

CARBON AND NITROGEN REQUIREMENT OF COLLETOTRICHUM GLOEOSPORIOIDES PENZ

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Effect of various carbon and nitroged compounds on the growth and sporulation of *Colletotrichum gloeosporioides* Penz., is stated from diseased leaves of *Manihot utilissima* Pohl. (a plant of great economic value for its starchy tuberous roots), was studied in liquid cultures. Of the various carbon compounds used starch, glycerine, sucrose and maltose supported good growth of the organism. Fructose, glucose and galactose were comparatively poor supported while lactose supported least growth of the organism. The sporulation of the fungus was satisfactory on all the carbon sources used in the investigation, best being on starch. Comparatively poor sporulation was recorded on fructose and galactose. Among organic sources of nitrogen, which were better utilized by the fungus than inorganic ones, tryptophane, aspartic acid and asparagin showed good growth of fungus. Among inorganic sources potassium nitrate was the best. No growth was recorded on sodium nitrite. There was no correlation between the sporulation and growth of the fungus in relation to the source from which nitrogen was obtained.

Species of *Colletotrichum* Cda. are of wide occurrence and have been reported from every part of this country to cause various diseases like leaf-spots, anthracnoses, red-rots, die-back etc. in plants. *Colletotrichum gloeosporioides* Penz. has been isolated from diseased leaves of *Manihot utilissima* Pohl. the healthy foliage of the plant has a direct influence over the production of starch by its roots. Some physiological studies on different strains of this fungus have been done by Tandon & Verma¹ and Tandon & Chandra² but the strain isolated from *M. utilissima* Pohl. has not so far been investigated.

Carbon and nitrogen occupy the premier position amongst the essential elements required by micro-organisms. Brock³ Wolf^{4, 5}, Tandon & B. Igrami⁶, Agnihotri⁷, Sedlmayr et al⁸ and Lilly & Barnett⁹ have shown the importance of carbon and nitrogen in the nutrition of fungi. In the present investigation the effect of various carbon and nitrogen compounds on the growth and sporulation of *Colletotrichum gloeosporioides* Penz. has been studied.

M A T E R I A L A N D M E T H O D

A single spore culture of *C. gloeosporioides* Penz., isolated from diseased leaves of *M. utilissima* was prepared. Richard's solution was taken as basal medium (potassium nitrate 10.0 gm., potassium dihydrogen phosphate 5.0 gm., magnesium sulphate hydrated 2.5 gm, ferric chloride 0.01 gm., cane sugar 50 gm, distilled water 1 litre). To study the effect of different carbon and nitrogen sources, the amount of individual substance (carbon and nitrogen) in the basal medium was calculated and the substance was so adjusted as to contain the same amount of carbon or nitrogen present in the basal medium. 50 ml. of nutrient solution, fractionally sterilized by steaming for half an hour daily for three successive days in 150 ml. Erlenmeyer flasks, was used to culture the organism. The pH of the

TABLE 1

DRY WEIGHT AND SPORULATION OF *C. gloeosporioides* ON DIFFERENT CARBON SOURCES

S. No.	Carbon compounds	Dry weight (mg.)	Sporulation
1	Starch	1263	Excellent
2	Glycerine	1193	Good
3	Sucrose	985	Fair
4	Maltose	920	Good
5	D (-)—Fructose	787	Poor
6	D—Glucose	737	Good
7	D—Galactose	717	Poor
8	Lactose	690	Fair
9	Control (no carbon)	000	—
10	General mean	810.22	

Summary of dry weight results and conclusion at 5% level of P

Treatment	Highly significant
Replicates	Non significant
S.E.	12.7
C.D. at 5%	38.1

1 > 2 > 3 > 4 > 5 > 6 7 8

TABLE 2

DRY WEIGHT AND SPORULATION OF *C. gloeosporioides* ON DIFFERENT NITROGEN SOURCES

S. No.	Nitrogen compounds	Dry weight (mg.)	Sporulation
1	DL—Tryptophane	1325	Excellent
2	L—Aspartic acid	1293	Poor
3	Potassium nitrate	985	Fair
4	L—Asparagin	908	Poor
5	Sodium nitrate	888	Poor
6	Ammonium nitrate	728	Good
7	Ammonium Chloride	465	Fair
8	Sodium nitrate	000	Nil
9	Control	000	—
10	General mean	823.00	

Summary of dry weight results and conclusion at 5% level of P

Treatments	Highly significant
Replicates	Nonsignificant
S.E.	12.9
C.D. at 5%	39.1

1 > 2 > 3 > 5 > 6 > 7 > 8

medium was adjusted to 8.5 before sterilization. The fungus was allowed to grow at $25 \pm 2^\circ\text{C}$ for 15 days after which it was harvested on oven dry Whatman's filter paper No. 42. Three replicates of each were taken. Degree of sporulation was classified on the basis of visual observation (viz. excellent, good, fair or poor). The following carbon and nitrogen compounds were used for study :—

Carbon sources

Monosaccharides	— D-Glucose, D(—)-Fructose & D—Galactose.
Disaccharides	— Maltose, Lactose & Sucrose.
Polysaccharide	— Starch.
Alcohol	— Glycerine.

