 | 23.0 | 37.0 | 31.0 | 43.0 | 51.0 | 16.0 | 28.0 | 33.0 | 12.0 | 53.0 | 77.5 |
| FOC-7 | 15.5 | 19.7 | 26.9 | 42.0 | 67.0 | 76.0 | 24.0 | 29.5 | 35.0 | 19.0 | 59.0 | 89.0 |
| FOC-8 | 23.0 | 42.0 | 57.0 | 47.0 | 55.0 | 65.0 | 21.5 | 22.5 | 39.0 | 21.0 | 60.0 | 88.5 |
| FOC-9 | 23.5 | 39.5 | 67.0 | 44.0 | 61.0 | 79.0 | 17.5 | 19.5 | 30.5 | 14.0 | 59.0 | 89.0 |
| FOC-10 | 16.0 | 35.5 | 60.4 | 35.0 | 55.0 | 79.0 | 12.0 | 14.0 | 19.0 | 11.5 | 61.0 | 86.5 |
| FOC-11 | 12.5 | 21.7 | 43.4 | 6.0 | 14.0 | 32.0 | 19.5 | 26.0 | 30.0 | 12.0 | 70.5 | 90.0 |
| FOC-12 | 26.5 | 43.1 | 59.7 | 42.0 | 53.0 | 80.0 | 11.0 | 15.0 | 23.5 | 13.7 | 73.6 | 89.5 |
| FOC-13 | 18.5 | 42.5 | 72.3 | 46.5 | 56.0 | 71.5 | 31.5 | 40.5 | 48 | 19.0 | 75.0 | 89.0 |
| FOC-14 | 24.0 | 35.5 | 87.7 | 42.0 | 55.0 | 83.0 | 22.5 | 36.5 | 43.5 | 15.5 | 78.0 | 89.8 |

| Nitrogen Source | SEM (±) | 3 rd Day | 6 th Day | 9 th Day |
|----------------------------|---------|---------------------|---------------------|---------------------|
| | | CD _{0.05} | 0.237 | 0.113 |
| Isolate | SEM (±) | 0.671** | 0.320** | 2.6241** |
| | | CD _{0.05} | 0.444 | 0.2121 |
| Nitrogen source × Isolates | SEM (±) | 1.2581** | .6009** | 8.50557** |
| | | CD _{0.05} | 0.8882 | .4242 |
| | | 2.5168** | 1.2021** | 9.8187** |

| Isolates | 3 DAI | | Isolates | 6 DAI | | Isolates | 9 DAI | |
|----------|-------|------|----------|-------|------|----------|-------|------|
| | PDA | CDA | | PDA | CDA | | PDA | CDA |
| Foc-1 | 44 | 10 | Foc-1 | 70 | 21 | Foc-1 | 85 | 35 |
| Foc-2 | 44 | 30.5 | Foc-2 | 67 | 46 | Foc-2 | 80 | 65 |
| Foc-3 | 48.5 | 17.5 | Foc-3 | 76.5 | 26.5 | Foc-3 | 90 | 43.5 |
| Foc-4 | 39.5 | 62.5 | Foc-4 | 66.5 | 89.5 | Foc-4 | 80 | 90 |
| Foc-5 | 35 | 18 | Foc-5 | 55 | 25 | Foc-5 | 75 | 47 |
| Foc-6 | 35.5 | 17 | Foc-6 | 62.5 | 23 | Foc-6 | 83 | 37 |
| Foc-7 | 45 | 15.5 | Foc-7 | 71 | 19.7 | Foc-7 | 89.5 | 26.9 |
| Foc-8 | 40.5 | 23 | Foc-8 | 68.5 | 42 | Foc-8 | 89.5 | 57 |
| Foc-9 | 39.5 | 23.5 | Foc-9 | 63.5 | 39.5 | Foc-9 | 80.5 | 67 |
| Foc-10 | 42.5 | 16 | Foc-10 | 70.5 | 35.5 | Foc-10 | 90 | 60.4 |
| Foc-11 | 43 | 12.5 | Foc-11 | 70 | 21.5 | Foc-11 | 89.5 | 43.4 |
| Foc-12 | 37 | 26.5 | Foc-12 | 56.5 | 43.5 | Foc-12 | 82.5 | 59.7 |
| Foc-13 | 35 | 18.5 | Foc-13 | 50 | 42.5 | Foc-13 | 88 | 72.3 |
| Foc-14 | 46.5 | 24 | Foc-14 | 70.5 | 35.5 | Foc-14 | 89.5 | 87.7 |

| Source | SEm(±) | 3 rd Day | 6 th Day | 9 th Day |
|----------------|--------|---------------------|---------------------|---------------------|
| | | CD _{0.05} | 0.9445 | 0.0711 |
| Medium | SEm(±) | 2.735** | 0.2059** | 0.2827** |
| | | CD _{0.05} | 0.4066 | 0.3059 |
| Isolate | SEm(±) | 1.17756** | 0.8861** | 1.2168** |
| | | CD _{0.05} | 0.6838 | 0.5145 |
| Isolate×Medium | SEm(±) | 1.9804** | 1.4903** | 2.0464** |
| | | CD _{0.05} | | |

