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Bioconversion of molasses to ergot alkaloids by *Claviceps purpurea* BPS-1660 exposed to Dioctadecyl dimethylammonium chloride (DODAC)

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Abstract: The influence of dioctadecyl dimethylammonium chloride (DODAC) on bioconversion of molasses to ergot alkaloids by *Claviceps purpurea BPS-1660* has been assessed. It has been found that micelle, i.e., dioctadecyl dimethylammonium chloride (DODAC) under trial has enhanced the yield of ergot alkaloids to an extent of 10.579% higher in comparison to control

(Key words: Ergot alkaloids fermentation, dioctadecyl dimethylammonium chloride (DODAC) and *Claviceps purpurea* BPS-1660).

Introduction

Micelles are widely used in industrial and biological fields for their ability to dissolve and move non polar substances through an aqueous medium, or to carry drugs which are, often, scarcely soluble in water. The carrying ability of micelles can be altered if parameters determining their size and shape are changed. However, the shape and size of the micelle are functions of the nature of the system.[1-11] It is well known that nature provides some versatile compounds without which it would have been difficult for the life to persist. Surfactants are one such group of compounds which are used in various fields of science from electronics to biology.[12-16]

Thus, from the above brief review it is evident that there is no definite opinion regarding the influence of micelles on the production of ergot alkaloids and in view of this the author has studied the influence of dioctadecyl dimethylammonium chloride (DODAC) on bioconversion of molasses to ergot alkaloids by *Claviceps purpurea* BPS-1660.

Experimental

The effect of micelle, i.e., dioctadecyl dimethylammonium chloride (DODAC) on bioconversion of molasses to ergot alkaloids by *Claviceps purpurea BPS-1660* has been studied. The experimental results have been described in table -1.

Temperature : $27^0C \pm 1^0C$
Assay method :
Evaluation of ergot alkaloids formed was made colorimetrically[17-19].
Sterilization :
At 15 lbs steam pressure for 30 min. in an autoclave
Organism used :
Claviceps purpurea BPS-1660 was used as source of enzymes for production of ergot alkaloids.
Age of inoculum : 50h
Quantum of inoculum : 10ml
Incubation periods : 6, 8 and 10 days
Optimum incubation period : 8 days
Vegetative medium[20] The composition of the vegetative medium was as follows :
Dextrose : 125.00 g; Citric acid : 15.00 g; Yeast extract : 0.15 g; KH_2PO_4: 0.60g; $MgSO_4.7H_2O$:0.35g; $FeSO_4.7H_2O$: 0.010g; $ZnSO_4.7H_2O$: 0.020g; pH : 5.4; Distil water : 1000 ml.

The pH of the medium was adjusted to

Table - 1
Bioconversion of molasses to ergot alkaloids by *Claviceps purpurea BS-1659* exposed to Dioctadecyl dimethylammonium chloride (DODAC)

Concentration of micelle a x 10^{-3} M	*Yield of Ergot -alkaloids in mg/litre			% of ergot alkaloids increase (+) in 8 days of incubation period
	6 days	8 days	10 days	
1.0 x 10^{-3} M (+) micelle	680	918	890	(+) 2.227
2.0 x 10^{-3} M (+) micelle	689	935	898	(+) 4.120
3.0 x 10^{-3} M (+) micelle	694	946	911	(+) 5.345
4.0 x 10^{-3} M (+) micelle	701	963	915	(+) 7.238
5.0 x 10^{-3} M (+) micelle	715	984	925	(+) 9.576
6.0 x 10^{-3} M** (+) micelle	679	993***	920	(+) 10.579
7.0 x 10^{-3} M (+) micelle	660	989	913	(+) 10.133
8.0 x 10^{-3} M (+) micelle	651	973	901	(+) 8.351
9.0 x 10^{-3} M (+) micelle	640	940	893	(+) 4.677
10.0 x 10^{-3} M (+) micelle	632	930	890	(+) 3.563
Control (-) micelle	670	898	880	—

* Mean of three observations. ** Optimum concentration of micelle used, *** Optimum yield of ergot alkaloids (+) values indicate % increase (+) after 8 days. Experimental deviation (±) 1.5% to 3.5%

5.4 by adding requisite amount of NH_4OH solution. Now 100 ml of the vegetative medium was taken into 250 ml conical flask. The flasks were then plugged and sterilised in an autoclave at 15 lbs steam pressure for 30 minutes.

