

## POTENTIALS OF YEAST ISOLATES FOR BIOSURFACTANT PRODUCTION

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### ABSTRACT

Twenty yeast isolates were screened for potential to produce biosurfactants using three different methods namely: collapse of oil drops, oil spreading or displacement and emulsification potential. Of the twenty yeast isolates screened, sixteen (80.0 %) showed collapse of oil drops. Eleven isolates (55 %) were able to displace the oil with *Saccharomyces cerevisiae* H02, *Candida boleticola* H09 and *Rhodotorula bogoriensis* H15 having the highest diameter of displacement of 6.2 cm, 5.3 cm and 4.7 cm respectively. The isolates had emulsifying capacities that ranged from 6.67 % to 33.33 %. The results showed that *Saccharomyces cerevisiae* H02, *Candida boleticola* H09 and *Rhodotorula bogoriensis* H15 were considered efficient potential candidates for biosurfactants production.

**Keywords:** yeast, biosurfactants, oil collapse, oil displacement, emulsification

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### INTRODUCTION

Biosurfactants are amphiphilic compounds, which are secreted extracellularly by microorganisms (Luna *et al.*, 2013). They can be used in many industries as bio-emulsifiers in the food industries; as antimicrobial agent; in detergent formulation; in bioremediation processes as well as in enhanced crude oil recovery. Even though yeasts are known for producing biosurfactants in higher concentrations than bacteria, which is an advantage regarding these microorganisms, minimal studies have reported biosurfactants synthesized by yeasts (Hua *et al.*, 2003). Hence the need to explore yeasts in biosurfactant production, which was the basis for this study.

### MATERIALS AND METHODS

#### Yeast strains

Twenty yeast isolates were used. The yeasts were obtained from Department of Microbiology, Federal University of Technology, Minna, Nigeria.

**Drop collapse test:** Oil drop collapse assay developed by Jain *et al.* (2011) was adopted.

**Oil displacement test:** The oil displacement test was done according to methods described by Rodrigues *et al.* (2006)

**Emulsification activity:** Emulsification ability of biosurfactants towards kerosene was studied using methods of Youssef *et al.* (2004).

**Statistical analysis:** Data was analyzed statistically using error mean square and

correlation analysis. Computer statistical package SPSS 9.0 was used.

## RESULTS

Of the twenty yeast isolates screened, sixteen (80.0 %) showed positive result (that is, dispersed the oil around the

liquid droplet). Of the sixteen positive isolates, only seven (43.75 %) were able to collapse the oil droplet (that is, caused it to spread out and appeared flat on the solid surface within one minute. The results of the oil drop collapse caused by yeast isolates are shown in Table 1

Table 1: Extent of collapse of oil drops caused by yeast isolates

Coded yeast isolates	Reaction
<i>Pichia strasburgensis</i> H01*	+
<i>Saccharomyces cerevisiae</i> H02	++
<i>Saccharomyces cerevisiae</i> H03	++
<i>Cryptococcus laurentii</i> H04	+
<i>Cryptococcus skinneri</i> H05	+
<i>Yarrowia lipolytica</i> H06	++
<i>Candida zelandoides</i> H07	+
<i>Candida apis</i> H08	-
<i>Candida boleticola</i> H09	++
<i>Candida spandovensis</i> H10	+
<i>Candida spandovensis</i> H11	+
<i>Candida tropicalis</i> H12	+
<i>Candida tropicalis</i> H13	++
<i>Rhodotorula glutinis</i> H14	++
<i>Rhodotorula bogoriensis</i> H15	++
<i>Rhodotorula bogoriensis</i> H16	-
<i>Candida acuta</i> H17	-
<i>Candida acuta</i> H18	-
<i>Candida acuta</i> H19	+
<i>Candida acuta</i> H20	+
Control	-

Key: + = positive only; ++ = positive and collapsed; - : negative. Control: distilled water.

\*Code assigned to yeasts in our laboratory.

### Oil spreading or displacement potential of the yeast isolates

The twenty yeast isolates were also screened for their potential to displace and spread crude oil. Of this number, 11 isolates (55 %) were able to displace the oil. The diameter of oil spread ranged from 1.2 cm to 6.2 cm within 5 to 35 seconds (Table 2). It was observed that *Saccharomyces cerevisiae* H02, *Candida boleticola* H09 and *Rhodotorula bogoriensis* H15 had the highest diameter

of displacement of 6.2, 5.3 and 4.7 respectively within 10 seconds, meaning that they had strong ability to displace crude oil.

### Emulsification potential of the yeast isolates

The emulsification capacities of the yeasts were tested and it was observed that the organisms had varying emulsifying capacities which ranged from 6.67 % to 33.33 % (Table 3). Twelve (60 %) of the

isolates (*Pichia strasburgensis*, *Saccharomyces cerevisiae* H03, *Candida zelandoides* H07, *Candida apis* H08, *Candida spandovensis* H10, *Candida spandovensis* H11, *Candida tropicalis* H12, *Rhodotorula glutinis* H14, *Rhodotorula bogoriensis* H16, *Candida acuta* H17, *Candida acuta* H18, and *Candida acuta* H20) had no emulsifying capacity, 5 (25 %) of the isolates (*Yarrowia lipolytica* H06, *Candida acuta* H19, *Cryptococcus laurentii* H04, *Cryptococcus skinneri* H05, and *Candida*

*tropicalis* H13) had very low emulsification indexes of 6.67 %, 8.11 %, 16.67 %, 16.67 % and 16.67 % respectively, 3 (15 %) isolates (*Saccharomyces cerevisiae* H02, *Candida boleticola* H09 and *Rhodotorula bogoriensis* H15) had emulsification indexes of 33.33 %, 27.33 % and 24.20 % which is considered moderate. However none of the isolates showed percentage emulsification that was significantly high (50 % to 100 %).

