

Impact of acriflavine on bioproduction of microbial lactic acid

Shivankari

Department of Chemistry, Magadh University, Bodh-Gaya (Bihar) India

Email : shivankariroy@gmail.com

Manuscript received online 26 June 2020, accepted on 13 August 2020

Abstract: The impact of acriflavine on bioproduction of lactic acid by some lactic acid producing bacteria such as *Lactobacillus jugrti* NCIM- 2367, *Lactobacillus lactis* NCIM-2369, *Lactobacillus plantarum* NCIM-2373, *Lactobacillus brevis* NCIM-2436 and *Lactobacillus delbrueckii* NCIM-2025 has been studied. The bacterial strain *Lactobacillus delbrueckii* NCIM-2025 has been found quite effective and useful for maximum bioproduction of lactic acid, i.e., 10.745 g/100 ml of lactic acid which is 12.360% higher in comparison to control fermentor flasks, i.e., 9.563 g/100ml at 4.0×10^{-5} M molar concentration of acriflavine mutagene. It has been found that bioproduction of lactic acid in the presence of acriflavine at optimum molar concentration, i.e; at 6.0×10^{-5} M attains its best activity when 24% (w/v) molasses solution is allowed to ferment for 5 days of optimum incubation period, at 38°C temperature by maintaining the pH value of the fermentation medium at 5.9 along with other nutritional ingredients required by the lactic acid bacteria.

(Keywords: Lactic acid fermentation, acriflavine, molasses, *Lactobacillus delbrueckii* NCIM-2025, pH, temperature and incubation period.)

Introduction

Chemical Mutagens: Environmental effects on biological systems brings together relevant facts about synthetic and naturally occurring mutagenic chemicals¹⁻¹⁰. Chemical mutagens induce in the various test systems that have been utilized.

Mutation can result in several different types of change in deoxyribo nucleic acid sequences; these can either have no effect, alter the product of a gene, or prevent the gene from

functioning properly or completely¹⁻¹⁶. Mutations can involve large sections of deoxyribo nucleic acid becoming duplicated, usually through genetic recombination. The influence of mutagens in fermentation technology specially lactic acid fermentation has been studied extensively.

Some mutagens where tested by many workers¹⁷⁻³⁰ for different fermentations process and were found most of them were found useful for higher production of the desired products. It is obvious from above review of literature that various chemical mutagens and some other mutagenic agents are used to produce mutants. Thus, it is concluded that a large number of mutagens have been employed to generate the mutants of different microbes but still there are some chemical mutagens whose impact on production of lactic acid by *Lactobacillus delbrueckii* NCIM-2025 have not been well studied and established. Moreover, survey of the literature reveals that there has been not enough mention to study the lactic acid fermentation exposed to mutagens specially chemical mutagens. Therefore, in the present investigation the authoress has made an attempt to study the bioproduction of lactic acid by *Lactobacillus delbrueckii* NCIM-2025 exposed to exposed to acriflavine

Experimental

Compositions of the production medium :

The composition of the production medium for the bioproduction of lactic acid by *Lactobacillus delbrueckii* NCIM-2025 exposed to acriflavine is as follows :

