Role/Effect of Deproteinised Juice (DPJ) from Some Wild and Cultivated Plants on Yield of Biomass of Different Fungi.

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Abstract:

The deproteinised juice (DPJ) is also known as 'Whey' or 'Liquor' which is left after the extraction of protein from juice. It is well known that, the DPJ contains biologically active substances like sugars, amino acids and vitamins. These are essential components of the nutrient media useful for the cultivation of microorganisms. The presence of carbohydrates in this product makes it suitable for microbial biomass production. During the present investigation some wild and cultivated plant species (*Brassica juncea, Brassica napus, Chenopodium album, Goniocaulonindicum, Brassica oleracea, Celosia argentea, Vign atrilobata, Digera muricata, Tridax procumbens* and *Ocimum americanum*) have been used for the preparation of deproteinised juice (DPJ) and these DPJ has been utilised on the fungal biomass production.

Keywords:
Deproteinised Juice (DPJ), fungal growth, microorganisms etc.

Introduction:

The technique for extraction of protein from green leaves has been suggested by Pirie (1942) now becoming popular as "Green Crop Fractionation" (GCF). Piries method basically includes pulping the green material, expressing the juice and precipitating the proteins by heat. It means it involves the separation of proteins in leaf from the indigestible fibrous material with which it is associated. This method for separating protein from the leaf extract, sudden heating to 70-80^{O}C (Morrison and Pirie, 1961) is the most convenient method. Thus the process of GCF results into four fractions, namely Leaf juice (Leaf extract), Pressed crop residue (PC), Leaf protein concentrate (LPC) and Deproteinised juice (DPJ).

The DPJ is the fourth and last product of green crop fractionation process. During preparation of LPC, the LPC can be separated from remaining part of the juice i.e. deprotenised juice (DPJ), by filtration through a simple cotton or canvas cloth. The DPJ is a by-product of GCF system, which is produced in large volume. This brown colored watery juice is also known as "Whey" or "Liquor."In order to avoid local environmental bio-pollution due to the random disposal of DPJ and to make the process of GCF more economical and efficient, its proper use has to be made (Pirie, 1942).

It is well known that, the DPJ contains biologically active substances like sugars, carbohydrates, free amino acids, amides, minerals, vitamins and other water soluble components. The dry matter of the DPJ contains 40% carbohydrates and 3% nitrogen as reported by Pirie (1971). The glucose and fructose are the dominant monosaccharide present in the DPJ. It contains proximate amount of non protein nitrogen, soluble carbohydrates, calcium and potassium as suggested by Reddy (1986). DPJ can be successfully utilized as a culture medium for microbial growth such as yeast and *Aspergillus* (Shah *et al.* 1970; Deshpande and Joshi, 1969a); it can also be exploited for production of single cell protein (Reddy, 1986).

In the present investigation attempts were made to study the effect of different DPJ (*Brassica juncea, Brassica napus, Chenopodium album, Goniocaulon indicum, Brassica oleracea, Celosia argentea, Vigna trilobata, Digera muricata, Tridax procumbens* and *Ocimum americanum*) on yield of biomass of three fungi i.e. *Aspergillus niger*, *Fusarium moniliformae* and *Penicillium chrysogenum.*

Material and methods:

Preparation of potato dextrose agar (PDA) Medium

Potato (Peeled) - 200.0gm
Dextrose - 20.0gm
Distilled water - 1000ml

Extract was prepared from 200gm potatoes. Before weighing, potatoes were peeled and sliced. After weighing 200gm, they were suspended in 500ml sterilized distilled water and boiled for 10min. the boiled potatoes with water were filtered through muslin cloth.

The dextrose were mixed in a potato extract. The potato extract was made up to 1 liter by distilled water and heated slowly until gets dissolved properly. It was then dispensed in suitable conical flask and autoclaved at 121^{0}C for 15min.

ii) 2% DPJ solution: - 2% DPJ solution was used for fungal growth and it was prepared by dissolving 2gm of dry DPJ in 100ml distilled water, filtered and used.

Procedure

i) Sterilization: - 60ml of both PDA broth medium and the solution of DPJ were poured into 150ml small-mouthed conical flask. The flasks were then plugged with non-absorbent cotton and autoclaved at 15lbs pressure for 30min. Three replicates of each treatment were taken for the study.

ii) Inoculation: - The autoclaved flasks were transferred to the inoculation chamber. The stock cultures of fungi namely *Penicillium chrysogenum* (NCIM-739), *Aspergillus niger* (ATTC No.-545) and *Fusarium moniliformae*(ATCC No.-1100 (14164)) were used. The inoculation was always done in UV chamber

under aseptic condition with the help of sterilized inoculating needle. The inoculated flasks were incubated at room temperature for 7 days.

iii) Harvesting of microbial biomass: - After incubation period, the fungal biomass was harvested by filtration through whatman filter paper No.1. The mycelial biomass was dried along with the filter paper in an oven at 65 ± 5^{0}C till constant weight. The mycelial dry weight (MDW) was then recorded by subtracting the weight of filter paper.

