

# Bioenergetic Transformation of Molasses Pollutant to Ethanol by *Saccharomyces Cerevisiae* Ncim- 2086 Exposed to 5- Nitroindole

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## ABSTRACT

The effect of 5- Nitroindole on bioenergetic transformation of molasses pollutant to ethanol by *saccharomyces cerevisiae* NCIM-2086 has been studied. It has been found that the mutagen i.e 5- Nitroindole under trail has stimulatory effect on bioenergetic transformation of molasses pollutant to ethanol by *saccharomyces cerevisiae* NCIM-2086 and enhances the yield of ethanol to an extent of 7.89473% higher in comparison to control fermentor flasks i.e 5.70ml/100ml in 46 hrs of incubation period 4.8pH and 32°C temperature with 16%(W/V) molasses Solution.

**Keywords :** Molasses, mutagen, ethanol, 5-Nitroindole, *saccharomyces cerevisiae* NCIM-2086)

## I. INTRODUCTION

Chemical mutagens were not demonstrated to cause mutation until the 1940s, when Charlotte Auerbach and J.M.Robson found that mustard gas can cause mutations in fruit files. A large number of chemical mutagens have since been identified, especially after the development of the Ames test in the 1970s by Bruce Ames that screens for mutagens and allows for preliminary identification of carcinogens . Early studies by Ames showed around 90% of known carcinogens can be identified in Ames test as mutagenic and -80% of the mutagens identified through Ames test may also be carcinogens . Mutagens are not necessarily carcinogens, and vice versa. Sodium azide for example may be mutagenic (and highly toxic), but it has not been shown to be carcinogenic.

Chemical mutagens are compounds that increase the frequency of some types of mutation<sup>1-3</sup>. Mutagenic agents are used to induce favourable mutations at high frequency that include ionizing radiation and chemical

mutagens<sup>4-6</sup>. Chemical mutagens are the one cause of mutations in living organism. It is known that various chemicals have positive or negative effects on living organisms. Many of these chemicals have clastogenic (chromosome damaging) effects on plants via reactive oxygen-derived radicals<sup>7</sup>. Many researchers compared the mutagenic efficiencies of different mutagens on different crops and their results seem to be entirely specific for particular species and even varieties. While many researchers found chemical mutagens are to be more effective than physical ones<sup>8-9</sup>. and many others researchers found the reverse case<sup>10</sup> .A number of workers<sup>11</sup>. have reported the role of chemical mutagens in enhancing genetic variability in higher plants because it is the fundamental characteristics to successful breeding programs in vegetatively and sexually propagated plants<sup>12</sup>. the mutants so produced facilitate the isolation, identification and cloning of genes used in designing crops with improved yield and quality traits. A large number of chemical mutagenes are recorded as a good agent for different microbes and microbial processes.

Thus, from the above brief review it is evident that chemical mutagens are required for genetic manipulation and exploitation specially for alcoholic fermentation and in view of this the authors have studied the influence of 5-nitroindole on bioenergetic transformation of molasses pollutant to ethanol by *Saccharomyces cerevisiae* NCIM-2086.

## II. EXPERIMENTAL

The influence of 5-Nitroindole on bioenergetic transformation of molasses pollutant to ethanol by *Saccharomyces cerevisiae* NCIM- 2086 .

The composition of production medium for the bioenergetic transformation of molasses pollutant to ethanol by *Saccharomyces cerevisiae* NCIM- 2086 is prepared as follows :

Molasses	:	16% (w/v)
Malt extract	:	1.25%
Yeast extract	:	1.25%
Peptone	:	1.25%
(NH <sub>4</sub> ) <sub>2</sub> HPO <sub>4</sub>	:	1.25%
pH	:	4.8

Distilled water was added to make up the volume up to '100 ml'.

The pH of the medium was adjusted to 4.8 by adding requisite amount of lactic acid.

Now, the same production medium for bioenergetic transformation of molasses pollutant to ethanol by *Saccharomyces cerevisiae* NCIM- 2086 was prepared for 99 fermentor-flasks, i.e., each containing 100 ml of production medium. These fermentor-flasks were then arranged in 10 sets each comprising 9 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.

Now, M/1000 solutions of 5-Nitroindole 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 to the fermentor-flasks of first 10 sets respectively. The control fermentor-flask contained no chemical mutagens. The total volume in each fermentor-flask was made upto '100 ml' by adding requisite amount of distilled water.