Nitrogen sources

Inorganic	— Ammonium chloride, Sodium nitrate, Sodium nitrite, Ammonium nitrate & Potassium nitrate.
Organic	— DL—Tryptophane, L—Aspartic acid & L—Asparagin.

OBSERVATIONS

The dry weights and sporulation of the fungus on different sources of carbon and nitrogen are recorded in Table 1 and 2.

RESULTS AND DISCUSSION

“The difference in the extent of growth of various fungi on a particular carbon compound may be attributed either to the difference in permeability of the cell wall or to the presence or absence of the specific enzyme necessary for the respiratory steps followed by that compound during its assimilation,” (Steinberg¹⁰). The present organism was able to utilize starch, glycerine, sucrose, maltose, fructose, glucose, galactose and lactose as carbon sources when they were replaced separately for sources in the basal medium. Starch gave the optimum growth of the fungus and was significantly superior to other carbon sources used in the investigation. Tandon & Verma¹ working with *C. gloeosporioides*, isolated from the diseased leave of *Prunus persica* and Cantino¹¹ working with *Blastocladia pringsheimii* also found starch to be good source of carbon. Glycerine was also a good source for the present organism. Grewal¹² also regarded it as a good source of carbon for *C. phomoides*, *C. lindemuthianum*, and *Alternaria tenuis*. Fairly good growth of the present organism was also recorded on different disaccharides except lactose. Sucrose was significantly better than maltose and lactose. Tandon & Chandra² working with *C. gloeosporioides* concluded that maltose and sucrose were favourably good supporters of growth of the fungus. Sucrose and maltose were also found to be good carbon sources for *Cercosporina ricinella*, *C. gloeosporioides* and *Curvularia penniseti* (Tandon & Chandra¹³). In the present study lactose was found to be the poorest source of carbon giving least growth of the fungus. It was found to be a poor source of carbon for *B. pringsheimii* and *Glomerella cingulata* by Cantino¹¹ and Saksena¹⁴ respectively. But Grewal¹² working with *C. phomoides*, *C. lindemuthianum* and *A. tenuis* found lactose as a good source of carbon. Sucrose and maltose were readily utilized by the present organism as it is believed that the hydrolytic enzymes such as invertase or transfructosidase in case of sucrose and maltase

in case of maltose were available in the fungus (Tandon & Bilgrami⁶). In case of lactose the suitable hydrolytic enzyme (lactase) was not produced by the fungus resulting in its poor utilization as found by Tandon & Bilgrami⁶ in case of *Phyllosticta cycadina*, *P. artocarpina* and *Pestalotia mangiferae* and Saksena¹⁴ in case of *G. cingulate*.

Fructose, glucose and galactose also supported good growth of the present fungus. These sugars were also reported to be good sources of carbon by Grewal¹², Misra & Mahmood¹⁵ for *C. capsici* Wolf⁴ for *Ustilago zaeae* and Saksena¹⁴ for *G. cingulate*. Glucose and galactose showed almost identical rate of growth in the present investigation. However, the latter proved to be unsatisfactory for all the organisms studied by Lilly & Barnett¹⁶ and Hawker¹⁷. They believed that poor growth on galactose was due to structural configuration. Srivastava¹⁸ also recorded slow growth of *A. tenuis* on galactose. He explained that the organism takes time to adapt itself to this sugar which does not occur in the plants.

Lilly & Barnett¹⁶ stated "Not all carbon sources are equally suitable for fruiting of fungi, some which are favourable for mycelial growth do not favour sporulation". This was true, in general, in the present investigation also. The present organism showed excellent sporulation on starch and good sporulation on glycerine, maltose and glucose. Sucrose and lactose also showed fair sporulation. Grewal¹² also recorded good sporulation on maltose and lactose in the fungi used by him. But he recorded poor sporulation on glycerine. Thind & Randhawa¹⁹ also found in case of *C. capsici* that *di* and *poly*-saccharides yielded maximum sporulation. Poor sporulation of the present organism was recorded on fructose and galactose.

Organic sources of nitrogen proved to be better supporters of the growth of the present fungus than inorganic ones, best being tryptophane. Aspartic acid and asparagine were fairly good sources of nitrogen. Tandon & Verma¹ and Thind & Randhawa¹⁹ found asparagine and aspartic acid supporting best sporulation of *C. gloeosporioides* and *C. capsici* respectively. Manocha²⁰ and Misra & Mahmood¹⁵ regarded asparagine better than other inorganic sources of nitrogen for the growth and sporulation of *C. falcatum* and *C. capsici* respectively.

Among inorganic sources of nitrogen used in the present investigation potassium nitrate was the best. Sodium nitrate and ammonium nitrate were fairly good supporters of growth. Ammonium chloride was comparatively poor source while sodium nitrite did not support the growth of the fungus at all. Similar results were found by Wilson²¹ with *C. capsici*. Tandon & Chandra² concluded that nitrite sources of nitrogen were better than ammonium and nitrite sources for *C. gloeosporioides* Penz.

Table 2 showed that there was no coorelation of fruiting of the fungus in relation to nitrogen sources used as all grades of sporulation were recorded with significantly good or poor growth of the fungus.

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