Molasses : 24% (w/v), Malt Extract : 1.60%

Table - 1
Impact of acriflavine on bioproduction of microbial lactic acid

Concentration of mutagen used a x 10 ^{-x} M	Incubation period in days	Yield of lactic acid* in g/100 ml	Molasses substrate* left unfermented in g/100 ml	% of lactic acid increase in 5 days of incubation pd.
Control	4	7.649	4.438	—
(– mutagen)	5	9.563	2.527	—
	6	8.482	2.418	—
1.0 x 10 ⁻⁵ M	4	7.710	4.386	—
(+ mutagen)	5	9.644	2.454	+0.847
	6	8.538	2.359	—
2.0 x 10 ⁻⁵ M	4	7.729	4.369	—
(+ mutagen)	5	9.696	2.399	+1.390
	6	8.589	2.286	—
3.0 x 10 ⁻⁵ M	4	8.035	4.063	—
(+ mutagen)	5	10.075	1.984	+5.353
	6	9.065	1.875	—
4.0 x 10 ⁻⁵ M	4	8.184	3.885	—
(+ mutagen)	5	10.257	1.802	+7.257
	6	9.155	1.710	—
5.0 x 10 ⁻⁵ M	4	8.337	3.720	—
(+ mutagen)	5	10.458	1.567	+9.358
	6	9.356	1.464	—
6.0 x 10 ⁻⁵ M**	4	8.566	3.493	—
(+ mutagen)	5	10.745***	1.315	+12.360
	6	9.640	1.211	—
7.0 x 10 ⁻⁵ M	4	8.361	3.686	—
(+ mutagen)	5	10.352	1.715	+8.250
	6	9.259	1.613	—
8.0 x 10 ⁻⁵ M	4	7.955	4.103	—
(+ mutagen)	5	9.965	2.093	+4.203
	6	8.869	2.081	—
9.0 x 10 ⁻⁵ M	4	7.786	4.273	—
(+ mutagen)	5	9.748	2.294	+1.934
	6	8.637	2.184	—
10.0 x 10 ⁻⁵ M	4	7.726	4.332	—
(+ mutagen)	5	9.670	2.428	+1.118
	6	8.566	2.316	—

* Each value shows mean of three observation ** Optimum concentration of mutagens under observation

*** Maximum production of lactic acid (LA) Positive Values indicate percent enhancement in the production of CH₃CHOHCOOH Experimental deviation $\pm 2.5 - 3.5\%$

Yeast Extract : 1.60%, Peptone : 1.60%, $(\text{NH}_4)_2\text{HPO}_4$: 1.60%, CaCO_3 : 11.0%, pH : 5.9
(The pH was Adjusted by adding requisite amount of phosphate-buffer solution). Distilled water : To make up 100 ml.

Assay methods :

Evaluation of lactic acid³¹ formed and molasses³² left unfermented was made colorimetrically

Sterilization : The growth and production medium was sterilized in an autoclave maintained at 15 lbs steam pressure for 30 minutes.

Strain : *Lactobacillus delbrueckii* NCIM-2025 has been employed in the present study. The strain was procured from NCL - Pune, India

Age of the inoculum : 48 hours old.

Quantum of the inoculum: 0.5 ml bacterial suspension of *Lactobacillus delbrueckii* NCIM-2025.

Incubation period : 4, 5 and 6 days

Concentration of acriflavine used :

M/1000 solution of acriflavine under trial has been prepared and $1.0 \times 10^{-5}\text{M}$ to $10 \times 10^{-5}\text{M}$ molar concentration of acriflavine has been employed.

Results and Discussion

The influence of acriflavine on production of microbial lactic acid by *Lactobacillus delbrueckii* NCIM-2025

The data recorded in the table-1 shows that the chemical mutagen acriflavine has detrimental effect on production of microbial lactic acid by *Lactobacillus delbrueckii* NCIM-2025. The yields of lactic acid obtained in the control fermenter flasks has been found to be higher than that obtained from each of the fermentor flasks containing chemical mutagen, i.e., acriflavine

The maximum yield of lactic acid, i.e., 10.745 g/100 ml. in the presence of acriflavine, i.e., at $6.0 \times 10^{-5}\text{M}$ was found in 5 days of optimum incubation period, which is 12.360% higher in comparison to control i.e., 9.563 g/100ml.

However, at higher concentrations of the chemical mutagen, i.e., acriflavine the production of lactic acid was found almost negligible and insignificant. Thus, it is obvious from the results that the chemical mutagen acriflavine under trial is much detrimental and inhibitory for production of microbial lactic acid by *Lactobacillus delbrueckii* NCIM-2025

