

# Biokrishi Technologies Private Limited



# Case Study



Jakraya Sugar pvt ltd a Molasses /Cane syrup ethanol plant in Solapur, Maharashtra, India.

#### It shows:

- How the Antibacterial products and Bacterial contamination control methodology would work
- Decisions on commercial criteria
- Challenges and Questions that remain
- Trial results



Biokrishi Technologies Private Limited DSK Vishwa,Varun D, Flat N0 703, SN 126, Dhayari Pune 411041 ,Maharashtra ,India



15th March ,2023

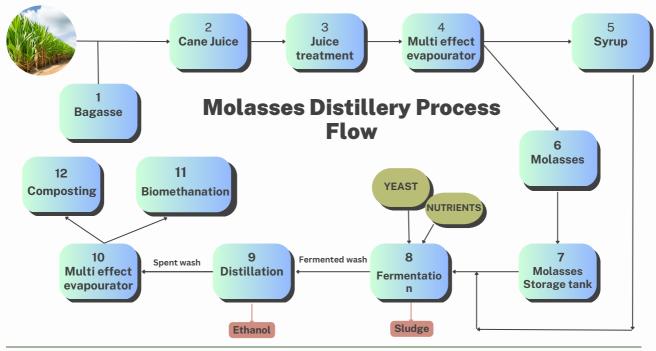


#### Introduction

To promote the country's fuel grade ethanol producing distilleries. The Indian government has approved the manufacturing and procurement of ethanol from sugarcane-based raw materials such as C & B heavy molasses, sugarcane juice / sugar / sugar syrup, surplus rice with Food Corporation of India (FCI), and maize. Although most distilleries are familiar with ethanol production from C molasses, changes in process substrate, such as B molasses and sugarsyrup substrate, necessitate the optimization of fermentation media as well as microbial contamination control strategies.

Several scientific studies in the microbiological field have been critical for the evolution of the ethanol industry in India, including the <u>improvement and development of new fermentation processes</u>, the <u>selection of commercial industrial yeast strains</u>, better control of bacterial contaminants, and the <u>improvement of chemical and microbiological control at distilleries</u>.

Bacterial infections are one of the most significant contributors to poor fermentation. Infections prevent yeast from fermenting sugar into ethanol. Annually, ethanol plants might spend a lot of money on products that are used to prevent the spread of infection. Furthermore, expenditures in research on first-generation ethanol have encouraged certain corporations to participate in the production of ethanol from sugarcane juice/syrup and B heavy molasses. The industry is being confronted with new scientific and technological boundaries in order to improve the fermentation process for ethanol production in India.



# Problem statement



#### Jakraya Sugar distillery Case:

In all, 150 KLPD The ethanol plant at Jakraya distillery in Solapur, Maharashtra, was facing contamination difficulties during ethanol production. Despite the use of enzyme-based antibacterial solutions, it was established that they had a serious problem with a bacterial infection that was impacting the plant's functioning and daily ethanol production. Jakraya distilleries differ in terms of fermentation procedure and B-heavy molasses must make up. Jakraya Distillery buys molasses from distilleries connected to sugar factories and usually ferments B-heavy musts of molasses diluted with water due to their large factory capacity and their own B-heavy molasses from sugar factories.

When B heavy molasses arrived at their company's operations it was stored for several weeks, and ethanol fermentations could take place 9 to 10 months of the year. Unlike C molasses, B molasses must be stored in a storage tank, and a strategy must be developed to avoid loss due to microbial contamination. Furthermore, due to the summer season, the industry faces bacterial contamination difficulties throughout the month of March to June.

Along with bacterial contamination issues, it also has <u>difficulties with alcohol</u> recovery, process, and molasses medium optimization due to raw material <u>fluctuations</u>, and stuck fermentation due to low pH in the fermentation process.

# Biokrushi team solves the Microbial contamination and Alcohol recovery problem for ethanol plant

The bacterial contamination <u>problems began in February 2023</u>, when the ethanol factory began operations and used B-heavy molasses for the fermentation process. By treating <u>500 m3 of B-heavy molasses feed each day</u>, this facility may produce up to 1300 m3/day of fermented wash. Based on the distillery management's request, the Biokrushi team visited their plant to monitor the overall manufacturing process and identify the root causes. The objectives for resolving the problem have been set based on plant monitoring.

