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Statistical Optimization of Temperature, Concentration, RPM and pH for the Surface Tension of Biosurfactant by *Achromobacter* Xylos GSR21

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Authors' contributions

Authors BSU, GSR and MMR conceived of the presented idea. Authors BSU, GSR and MMR developed the theory and performed the computations. Authors BSU, GSR and MMR verified the analytical methods. Authors MMR encouraged authors BSU and GSR to investigate [statistical optimization of temperature, concentration, RPM and pH for the surface tension of biosurfactant by Achromobacter xylos GSR21] and supervised the findings of this work. All authors discussed the results and contributed to the final manuscript.

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ABSTRACT

Achromobacter xylos strain GSR21 plays a crucial role in bioremediation of fossil fuel contamination, biopharmaceutical, cosmetics, chemical, petroleum refining, petrochemical, food industries and tertiary oil recovery (Microbial enhanced oil recovery). Response surface quadratic models (RSQM) was applied to reinforce the censorious operating conditions for the assembly of Achromobacter xylos strain GSR21. The Response surface method (RSM) was application to determine the best degrees of cycle factors (Temperature, Concentration, RPM, pH). Central composite design (CCD) of RSM was used to contemplate the four factors at five levels, and strain GSR21 Achromobacter xylos fixation was approximate as a reaction. Relapse coefficients predicted by examination and therefore the model was settled. R^2 value regard for bio-surfactant

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(mN/m) attempted to be 0.81, showing that the model fitted well with the explorative results. The mathematical model predicted by simulation of the foreseen updated values, and bio-surfactant surface tension was found 50 mN/m. The foreseen model was matched at 98.8% with the test outcomes coordinated under the perfect conditions. Based on the finding research, temperature-40°C, concentration-1.8 g/l, RPM-180 rev/mint and pH-4 was perceived as compelling fragments for *Achromobacter xylos* GSR21.

Keywords: Achromobacter xylos GSR21; response surface methodology; central composite design.

1. INTRODUCTION

Achromobacter xylos GSR21 are amphiphilic intensify present in living surfaces, for the first part on microbial cell surfaces or delivered extracellular hydrophobic and hydrophilic moieties that present the adaptability to amass between liquid stages, from now on the diminishing surface and interfacial bear the surface and interface separately [1-5]. They need the name of diminishing the face and interfacial strain utilizing similar instruments as produced blends surfactants. Surfactants are the dynamic decorations found in synthetic compounds and synthetic substances with the adaptability to assemble at the air-water interface and are typically wont to isolate smooth materials from a particular media. So they will build fluid dissolvability of Non-Fluid Phase Liquids (NAPLS) by lessening their surface/interfacial suffer air–water parcels oil interfaces [6-10]. *Achromobacter xylos* GSR21 are on a necessary level portrayed by their substance structure and their microbial inception. The standard classes of *Achromobacter xylos* GSR21 are glycolipids, phospholipids, polymeric biosurfactants and lipopeptides (surfactin) [10-15]. The preeminent standard glycolipids are rhamnolipids, sophorolipids and trehalolipids [16-21]. Surfactants are broadly utilized for the present day, developing, food, beautifiers and medications application regardless by a wide margin a large portion of those mixes are blended misleadingly and perhaps cause organic and toxicology issue because of the unmanageable and persevering nature of those substances [22-29]. With current advances in biotechnology are the thought to the choice great cycle for assembly of different kinds of biosurfactants from microorganisms [28-34].

The objective of the present paper is to estimate the best operating conditions of *Achromobacter xylos* strain GSR21 using response surface quadratic model.

2. MATERIALS AND METHODS

2.1 Microorganism

The microorganism *Achromobacter xylos* GSR21 used in this examination was gotten from Biochemical designing Laboratory culture assortment of the Biotechnology Department at Koneru Lakshmaiah Education Foundation, Andhrapradesh, India. The way of life is kept out in LB agar plates hatched at 37°C and sub-refined at normal's spans. Inoculums was set up by moving a loopful of culture to 100 mL of cleaned Luria Bertani (LB) stock and kept in rotational shaker hatchery at 200 rpm at 30 and 35°C for 48 h. All the synthetic substances utilized in the examination are of systematic evaluation and obtained from Quality-control, India.

2.2 Experimental Design

Four medium factors (Temperature, concentration, rpm, pH) were chose for Response surface methodology [5, 6, 30] improvement considers upheld starter screening contemplates. The fourth level scope was in Table 1. Thirty investigations were managed steady with a focal composite plan (Central Composite Design) appeared in Table 2.The correlation between the factors and thusly the reaction is generally speak the continuously arrange polynomial condition (Eqn. 1).

