

Fungal bioproduction of citric acid exposed to lithium 2-dodecylbenzenesulfonate

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Abstract : The efficacy of lithium 2-dodecylbenzenesulfonate on fermentative production of citric acid by some fungal strains such as *Aspergillus carbonarius* NCIM -953, *Aspergillus saitoi* NCIM - 951, *Aspergillus usumii* NCIM - 925, *Aspergillus wentii* NCIM - 905 and *Aspergillus oryzae* NCIM - 929 has been assessed. It has been found that the fungal strain *Aspergillus oryzae* NCIM-929 has been found suitable to give higher yield of citric acid. The micelle lithium 2-dodecylbenzenesulfonate 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 11.597% higher in comparison to control fermentor flasks, i.e., 7.536g/100 ml under the optimized conditions.

(Key words : Citric acid fermentation, lithium 2-dodecylbenzenesulfonate)

Introduction

Surfactants and lipids, small molecules consisting of a hydrophilic head and a hydrophobic tail, self-assemble into micelles. Surfactants and lipids are usually found in aqueous solutions. When the concentration of the surfactant reaches and exceeds the critical micelle concentration, the hydrophobic tails cluster together to minimize their interactions with the solvent while the hydrophilic heads interact with the polar solvent to increase the entropy of and stabilize the system. As a result, the surfactant molecules form micelles (spherical structures with the hydrophilic heads on the outside and hydrophobic tails on the inside). Micelles are in dynamic equilibrium such that molecules are continuously joining or leaving micelles.¹⁻⁸

A number of recent review articles have

been written on the rheology and applications of wormlike micelle solutions⁹⁻¹³. 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 junctions¹⁴⁻¹⁸. Thus, from the above brief review it is evident that micelles are required for exploitation specially for citric acid fermentation¹⁹ and in view of this the author has studied the influence of lithium 2-dodecylbenzenesulfonate on citric acid production by *Aspergillus oryzae* NCIM-929

Experimental

The influence of lithium 2-dodecylbenzenesulfonate 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 by *Aspergillus oryzae* NCIM-929 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,

Table - 1
Fungal bioproduction of citric acid exposed to lithium 2-dodecylbenzenesulfonate

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	7.536	2.468	-
$1.0 \times 10^{-5} M$	11	7.596	2.406	+0.796
$2.0 \times 10^{-5} M$	11	7.626	2.379	+ 1.194
$3.0 \times 10^{-5} M$	11	7.739	2.269	+ 2.693
$4.0 \times 10^{-5} M$	11	7.875	2.129	+ 4.498
$5.0 \times 10^{-5} M$	11	8.010	1.996	+ 6.289
$6.0 \times 10^{-5} M$	11	8.176	1.829	+8.492
$7.0 \times 10^{-5} M^{**}$	11	8.410 ^{***}	1.596	+11.597
$8.0 \times 10^{-5} M$	11	8.101	1.896	+7.497
$9.0 \times 10^{-5} M$	11	7.778	2.229	+3.211
$10.0 \times 10^{-5} M$	11	7.664	2.336	+1.698

* 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%

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 lithium 2-dodecylbenzenesulfonate 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 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 fermentor flasks was made upto 100 ml by adding requisite amount of distilled water. Thus, the molar concentration of lithium 2-dodecylbenzenesulfonate 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 micelle solution

The above fermentor flasks were then sterilized, cooled inoculated, incubated at 27°C and analysed after 9, 11 and 13 days for citric acid²⁰ formed and molasses²¹ left unfermented.

Results and Discussion

The influence of lithium 2-dodecylbenzenesulfonate

The data given in the table-1 shows that the micelle lithium 2-dodecylbenzenesulfonate has also been found stimulatory for the production of citric acid by *Aspergillus oryzae* NCIM-929. From the data given in the table-1 it is obvious that lithium 2-dodecylbenzenesulfonate influences the citric acid fermentation process in

different phases. The main characteristics of the lithium 2-dodecylbenzenesulfonate is as follows :

(i) Lithium 2-dodecylbenzenesulfonate is stimulatory at its all molar concentrations used during the course of fermentation for production of citric acid by *Aspergillus oryzae* NCIM-929 i. e., from $1.0 \times 10^{-5} \text{M}$ to $10.0 \times 10^{-5} \text{M}$.

(ii) The molar concentration from $1.0 \times 10^{-5} \text{M}$ to $7.0 \times 10^{-5} \text{M}$ of lithium 2-dodecylbenzenesulfonate influence the yield of citric acid in a approximately regular increasing order after each state, i. e., 0.796%, 1.194%, 2.693% , 4.498%, 6.289%, 8.492% and 11.597%

(iii) The molar concentration $8.0 \times 10^{-5} \text{M}$ to $10.0 \times 10^{-5} \text{M}$ of lithium 2-dodecylbenzenesulfonate also influences the productivity of citric acid in a regular decreasing manner. The % increase in the yield of citric acid at respective molar concentration of lithium 2-dodecyl benzene-

sulfonate has been found to be as follows :

7.497%, 3.211%, and 1.698%.

(iv) The maximum yield of citric acid, i.e; 8.410g/100 ml in the presence of lithium 2-dodecylbenzenesulfonate was observed at $7.0 \times 10^{-5} \text{M}$ molar concentration in 11 days of optimum incubation period which is 11.597% higher in comparison to control fermentor flasks, i.e; 7.536 g/100 ml in the same times course and other same experimental parameters.

(v) The higher molar concentrations of lithium 2-dodecyl-benzenesulfonate were not much favourable for the production of citric acid by *Aspergillus oryzae* NCIM-929 So the gradual addition of the micelle lithium 2 dodecyl-benzenesulfonate after molar concentrations $8.0 \times 10^{-5} \text{M}$ and onwards were not beneficial for the citric acid fermentation process.

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