### **Process Overview**



# Process monitoring, challenges and Objectives

### Fermenter Cleaning

In fed-batch fermentations, each tank is filled, managed, and cleaned separately from the other with the help of raw water, clean water followed by steam. The <u>steam is applied to the fermenter for a half hour</u> and the water has been drained from the bottom of the fermenter. It was observed that the process of cleaning the fermenter was very efficient as no contamination has been observed in the fermenter run with the uncontaminated B-heavy molasses

#### Fermentation Process

The overall factory contains a **total of Eleven fermenters**, out of which five fermenters were newly installed .while the remaining six fermenters include three fermenters with a working capacity of 275m3 and the remaining three with 295 m3 capacity.

	Old fermeters	Old fermeters	New fermenters
Number of Fermneters	3 NOS	3 NOS	5 NOS
Fermenter volume (m3)	350	400	600
Working Capacity (m3)	275	296	496

All of the fermenters in the factory experience bacterial contamination issues as a result of contaminated <u>B-heavy molasses from the Jakraya factory</u>. They received an average recovery of 250-260 L per MT of B-heavy molasses.

The factory receives an average ethanol recovery of 295-300 L per MT of B-heavy molasses from the other sugar refinery in newly installed fermenters. When considering the quality of molasses, we discovered that this recovery is relatively poor.

# Microscopic and Chemical Observations of B-Molasses



#### Contamination of Bacteria in B-heavy Molasses from Jakraya factory

The primary bacterial communities were represented by Gram-positive rods and a little amount of Gram-positive cocci, according to the microscopic examination of prefermenter (PF) and fermenter samples. Despite the fact that Gram-positive bacteria are the most common cause of bacterial contamination, a case of severe loss in fermentation yield was documented. Because of the contamination, the <u>initial bacterial count in the PF was discovered to be higher than the yeast cell count.</u>

#### Chemical composition of B-heavy Molasses from the Jakraya factory

All of the other criteria, such as TRS, FS, and UFS, were found to be acceptable, however the TVA concentration of molasses was discovered to be <u>extremely high, as 9600 ppm.</u>

#### Contamination of Bacteria in B-heavy Molasses from other sugar factory

<u>Gram-positive rod shape bacteria</u> were found in very modest quantities in the PF and fermenter samples

#### Chemical composition of B-heavy Molasses from the other sugar factory

All of the other criteria, such as TRS, FS, and UFS, were found to be satisfactory. Molasses' <u>TVA content was measured to be 3000 ppm</u>

## Problem findings and challenges

#### Jakraya B-heavy Molasses

- 1.Bacterial contamination in fermentation caused by <u>contaminated B-heavy molasses</u> from the Jakraya factory
- 2.A high TVA content in the molasses results in a very high initial VA in the fermentation environment, which <u>stresses the yeast and inhibits it</u>. This also provides more opportunities for bacteria to grow in the media.
- 3.Increased bacterial contamination and TVA <u>reduce the ethanol output per tonne of molasses</u>, which is 250-260 litres per tonne of molasses.

#### Other sugar factory B-heavy Molasses

- 1. <u>Poor alcohol recovery per metric tonne of B molasses</u> ( Molasses purchases from the other outside sugar factory )
- 2.Despite the fact that the molasses quality was good, the plant management attempted to increase the molasses in the fermentation media in order to obtain more alcohol per fermeter. They obtained more ethanol each fermenter by doing so, but the <u>overall alcohol recovery was reduced to 295-300 Liter per MT of molasses.</u>

#### Commercial challenges

1.Because the existing cost of antibacterial products was shown to be excessive, management was primarily concerned with <u>minimizing the cost of these items while</u> <u>maximizing alcohol recovery.</u>

# **Propose solutions**



# Biokrushi Cost effective, efficient enzyme based antibacterial booster and process optimization Solutions

The Biokrushi team has proposed the following product to control bacterial contamination based on <u>process monitoring</u>, <u>microscopic and chemical examinations</u> of fermentation and molasses samples.

