

Production of citric acid by *Aspergillus oryzae* NCIM-929 exposed to tris(dodecylbenzenesulfonate)isopropoxide

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Abstract: The efficacy of tris(dodecylbenzenesulfonate)isopropoxide on fermentative production of citric acid has been assessed. It has been found that tris(dodecylbenzenesulfonate)isopropoxide has stimulatory effect on fermentative production of citric acid by *Aspergillus oryzae* NCIM-929 and enhances the yield of citric acid to an extent of 8.693% higher in comparison to control fermentor flasks, i.e., 7.109 g/100 ml under the optimized conditions.

(Key words : Citric acid fermentation, tris (dodecylbenzenesulfonate)isopropoxide.)

Introduction

Micelles are approximately spherical in shape. Other phases, including shapes such as ellipsoids, cylinders, and bilayers, are also possible. The shape and size of a micelle are a function of the molecular geometry of its surfactant molecules and solution conditions such as surfactant concentration, temperature, pH, and ionic strength. The process of forming micelles is known as micellisation and forms part of the phase behaviour of many lipids according to their polymorphism¹⁻³. A number of recent review articles have been written on the rheology and applications of wormlike micelle solutions⁴⁻¹⁶. However, as the salt concentration of micelles is increased and the head group charges are sufficiently screened, the wormlike micelles can form three-point or four-point junctions. Thus, from the above brief review it is evident that micelles are required for exploitation specially for citric acid fermentation¹⁷ and in view of this the author has studied the influence of tris(dodecylbenzenesulfonate)isopropoxide. on

citric acid production by *Aspergillus oryzae* NCIM-929

Experimental

The influence of tris(dodecylbenzenesulfonate)isopropoxide on production of citric acid by *Aspergillus oryzae* NCIM-929.

The composition of citric acid production medium is prepared as follows:

Molasses : 24% (w/v) NH_4NO_3 : 0.55%
 KH_2PO_4 : 0.55% $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$: 0.55%, pH : 2.0

The pH of the production medium was adjusted to 2.0 by adding requisite amount of KCl-HCl buffer solution and this pH was also ascertained by a pH meter. The above composition medium represents volume of a fermentor flask, i.e., "100ml" production medium of citric acid by *Aspergillus oryzae* NCIM-929. Now, the same production medium for production of citric acid was prepared for 99-fermentor flask, i.e.; each contained '100ml' of production medium. The above 99-fermentor flasks were then arranged to 11-sets each comprising of 9-fermentor flasks. Each set was then rearranged in 3-subsets, each consisting of 3-fermentor flasks. The remaining 9-fermentor flasks out of 99-fermentor flasks were kept as control and these were also rearranged in 3-subsets each consisting of 3-fermentor flasks.

After preparing the above sets of fermentor flasks M/1000 solution of tris(dodecylbenzenesulfonate)isopropoxide. was prepared and from the above micelles solution 1.0, 2.0, 3.0, 4.0, 5.0, 6.0, 7.0, 8.0, 9.0 and 10 ml was

Table - 1
Production of citric acid by *Aspergillus oryzae* NCIM-929 exposed to
tris(dodecylbenzenesulfonate)isopropoxide

Concentration of micelle used $A \times 10^{-x} M$	Incubation Period in days	Yield of citric acid* in g/100 ml	Molasses* left Unfermented in g/100 ml	% of citric acid increased after 11 days
Control	11	7.109	2.893	-
$1.0 \times 10^{-5} M$	11	7.151	2.846	+0.590
$2.0 \times 10^{-5} M$	11	7.236	2.764	+1.786
$3.0 \times 10^{-5} M$	11	7.322	2.673	+2.996
$4.0 \times 10^{-5} M$	11	7.415	2.586	+4.304
$5.0 \times 10^{-5} M$	11	7.571	2.423	+6.498
$6.0 \times 10^{-5} M^{**}$	11	7.727***	2.276	+8.693
$7.0 \times 10^{-5} M$	11	7.478	2.526	+5.190
$8.0 \times 10^{-5} M$	11	7.336	2.669	+3.193
$9.0 \times 10^{-5} M$	11	7.244	2.753	+1.899
$10.0 \times 10^{-5} M$	11	7.165	2.836	+0.787

* Each value represents mean of three trials ** Optimum concentration of micelle used

*** Optimum yield of citric acid (+) values indicate % increase in the yield of citric acid after 11 days. Experimental deviation ($\pm$) 1.5-3%

added to the fermentation flasks of above 1st to 10th sets respectively. The control fermentor flasks contained no micelle.

Now, the total volume in each flat bottom fermentor flasks was made upto 100 ml by adding requisite amount of distilled water. Thus, the molar concentration of tris(dodecylbenzenesulfonate)-isopropoxide in 1st, 2nd, 3rd, 4th, 5th, 6th, 7th, 8th, 9th and 10th subsets were approximately as given below :

$A \times 10^{-x} M$ i.e.,

$1.0 \times 10^{-5} M$ to $10.0 \times 10^{-5} M$ respectively.

Where : A = amount of micelle,

in ml, i.e. 1.0 ml to 10 ml.

x = Molarity of the micellar solution

The above fermentor flasks were then sterilized, cooled inoculated, incubated at 27°C and analysed after 9, 11 and 13 days for citric acid¹⁸ formed and molasses¹⁹ left unfermented.

Results and Discussion

The Influence of tris(dodecyl benzenesulfonate) isopropoxide.

The data recorded in the table 1 shows that tris (dodecyl benzenesulfonate)isopropoxide has stimulatory and beneficial effect on production of citric acid by *Aspergillus oryzae* NCIM-929.

The maximum yield of citric acid, i.e; 7.727g/100 ml in the presence of tris (dodecylbenzenesulfonate) isopropoxide was observed at $6.0 \times 10^{-5} M$ molar concentration in 11 days of optimum incubation period which is 8.693% higher in comparison to control fermentor flasks, ie; 7.109g/100 ml in the same times course and other same experimental parameters.

The higher molar concentrations of tris (dodecylbenzenesulfonate) isopropoxide were not

much effective and favourable for the production of citric acid by *Aspergillus oryzae* NCIM-929. So the gradual addition of the micelle tris-(dodecylbenzenesulfonate)isopropoxide after certain concentrations were not effective and beneficial for the production of citric acid by *Aspergillus oryzae* NCIM-929.

It has been observed that molar concentration of the micelle, i.e., tris(dodecyl-

benzene sulfonate)isopropoxide from 1.0×10^{-5} M to 6.0×10^{-5} M enhances the yield of citric acid to a certain order being 0.590%, 1.786%, 2.996%, 4.304%, 6.498% and 8.693% higher in comparison to control flasks but at 7.0×10^{-5} M to 10.0×10^{-5} M the yield of citric acid shifted 5.190%, 3.193%, 1.899% and 0.787% higher in comparison to previous concentrations of tris (dodecylbenzenesulfonate)isopropoxide taken into experimental trials.

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