References

1. J.W. Drake, J.J. Holland : *Proc. Natl. Acad. Sci. U.S.A.* **96** (24): 13910 (1999).
2. J. S. Felton *et al.*, *Food Chemical Toxicology* **32** (10), 897 (1994).
3. P. Sniegowski, P. Gerrish, T. Johnson, A. Shaver : *Bioessays* **22** (12): 1057 (2000).
4. SA Sawyer, J. Parsch, Z. Zhang, DL. Hartl : *Proc. Natl. Acad. Sci. U.S.A.* **104** (16): 6504 (2007).
5. B. Commoner *et al.*, *Science* **210**, 913 (1978).
6. G. A. Gross, *Carcinogenesis* **11**, 1597 (1990).
7. J. Bertram : *Mol. Aspects Med.* **21** (6): 167, (2000)
8. J. Holland, K Spindler, F. Horodyski, E. Grabau, S. Nichol, S. VandePol : *Science* **215** (4540): 1577 (1982).
9. B. N. Ames, J. McCann, and E. Yamasaki " *Mutation Research* **31**, 347 (1975).
10. T. Sugimura *et al.*, in *Origins of Human Cancer*, H. H. Hiatt, J. D. Watson, and J. A. Winsten, Eds., (Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, , 1561 (1977)
11. Y.P. Lai, J. Huang, L. Li and Z. R. Wu, *Biotechnology and Bioengineering*, **86**(6), 622 (2004).
12. P J. Hastings, ; J. R. Lupski, ; S. M. Rosenberg, ; G Ira, : *Nature Reviews. Genetics* **10** (8): 551 (2009)
13. SB. Carroll, J. Grenier, SD Weatherbee : *From DNA to Diversity: Molecular Genetics and the Evolution of Animal Design. Second Edition.* Oxford: Blackwell Publishing. (2005).

14. P. Harrison, M. Gerstein : *J Mol Biol* **318** (5): 1155 (2002).
15. CA Orengo , JM Thornton : *Annu. Rev. Biochem.* **74**: 867 (2005).
16. M. Long, E. Betrán, K. Thornton, W. Wang : *Nat.Rev. Genet.* **4** (11): 865 (2003).
17. Bireshwar Mitra, M. Ziauddin Shahzada , Md. Shahabuddin, Afsar Nasim, Akhouri Prashant Kumar and Manjushree Prasad *J.Chemtracks*, **17** (1), 47 (2015)
18. H. K. Satapathy, V. Mandal, J. P. Kumar, Sanjit K. Anand, Jyotimala and S.P. Singh *J.Chemtracks*, **17** (1), 141 (2015)
19. S. Kumar, Ejaz Jawaid, Nagmani Kumar, Md. Shahabuddin, Afsar Nasim and S. P. Singh *J.Chemtracks*, **17** (1), 181 (2015)
20. R. Mishra, L. Kumari ,M. Prasad, Mona Kumari, P. K. Sinha and S. P. Singh, *J. Chemtracks*, **17** (2), 247 (2015)
21. P. Singh, J. P. Kumar, Sheela Kumari, D. K. Singh, Vijay Kumar, U. S. Singh, and S. P. Singh *J. Chemtracks*, **17** (2), 333 (2015)
22. R. Kumar, J. Prakash Kumar, S. Kumari, Vijay Kumar , U.S. Singh and S. P. Singh *J.Chemtracks*, **17** (2), 369 (2015)
23. M. Kumari, A. Singh, L. Kumari, P.K. Sinha, S. S. Kumari and S. P. Singh *J. Chemtraks* **18**(1), 85 (2016).
24. R.S. Sharma, B.K. Choudhary, Neeta Sinha, and B.B. Sharma *J. Chemtraks* **18**(2), 319 (2016).
25. R. Kumari, S. Singh and S. P. Singh *J. Chemtraks* **18**(2), 329 (2016).
26. P. Singh, F. Qmar, B.K. Choudhary, B.B. Sharma and S.P. Singh *J. Chemtraks* **18**(1), 2016 (2016).
27. K. Sangita, Lalan Kumar and S.P. Singh *J. Chemtraks* **19**(1), 71 (2017)
28. Shivankari, Madhurendra, Bijay Kumar and S. P. Singh *J. Chemtraks* **19**(2), 237 (2017)
29. Shivankari, A. Suraiya and S. P. Singh *J. Chemtraks* **21**(1&2), 5-8 (2019)
30. Shivankari, S. Singh and S. P. Singh *Biotech Today* **10**(1), 44 (2020)
31. S. B. Barker, and W. H. Summerson, *J. Biol. Chem.* **138**, 534 (1941)
32. M. Dubois, K. A. Gills, J. K. Hamilton, P. A. Rebers and F. Smith, *Anal. Chem.* **28**, 350 (1956).