Result and discussion:

The DPJ released during filtration of the heated juice leaving behind LPC is rich in plant nutrients which are soluble in water; these include carbohydrates, free amino acids, amides, minerals and other nutrients present in the cell. It is used as an animal nutrition and as a source of fertilizer. This fraction contain large proportion of soluble nutrients, it can also support the growth of microorganisms. In view of this, attempts were made during the present investigation for DPJ as a growth medium for cultivating fungi, as suggested by various workers (Ajaykumar and Mungikar, 1990a, 1990b, 1990c). The earlier reports suggested that the DPJ has a potential to support growth of various fungi and other microorganisms, and it can serves as an excellent medium for their growth. Thus the use of DPJ in microbial biotechnology is possible provided investigations on proper lines are under taken.

During the present study attempts were made to grow various fungi i.e. *Aspergillus niger*, *Fusarium moniliformae* and *Penicillium chrysogenum* on DPJ from ten selected plant species. These fungi were simultaneously cultivated on PDA (Potato Dextrose Agar) medium for comparison. The growth of fungi was evaluated on the basis of dry wt. of the mycelium harvested after 7 days of incubation period and the data is given in Table No. 1. The data obtained were statistically analyzed for standard deviation and analysis of variance (ANOVA) following Gomez and Gomez (1976) and Mungikar (2003).

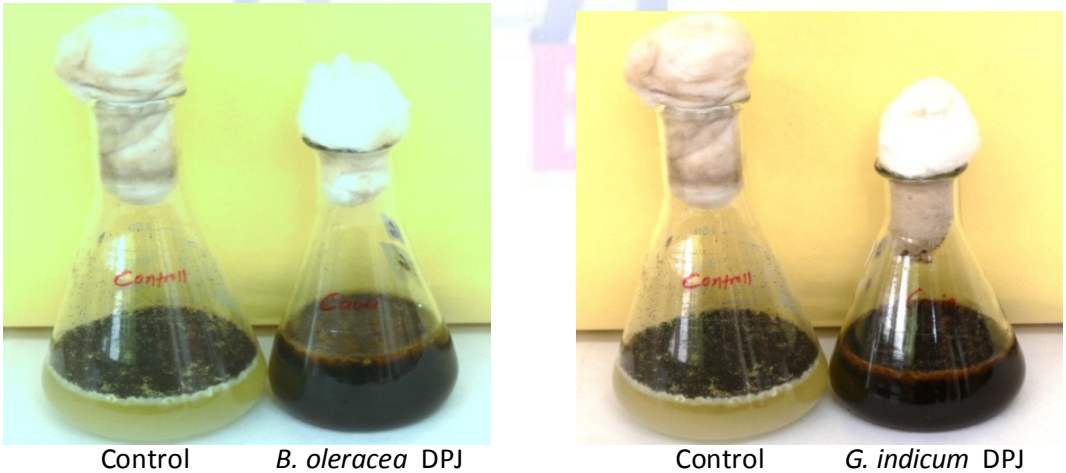
The yield of mycelial dry wt. (MDW) was 0.150g with *A. niger* when it was cultivated on PDA medium. The DPJ of *Goniocaulon indicum, Brassica oleracea, Brassica napus Brassica juncea* and *Ocimum americanum* yielded 0.270g (80%), 0.257g (71.33%), 0.200g (33.33%), 0.180g (20%) and 0.160g (6.67%) respectively were higher over the control whereas remaining DPJ did not show positive growth for *A. niger*(Plate no. 1A).

The similar trend was observed with *F. moniliformae* when it was cultivated on either PDA medium/ or on the DPJ solution. The mycelial dry wt. production (MDW) of *F. moniliformae* was 0.063g on PDA medium whereas mycelial dry wt. increased on 2% DPJ of *Chenopodium album* 0.096g (52.38%), *Tridax procumbens* 0.086g (36.51%), *Brassica napus* 0.081g (28.27%), *Brassica oleracea* 0.077g (22.22%), *Ocimum americanum* 0.069g (9.52%) and *Celosia*

argentea 0.064g (1.59%) (Plate no. 1B). This clearly indicates that, the DPJ can support the growth of fungi. Ajaykumar and Mungikar (1990a, 1990b and 1990c) as well as Gogle (2000) have also reported similar results with *Aspergillus* and *Fusarium* species. However, during the present study *P. chrysogenum* did not show any positive results. The DPJ isolated from plant species under investigation did not support to the growth of *P. chrysogenum* (Plate no. 1C). It indicates that effect of DPJ depends on source of the plant material and type of fungal species.