Table 2: Diameter and time of displacement caused by yeast isolates

Coded yeast isolates	Diameter (cm)	Time (s)	Interpretation	
<i>Saccharomyces cerevisiae</i> H02*	6.2	5	Positive	
<i>Candida boleticola</i> H09	5.3	7		
<i>Rhodotorula bogoriensis</i> H15	4.7	10		
<i>Pichia strasburgensis</i> H01	4.5	12		
<i>Cryptococcus laurentii</i> H04	3.8	35		
<i>Saccharomyces cerevisiae</i> H03	3.5	20		
<i>Candida acuta</i> H19	3.2	34		
<i>Yarrowia lipolytica</i> H06	2.7	13		
<i>Cryptococcus skinneri</i> H05	2.3	9		
<i>Candida zelandoides</i> H07	1.2	7		
<i>Candida tropicalis</i> H13	1.2	16		
<i>Candida apis</i> H08	-	-		Negative
<i>Candida spandovensis</i> H10	-	-		
<i>Candida spandovensis</i> H11	-	-		
<i>Candida tropicalis</i> H12	-	-		
<i>Rhodotorula glutinis</i> H14	-	-		
<i>Rhodotorula bogoriensis</i> H16	-	-		
<i>Candida acuta</i> H17	-	-		
<i>Candida acuta</i> H18	-	-		
<i>Candida acuta</i> H20	-	-		
Control A	-	-		
Control B	-	-		

Key: Control A=Distilled water; Negative=diameter less than 4.5 and occurred after 30 seconds; Control B=sterile broth; Positive=diameter from 4.5 and above and occurred within 30 seconds; cm=centimeter; s=seconds

Note: The results are arranged in ascending order of displacement with time.

\*Code assigned to yeasts in our laboratory

Table 1. Oil drop collapse index of 21 yeast strains

Yeast Strain	Oil Drop Collapse Index (%)
<i>Saccharomyces cerevisiae</i> H01*	0 ± 0.00 <sup>a</sup>
<i>Saccharomyces cerevisiae</i> H02	86.88 ± 1.27 <sup>b</sup>
<i>Saccharomyces cerevisiae</i> H03	0 ± 0.00 <sup>a</sup>
<i>Candida guilliermondii</i> H04	16.67 ± 0.72 <sup>c</sup>
<i>Candida guilliermondii</i> H05	16.67 ± 0.93 <sup>c</sup>
<i>Rhodotorula glutinis</i> H06	6.67 ± 0.24 <sup>d</sup>
<i>Candida guilliermondii</i> H07	0 ± 0.00 <sup>a</sup>
<i>Candida sp.</i> H08	0 ± 0.00 <sup>a</sup>
<i>Candida borealis</i> H09	27.33 ± 2.03 <sup>bc</sup>
<i>Candida spandorvansis</i> H10	0 ± 0.00 <sup>a</sup>
<i>Candida spandorvansis</i> H11	0 ± 0.00 <sup>a</sup>
<i>Candida tropicalis</i> H12	0 ± 0.00 <sup>a</sup>
<i>Candida tropicalis</i> H13	16.67 ± 1.14 <sup>c</sup>
<i>Rhodotorula glutinis</i> H14	0 ± 0.00 <sup>a</sup>
<i>Rhodotorula bogoriensis</i> H15	24.20 ± 0.87 <sup>bc</sup>
<i>Rhodotorula bogoriensis</i> H16	0 ± 0.00 <sup>a</sup>
<i>Candida acuta</i> H17	0 ± 0.00 <sup>a</sup>
<i>Candida acuta</i> H18	0 ± 0.00 <sup>a</sup>
<i>Candida acuta</i> H19	0 ± 0.00 <sup>a</sup>
<i>Candida acuta</i> H20	8.11 ± 0.45 <sup>d</sup>
Control	0 ± 0.00 <sup>a</sup>

Key: %E<sub>i</sub> = percentage emulsification index; control = distilled water; ± Se = standard error; the attached letters signifies significant difference (p < 0.05). Numbers bearing same letters within rows are not significantly different.

\*Code assigned to yeasts in our laboratory

## DISCUSSION

Saravana and Vijayakumar (2012) screened 243 and 802 isolates from oil contaminated soil samples using oil drop collapse for biosurfactant production and only 10 (4.12 %) and 40 (4.99 %) respectively gave positive result. These differences in results may be attributed to the physiological characteristics of the organisms as well as their genetic and molecular composition. Several researchers (Satpute *et al.*, 2008; Thavasi, *et al.*, 2010; Jain *et al.*, 2011; Tarango *et al.*, 2012; Ibrahim *et al.*, 2013; Pereira *et al.*, 2013; Padmapriya *et al.*, 2013) have proved the oil drop collapse method to be highly sensitive, very effective, and reliable in identification of potent biosurfactant producers

In a study conducted by Chandran and Das (2011), biosurfactants produced by *Rhodotorula muciliginosa* and *Candida rugosa* could effectively emulsify (86 % and 78 %) diesel oil respectively. Other researchers have reported similar results (Chander *et al.*, 2012; Padmapriya *et al.*, 2013).

## CONCLUSIONS

*Saccharomyces cerevisiae* H02, *Candida borealis* H09 and *Rhodotorula bogoriensis* H15 were considered efficient potential candidates for biosurfactants production

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