Thus, the concentration of 5-Nitroindole in first, second, third, fourth, fifth, sixth, seventh, eighth, ninth and tenth subsets were approximately as given below :

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

$$1.0 \times 10^{-5} M,$$

$$2.0 \times 10^{-5} M,$$

Where, A = amount of mutagens in ml, ie; from

$$3.0 \times 10^{-5} M, \quad 1.0 \text{ ml to } 10.0 \text{ ml.}$$

$$4.0 \times 10^{-5} M, \quad x = \text{molarity of the solution.}$$

$$5.0 \times 10^{-5} M, \quad \text{and respectively.}$$

$$6.0 \times 10^{-5} M,$$

$$7.0 \times 10^{-5} M,$$

$$8.0 \times 10^{-5} M,$$

$$9.0 \times 10^{-5} M,$$

$$10.0 \times 10^{-5} M$$

The fermentor-flasks were then steam sterilized, cooled, inoculated, incubated at 32<sup>0</sup> C and analysed colorimetrically after 40,46, and 50 hours for alcohol formed and molasses sugars left unfermented.

**Table-1.** Bioenergetic transformation of molasses pollutant to ethanol by *Saccharomyces cerevisiae* NCIM- 2086 exposed to 5-nitroindole

Concentration of mutagen Used A X 10* M	Incubation Period in hours	Yield of ethanol* in ml/100 ml	Molasses Sugars* left unfermented in g/100 ml	% Difference in yield of ethanol in comparison to control
1.0 × 10 <sup>-5</sup> M (+) Mutagen	46	5.74	2.25876	+ 0.70175
2.0 × 10 <sup>-5</sup> M (+) Mutagen	46	5.80	2.19880	+ 1.75438
3.0 × 10 <sup>-5</sup> M (+) Mutagen	46	5.91	2.08883	+ 3.68421
4.0 × 10 <sup>-5</sup> M (+) Mutagen	46	6.03	1.96884	+ 5.78947
5.0 × 10 <sup>-5</sup> M (+) Mutagen	46	6.08	1.91888	+ 6.66666
6.0 × 10 <sup>-5</sup> M** (+) Mutagen	46	6.15***	1.88380	+ 7.89473
7.0 × 10 <sup>-5</sup> M (+) Mutagen	46	6.02	1.97893	+ 5.61403
8.0 × 10 <sup>-5</sup> M (+) Mutagen	46	5.92	2.07896	+ 3.85964
9.0 × 10 <sup>-5</sup> M (+) Mutagen	46	5.82	2.17898	+ 2.10526
10.0 × 10 <sup>-5</sup> M (+) Mutagen	46	5.75	2.24890	+ 0.87719

\* Each value represents mean of three trials.

\*\* Optimum concentration of the chemical mutagen used.

\*\*\* Optimum yield of ethanol in 46 hours.

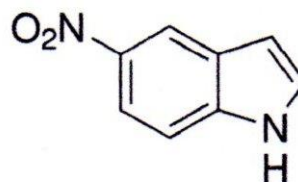
(+) Values indicate % increase in the yield of ethanol in comparison to control.

Experimental deviation (±) 1.5–3%.

### III. RESULTS AND DISCUSSION

The influence of 5-Nitroindole on bioenergetic transformation of molasses pollutant to ethanol by

*Saccharomyces cerevisiae* NCIM- 2086 5-Nitroindole



The data recorded in the table-1 shows that 5-Nitroindole has stimulatory effect on bioenergetic transformation of molasses pollutant to ethanol by *Saccharomyces cerevisiae* NCIM- 2086

The maximum yield of ethanol, i.e., 6.15 ml/100 ml in the presence of 5-Nitroindole was observed at  $6.0 \times 10^{-5}$  M molar concentration in 46 hours of optimum incubation period which is 7.89473% higher in comparison to control fermentor flasks, i.e.; 5.70ml/100ml in the same times course and other same experimental parameters.

The higher molar concentrations of 5-Nitroindole were not much favourable for the bioenergetic transformation of molasses pollutant to ethanol by *Saccharomyces cerevisiae* NCIM- 2086 . So the gradual addition of the mutagen 5-Nitroindole after certain concentrations were not beneficial for the bioenergetic transformation of molasses pollutant to ethanol by *Saccharomyces cerevisiae* NCIM-2086.

It has been observed that molar concentration of the mutagen, i.e., 5-Nitroindole from  $1.0 \times 10^{-5}$  M to  $6.0 \times 10^{-5}$  M enhances the yield of ethanol to an order being 0.70175%, 1.75438%, 3.68421%, 5.78947%, 6.6666%, 7.89473% higher in comparison to control flasks.

It has been observed further that after optimum concentration, i.e;  $6.0 \times 10^{-5}$  M, the addition of the same mutagen, i.e. 5-Nitroindole to the production medium causes fall in the yield of ethanol gradually and reached to 0.87719%. However, at all the experimental concentrations of mutagen, i.e. 5-Nitroindol used, the yield of ethanol by *Saccharomyces cerevisiae* NCIM- 2086 has been found higher in comparison to control fermentor flasks.

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