	Name of the Product	Product benefits	Application area
1	Promol	High protease	Fermenter
2	Promol plus	Blend of Enzymes	PF
3	Energy RetreatPlus	Blend of Enzymes with broad spectrum activity	PF and Fermenter

Along with the products mentioned above, we have suggested a few process improvements such as

- 1. Nutrient
- 2. Molasses factor (molasses to water dilution ratio)
- 3. Change in Molasses feeding rate

To obtain precise findings, <u>two separate trials</u> were carried out in two different fermenter sizes. In total, five test fermenters were run, and their findings were compared to five control fermenters in two independent trials.

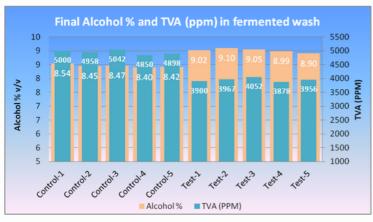
Sr no	Trial no	Fermenter set up	B-Molasses	Objective
1	Trial 1	Old fermentation setup	Molasses from Jakraya factory	To control bacterial contamination and to get more alcohol recovery
2	Trial 2	New fermentation set up	Molasses purchased from Other sugar factory	To optimize the process parameter to get more recovery of alcohol per ton of molasses

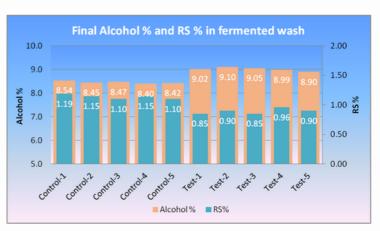
# Results:

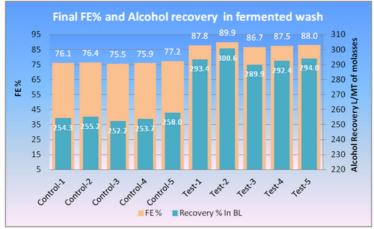




To control Contamination in fermentation of B-Heavy molasses from the Jakraya factory







- Graph I shows that the final alcohol percentage in the test(molasses factor of 0.30) increased by around <u>an average of 0.55%v/v</u> when compared to the control(molasses factor of 0.34). <u>This increase in alcohol is due to low molasses factor and low TVA formation</u>, as reduction in TVA formation has been observed in tests i.e 1000 ppm.
- Graph 2 shows that the <u>final RS% is less than 1%</u> in all test results, which is due to more conversion of RS into alcohol, as the increase in alcohol% has been observed in all test results when compared to control.
- Graph 3 results show that when compared to the control, both FE% and alcohol recovery increase in the test results. The FE% and alcohol recovery have both increased by around 10-11% and 37-39 L per tonne of B-heavy molasses respectively,
- The rate of fermentation in the PF and Fermenter was found to be faster in both processes, and the <u>retention time was reduced by 2 to 3 hours.</u>
- The <u>stuck fermentation problem in the factory is also solved</u> as a result of improvements in the fermentation process such as molasses feeding rate, correct nutrient delivery, and effective bacterial contamination control strategy.

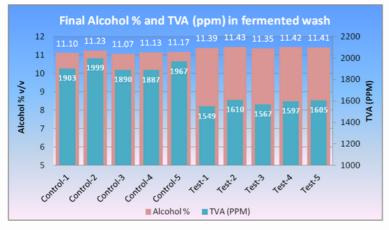
The above results clearly demonstrated the effectiveness of the product at their recommended doses, as well as the process change parameters recommended by the biokrushi team to the factory.