Table 8: Estimation of spore population & spore size of different *Foc* isolates grown in PDA medium

| Isolates | Spore population lakh/ml | Spore size (μ) | |
|---------------|--------------------------|------------------------------------|---------------------------------------|
| | PDA | Micro conidia | Macro conidia |
| <i>Foc-1</i> | 25.54 | 6.55-11.88x1.91-3.45 = (9.21x2.68) | 17.35-35.12x2.11-3.33 = (26.33x2.72) |
| <i>Foc-2</i> | 16.28 | 4.53-9.95x1.71-3.33 = (7.24x2.52) | 17.02-30.16x2.01-3.23 = (23.59x2.62) |
| <i>Foc-3</i> | 103.33 | 6.29-10.39x1.53-3.40 = (8.34x2.46) | 15.45-32.33x2.10-3.21 = (23.89x2.65) |
| <i>Foc-4</i> | 11.67 | 5.30-7.18x1.56-3.19 = (6.24x2.37) | 16.80-39.68x2.96-4.48 = (28.24x3.72) |
| <i>Foc-5</i> | 29.17 | 6.48-9.71x1.94-3.1 = (8.09x2.52) | 13.38-43.77x2.14-4.65 = (28.57x3.39) |
| <i>Foc-6</i> | 16.67 | 7.34-10.52x1.83-3.47 = (8.93x2.65) | 14.59-35.65x2.14-3.07 = (25.12x2.60) |
| <i>Foc-7</i> | 10.00 | 4.44-9.23x1.9-2.9 = (6.83x2.40) | 16.95-31.12x2.21-3.25 = (24.03x2.73) |
| <i>Foc-8</i> | 10.42 | 6.57-10.63x1.64-3.78 = (8.6x2.71) | 10.83-17.93x2.33-3.94 = (14.38x3.13) |
| <i>Foc-9</i> | 9.17 | 6.3-10.06x1.81-3.41 = (8.18x2.61) | 12.19-20.75x2.95-3.91 = (16.47x3.43) |
| <i>Foc-10</i> | 23.33 | 3.97-7.34x1.37-2.74 = (5.65x2.05) | 13.75-30.12x2.33-2.94 = (21.93x2.63) |
| <i>Foc-11</i> | 27.46 | 6.45-12.54x1.75-3.46 = (9.49x2.60) | 17.75-29.79x2.56 x3.10 = (23.37x2.83) |
| <i>Foc-12</i> | 43.98 | 6.30-10.64x1.92-3.71 = (8.47x2.81) | 12.79-21.57x2.10-3.97 = (17.18x3.03) |
| <i>Foc-13</i> | 13.79 | 6.75-11.21x1.41-3.10 = (8.98x2.25) | 13.78-42.77x2.41-4.56 = (28.27x3.48) |
| <i>Foc-14</i> | 73.57 | 5.97-12.56x1.78-2.87 (9.26x2.32) | 16.97-33.43x2.45- 3.97 = (25.20x3.21) |

Conclusion

Above studies revealed that panama wilt of banana caused by the fungus *Fusarium oxysporum* f.sp. *cubense* is the most important disease causing severe damage to the crop. Different isolates of pathogen were associated with the formation of the disease. These isolates of the pathogen collected from different varieties exhibited potatoes dextrose agar (PDA) as the best artificial media for growth & sporulation from their growth characteristics in different media. Maximum radial growth was obtained from *Foc-3*, *Foc-10* and minimum growth *Foc-5* in PDA medium at 9 days after inoculation. In CDA medium the highest growth was recorded in *Foc-4* and lowest *Foc-1*. Production of conidia and ratio of macro and micro conidia are also significantly differed among these 14 isolates of *Foc*. Different nitrogen sources (KNO_3 , NH_4Cl , Asparagin, and NaNO_3) were used in the medium to know the most suitable compound for their utilization during artificial culture. From the above experiment on the nutrient source of micro-organism it was recorded that there is choice on source of nitrogen for its growth and development in artificial culture. Out of the four chemicals tested against *Foc*, growth of the isolates was maximum on medium having asparagin as a source of nitrogen.

Thus it can be concluded that Asparagin is the most suitable nitrogen source for *Fusarium oxysporum* f sp *cubense* isolates as it supports for best growth & development of the pathogen.

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