Effect of DPJ Obtained from Various Preserved Leaf Juice Samples on Growth of *Rhizopus stolonifer*

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Abstract

Attempts have been made during present investigation to point out effect of the deproteinized juice (DPJ) obtained from various preserved leaf juice samples upto 48 hours on growth of *Rhizopus stolonifer* fungi. Six plant species, cauliflower, lucerne, spinach, coriander, cabbage and fenugreek were employed for experimental purpose. The deproteinized juice (DPJ), left after isolation of leaf protein concentrate (LPC) after every 04 hours was employed for the cultivation of *Rhizopus stolonifer*. The results obtained for growth of fungi on DPJ indicates that the DPJ is an ideal medium for growing fungi. The favorable growth of fungi on the DPJ expressed from the stored juice gives an indication of the availability of simple carbohydrates and nitrogenous compounds in the DPJ. The DPJ obtained from different leaf juices after 48 hours is found more suitable for the growth of fungi as a nutrient medium.

KEYWORDS: Spinach, Cauliflower, Fenugreek, Cabbage, Coriander, Lucerne, Leaf Juice, Mycelium, Rhizopus, DPJ.

Introduction:

Six plant species, cauliflower, lucerne, spinach, coriander, cabbage and fenugreek leaves were employed for extraction of leaf juice by mechanical fractionation. During mechanical fractionation as soon as the green foliage is harvested and pulped autolysis begins due to the activity of plant enzymes, which resemble the process of senescence (Singh, 1962, Batra et. al., 1976; Mungikar and Joshi, 1976). Singh (1962) observed that from 7 to 20 % of proteins was autolysed due to the incubation of leaf extracts for 2 hours at 37^0C. The autolysis process leads to catabolism which involves breakdown of complex chemical compounds including protein. As a result of this the recovery of protein decreases with the decreases in the yield of leaf protein concentrate (LPC). The leaf juice is prone to chemical change and its composition changes rapidly Nasi (1983). Enzymatically induced stability of protein in juice has been shown by several workers (Tracey, 1948; Macpherson, 1952; Brady, 1961; Batra et. al., 1976). The overall results indicate that storage of leaf juice for few hours leads to depletion of nutrient and other organic compounds. Attempts have been made during present investigation to point out the effect of deproteinized juice (DPJ) obtained from various preserved leaf juice samples upto 48 hours on growth of *Rhizopus stolonifer*. The DPJ obtained from different leaf juices upto 48 hours is employed for the growth of fungi as a nutrient medium along with the GN media.

Material and Methods:

Fresh leaves of six plant species, cauliflower (*Brassica oleracea var. botrytis*), spinach (*Spinacea oleracea* L.), coriander (*Coriandrum sativum* L.), cabbage (*Brassica*

oleracea L. var. *capitata*), Lucerne (*Medicago sativa* L.) and fenugreek (*Trigonella foenium-graceum* L.) were obtained early in the morning from local vegetable market. Fresh leaves of these plant species were immediately brought to the laboratory, washed with water to remove the adhering soil particles. The leaves were crushed and pressed to release the leaf extract. The leaf juice released during pressing was employed for the experimental purpose. The samples of juice were stored at room temperature (23 to 36 ^{o}C) in conical flask plugged with cotton and the samples of stored juice were opened after every 04 hours till 48 hours to obtain the DPJ.

One hundred ml of leaf juice from any one of these flasks was added in the beaker which contains 20 ml boiling water with stirring and heated to 95 ^{o} C for preparation of leaf protein concentrate (LPC). The LPC was then isolated from deproteinized juice by filtration through whatman filter paper. The deproteinized juice (DPJ), left after isolation of LPC was employed for the cultivation of *Rhizopus stolonifer*. For this purpose 25 ml DPJ was placed in 250 ml conical flask. The flask was plugged with non absorbent cotton and autoclaved at 15 Ibs for 15 minutes. The fresh and pure culture of *Rhizopus stolonifer* is obtained. The spore suspension is made with sterile distilled water for inoculation. The flask containing DPJ were inoculated with the spore suspension and inoculated flasks were incubated at room temperature (22 to 34 ^{o} C) for 7 days or till sporulation. After 7 days the content of the flask were filtered through whatman filter paper to isolate mycelium. Isolated mycelium was dried in oven at 60 ± 5 ^{o} C till constant weight and the mycelial dry weight (MDW) was recorded for each type of DPJ collected at every stage of storage. The data were statistically analyzed by following, Mungikar (1997).

Result Discussion:

The data on mycelial dry weight is given in table 1. The mycelial dry weight on cabbage DPJ was minimum (210 mg / 25 ml DPJ) while it was maximum on cauliflower DPJ (325 mg / 25 ml DPJ). The yield of mycelial dry weight (MDW) gradually increases with the increase in storage time. On spinach DPJ, mycelial dry weight increased from 250 to 390 mg / 25 ml of DPJ at the end of the 48 hours, similarly with the DPJ of cauliflower, fenugreek, cabbage, coriander and lucerne the increase was from 325 to 530, 266 to 410, 210 to 375, 280 to 450 and 290 to 515 mg 25 ml DPJ respectively. The mycelial dry weight of *Rhizopus stolonifer* on GN media was 84 mg / 25 ml which was far less than that obtained on DPJ. It is well known that the DPJ supports growth of microbes (Ajaykumar and Mungikar, 1990 a, 1990 b). The results obtained on growth of fungi on DPJ confirmed that the DPJ is an ideal medium for growing fungi. The favorable growth of fungi on the DPJ expressed from the stored juice gives an indication of the availability of simple carbohydrates and nitrogenous compounds in the DPJ. This confirms that during storage complex chemical compounds are disintegrated and subsequently lost in the DPJ due to that more fungal growth is found on DPJ which is obtained from different stored leaf juice.

Table 1: Effect of DPJ obtained from preserved leaf juice samples on growth of *Rhizopus stolonifer*.

Time of Storage of leaf Juice	Mycelial dry weight (MDW), (mg / 25 ml DPJ)						
	Spinach	Cauliflower	Fenugreek	Cabbage	Coriander	Lucerne	GN
00	250	325	266	210	280	290	84
04	255	340	274	317	295	305	
08	258	365	295	222	305	315	
12	262	390	308	233	318	330	
16	270	415	315	243	326	360	
20	280	432	327	255	340	395	
24	285	455	345	268	358	410	
28	292	470	358	275	370	434	
32	300	482	370	290	395	452	
36	320	495	395	310	410	475	
40	340	505	405	330	425	490	
44	365	520	408	345	438	505	
48	390	530	410	375	450	515	
Mean	297	440	344	275	362	406	
S. D.	44	69	51	52	57	79	
C. V.	15	16	15	19	16	19	

Summary and conclusion:

The overall results showed that the DPJ is a suitable and ideal medium for growth of *Rhizopus stolonifer* fungi. The favorable growth of fungi on the DPJ expressed from the stored juice gives an indication of the availability of simple carbohydrates and nitrogenous compounds in the DPJ. This confirms that during storage complex chemical compounds are disintegrated and subsequently lost in the DPJ which favorites the growth of fungi in large quantity. The overall increase in the amount of mycelium is of 15 to 19 % due to the storage of various preserved leaf juice upto 48 hours.

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