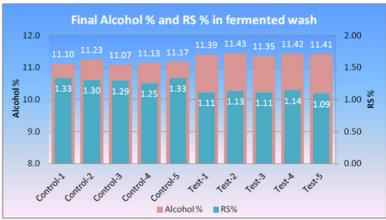
# Results:

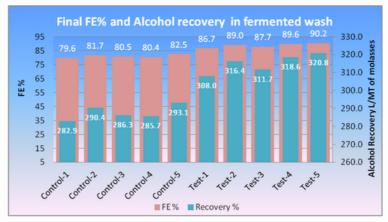
# TRIAL-2



To optimize the process for ethanol fermentation for B molasses from the other sugar factory







- Graph 1 shows that the final alcohol percentage in the test(molasses factor of 0.36) increased by around 0.25%v/v when compared to the control(molasses factor of 0.39). This increase in alcohol is due to the low molasses factor and low TVA formation, as less TVA formation has been observed in tests at 300-350 ppm.
- Graph 2 shows that the final <u>RS% is decreased by an average of 0.20%</u> in all test results, which is due to more conversion of RS into alcohol, as the increase in alcohol% has been observed in all test results when compared to the control.
- Graph 3 results show that when compared to the control, both FE% and alcohol recovery increase in the test results. The <u>FE% and alcohol recovery have both</u> <u>increased by around 7-8% and 25-30 L per tonne of B-heavy molasses</u> respectively,
- The rate of fermentation in the PF and Fermenter was found to be faster in both processes, and the retention time was reduced by 2 to 3 hours.
- The <u>stuck fermentation problem in the factory is also solved</u> as a result of improvements in the fermentation process such as molasses feeding rate, correct nutrient delivery, and effective bacterial contamination control strategy.

The above results clearly demonstrated the effectiveness of the product at their recommended doses, as well as the process change parameters recommended by the biokrushi team to the factory.

# Conclusion



## Profit calculations

# Bacterial Contamination Control Trial-1

Benefits of the Test product Promol, Promol plus and Energy Retreat plus are observed when compared to the Control .

Parameter	TEST AVERAGE	CONTROL AVERAGE
Fermenter Working Volume (lit)	296000	296000
Molasses in MT per fermenter	90.7	98.3
Alcohol % (AL)	9.01	8.46
Increase in Alcohol % (AL) compared to control	0.56	
Recovery/MTof B- heavy molasses (AL)	294.0	254
Extra alcohol in lit per MT of Molasses	39.4	
Extra alcohol in lit per fermenter	1645	
Price of extra alcohol (INR per lit)	60	
Price of extra alcohol (INR per fermenter )	98745	

Price of alcohol from the save molasses		
Molasses save per fermenter (MT)	7.56	
Alcohol produced from the save molasses (Liter)	2223	
Price of save alcohol (INR) per lit	60	
Price of alcohol from the save molasses	133378	

Profit earn calculations		
Price of extra alcohol (INR) per fermenter	98745	
Price of alcohol from the save molasses	133378	
Total price of Alcohol	232123	
Price of Enzyme products per fermenter	18100	
Profit earn per fermenter from the Product and Process change (INR)	214023	
Total per day profit earn in lakhs (per day capacity of 1300000 lit wash)	9.4	

# Conclusion



#### Profit calculations

# Process Optimization Trial-2

Benefits of the Test product Promol, Promol plus and Energy Retreat plus along with change in process parameters are observed when compared to the Control .

Parameter	TEST AVERAGE	CONTROL AVERAGE
Fermenter Working Volume (lit)	496000	496000
Molasses in MT per fermenter	179.5	192.1
Alcohol % (AL)	11.40	11.14
Increase in Alcohol % (AL) compared to control	0.26	
Recovery/MTof B- heavy molasses (AL)	315.0	287.7
Extra alcohol in lit per MT of Molasses	27.4	
Extra alcohol in lit per fermenter	1289	
Price of extra alcohol (INR per lit)	60	
Price of extra alcohol (INR per fermenter )	77376	

Price of alcohol from the save molasses		
Molasses save per fermenter (MT)	12.60	
Alcohol produced from the save molasses (Liter)	3970	
Price of save alcohol (INR) per lit	60	
Price of alcohol from the save molasses	238173	

Profit earn calculations	
Price of extra alcohol (INR) per fermenter	77376
Price of alcohol from the save molasses	238173
Total price of Alcohol	315549
Price of Enzyme products per fermenter	14200
Profit earn <u>per fermenter</u> from the Product and Process change (INR)	301349
Total per day profit earn in lakhs (per day capacity of 1300000 lit wash)	7.9