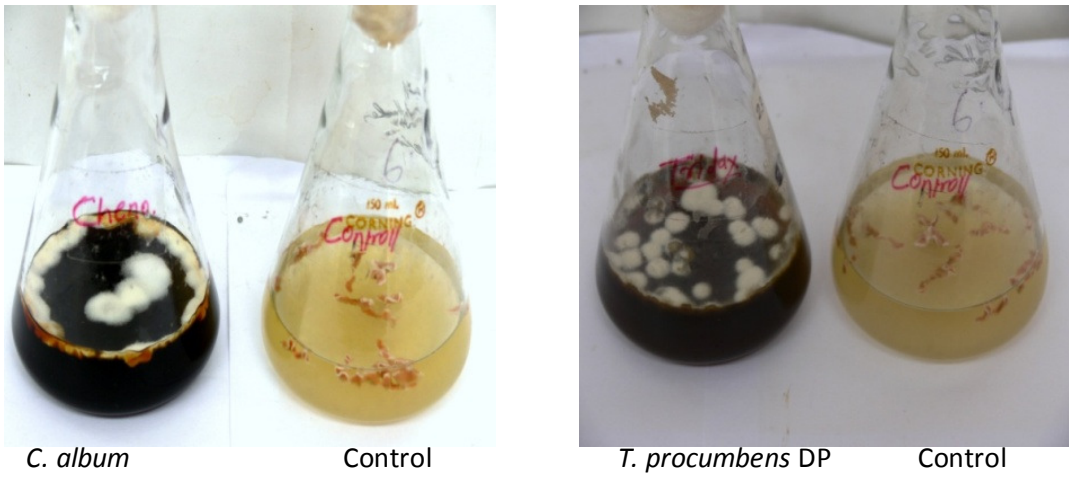
Table1: Effect of 2% DPJ of various plants on mycelial dry wt. (gm) of fungi

Name of DPJ (2%)	*Aspergillus niger*	*Fusariummonoliformae*	*Penicilliumcrysogenum*
Brassica juncea(L.) Czern. andCoss.	0.180	0.060	0.038
*Brassica napus*L.	0.200	0.081	0.032
Chenopodium album L.	0.070	0.096	0.023
Goniocaulonindicum (Klein ex willd). C.B. C.L.	0.270	0.063	0.040
*Brassicaoleraceavar. Botrytis*L.	0.257	0.077	0.050
*Celosia argentea*L.	0.060	0.064	0.007
Vignatrilobata(L.)Verde.	0.135	0.038	0.024
Digeramuricata(L.)Mart.	0.120	0.044	0.030
*Tridaxprocumbens*L.	0.127	0.086	0.028
*Ocimumamericanum*L.	0.160	0.069	0.016
Control	0.150	0.063	0.086
Mean	0.157	0.067	0.034
Critical Difference. C.D. (5%)	0.021	0.009	0.005
Critical Difference. C.D. (1%)	0.031	0.014	0.007
Coefficient of Variation. C.V. (%)	12.098	12.606	15.940



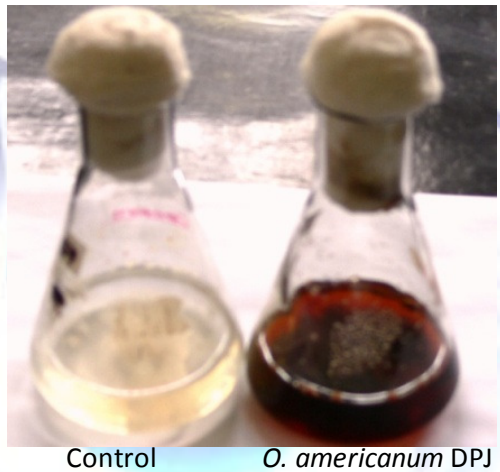
Control *B. oleracea* DPJ Control *G. indicum* DPJ

Figure. 1: Effect of 2% DPJ on *Aspergillusniger* (1-A)



C. album Control *T. procumbens* DP Control

Figure 2: Effect of 2% DPJ on *Fusariummoniliformae*(1-B)



Control *O. americanum* DPJ

Figure 2: Effect of 2% DPJ on *Penicilliumchrysogenum*(1-C)

Summary and conclusion:

During present study, the effect of different DPJ was studied on three fungi. The DPJ isolated from *Brassica juncea, Brassica napus, Goniocaulon indicum, Brassica oleracea* and *Ocimum americanum* showed positive growth for *Aspergillus niger* over the control and highest growth was observed with *Brassica oleracea* DPJ. *Fusarium moniliformae* showed positive growth from the DPJ isolated from *Brassica napus, Chenopodium album, Goniocaulon indicum, Brassica oleracea, celosia argentea, Tridax procumbens* and *Ocimum americanum* and highest with the DPJ of *Chenopodium album*. However, none of the DPJ was found to be suitable for the growth of *Penicillium chrysogenum*. From these observations it concludes that DPJ supports the growth of fungi and it can be successfully utilized for the cultivation of various microorganisms; however the suitability of the DPJ may depends on the plant species from which it was extracted.

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