Inoculation of the vegetative medium

The vegetative medium was inoculated by transferring 5 disc of mycelium (each of 10 mm dia) from 8days old slant.

Preparation of the production medium

The composition of the production medium employed throughout the investigation was as follows :

Sugar : 200.00 g; Citric acid : 20.00 g; Yeast extract : 0.15 g; KH_2PO_4: 0.55g; $MgSO_4.7H_2O$:0.55g; KCl : 0.15 g; $FeSO_4.7H_2O$: 0.010g; $ZnSO_4.7H_2O$: 0.008g; pH : 5.4; (Adjusted by NH_4OH solution)
Distil water to make up 1000 ml.

Now from the above composition 100 ml of the production medium was taken into 250 ml conical flask. The flasks were plugged and sterilized in an autoclave.

100 ml of vegetative medium was taken into 250 ml flasks. The flasks were plugged with non-absorbent cotton and were sterilized in an autoclave at 15 lbs steam pressure for 30 minutes and were left for cooling at room temperature. Now, two flasks were inoculated each with 5 discs (each disc of 10 mm dia). of 8 days old *Claviceps purpurea* B.S. 1659. The flasks were kept on a rotary shaker (230 rpm) at 27 ± 1^0C for 8 days. After 8 days the content of the flask was homogenised in a sterile mixer for 10 seconds. This is the vegetative stage I. For second vegetative stage the flasks were taken each containing 100 ml of the same vegetative medium. Now, each flask was inoculated with 10ml of inoculum from vegetative stage I. These flasks were put on a rotary shaker operating at 230 rpm for 50 hours. After 50 hours 10 ml of the inoculum was used to inoculate the production medium.

99-flasks, each containing 100 ml of the production medium was taken for production stage. These were arranged into 11 sets, each set consisting of 9-flasks.

Now, M/10 solution of dimethyl-ammonium chloride (DODAC) was prepared and 1.0, 2.0, 3.0, 4.0, 5.0, 6.0, 7.0, 8.0, 9.0 and 10.0 ml of this solution was added in the flasks from 1st to 10th set respectively. Remaining one set was kept as control and it contained no (DODAC). Each flask was inoculated with 10 ml of the inoculum from IInd vegetative stage. The flasks were kept on a rotary shaker operating at 230 rpm at temperature 27 ± 1^0C. Colorimetric analysis were carried out after 6, 8 and 10 days of incubation period.

Results and Discussion

The influence of dioctadecyl dimethylammonium chloride (DODAC)

The data recorded in the table - 1 shows that miceller efficacy of dioctadecyl dimethylammonium chloride (DODAC) on production of ergot alkaloids by fermentation has also been found stimulatory but not too significant. It has been found that there is a gradual and mild increase in the production of ergot alkaloids by fermentation with the stepping up of the micelle dioctadecyl dimethylammonium chloride (DODAC) till the maximum yield of ergot alkaloids, i.e., 993 mg/litre was obtained at 6.0×10^{-3}M concentration of dioctadecyl dimethylammonium chloride (DODAC) which is 10.579% higher in comparison to control fermenter flasks, i.e; 898mg/litre in 8 days of optimum incubation period. It has also been recorded that production of ergot alkaloids exposed to dioctadecyl dimethylammonium chloride (DODAC), i.e; at 1.0×10^{-3}M the yield of ergot alkaloids has been found a bit more in comparison to control fermentor flasks.

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