3. RESULTS AND DISCUSSION

3.1 Surface Tension Optimization Using Response Surface Methodology

Statistical optimization for biosurfactant surface tension was carried out according to the central composite design of RSM using Design expert software. The response, biosurfactant surface tension was estimated for thirty experiments and

$$Y = \alpha_0 + \alpha_1 X_1 + \alpha_2 X_2 + \alpha_3 X_3 + \alpha_4 X_4 + \alpha_{11} X_1^2 + \alpha_{22} X_2^2 + \alpha_{33} X_3^2 + \alpha_{44} X_4^2 + \alpha_{12} X_1 X_2 + \alpha_{13} X_1 X_3 + \alpha_{14} X_1 X_4 + \alpha_{23} X_2 X_3 + \alpha_{24} X_2 X_4 + \alpha_{34} X_3 X_4 \quad (1)$$

represented in Table.1.The response data were subjected to regression analysis to estimate the regression coefficient. The estimated coefficients were presented in (Table. 2). Final Equation in Terms of Coded Factors and Final Equation in Terms of Actual Factors for biosurfactant production was constructed by using the coefficients.

3.2 Final Equation in Terms of Actual Factors

The Model F-value of 2.82 implies the model is significant. There is only a 2.47% chance that a "Model F-Value" this large could occur due to noise. Values of "Prob > F" less than 0.0500 indicate model terms are significant. The result showed that B^2 was significant model terms. Values greater than 0.1000 indicate the model terms are not significant. If there are many not-significant model terms (not counting those required to support hierarchy), model reduction may improve your model.

The "Lack of Fit F-value" of 27.85 implies the Lack of Fit is significant. There is only a 0.01% chance that a "Lack of Fit F-value" this large could occur due to noise [5-9,28-30].

$$Surface\ tension\left(\frac{mN}{m}\right) = +8.04XA - 9.97XB + 46.55XC + 4.98XD + 50.41XAXB - 92.91XAXC - 7.42XAXD + 73.34XBXC + 86.06XBXD - 42.79XCXD + 1.58XA^2 + 80.27XB^2 + 110.62XC^2 + 44.24XD^2 \quad (2)$$

Table 1. Central composite design matrix with experimental values of bio-surfactant produced from achromobacter xylos strain GSR21

Run	Factor 1 A : Temperature (^{0}C)	Factor 2 B:Concentration (g/l)	Factor 3 C:RPM	Factor 4 D : pH	Response 1 Surface Tension (mN/m)
1	45	1.4	135	2.5	51
2	45	1.4	135	5.5	78
3	45	1	90	7	56
4	45	1.4	135	5.5	89
5	45	0.6	135	5.5	79
6	45	1.4	135	5.5	87
7	45	1.4	135	5.5	78
8	45	1.8	45	7	56
9	45	1.4	180	8.5	53
10	45	1.8	135	7	91
11	45	1.8	180	4	98
12	45	1.8	135	7	67
13	45	1.4	180	5.5	61
14	45	1	90	4	59
15	45	1	90	4	95
16	45	1	225	4	79
17	45	1	90	7	85
18	45	1.4	180	5.5	96
19	45	1.8	135	4	55
20	45	1.4	90	5.5	71
21	45	1.8	180	7	77
22	45	1	135	4	68
23	45	1.4	90	5.5	70
24	45	1.8	180	4	50
25	45	1.4	135	5.5	92
26	45	1.8	180	4	72
27	45	2.2	135	5.5	83
28	45	1	180	7	64
29	45	1.4	135	5.5	54
30	45	1	90	7	62

Table 2. ANOVA statistics for bio-surfactant production from achromobacter xylos GSR21

Source	Sum of squares	Df	Mean square	F-value	p-value	Significance
Model	1.170E+005	14	8353.97	2.82	0.0247	significant
A-Temperature	338.95	1	338.95	0.11	0.7394	
B-Concentration	280.78	1	280.78	0.095	0.7620	
C-RPM	3561.43	1	3561.43	1.20	0.2887	
D-pH	59.15	1	59.15	0.020	0.8893	
AB	1636.31	1	1636.31	0.55	0.4678	
AC	3905.33	1	3905.33	1.32	0.2674	
AD	55.53	1	55.53	0.019	0.8927	
BC	2612.52	1	2612.52	0.88	0.3613	
BD	2207.26	1	2207.26	0.75	0.4004	
COD	584.63	1	584.63	0.20	0.6626	
A^2	8.60	1	8.60	2.909E-003	0.9577	
B^2	14182.28	1	14182.28	4.80	0.0437	
C^2	7923.95	1	7923.95	2.68	0.1212	
D^2	3267.28	1	3267.28	1.10	0.3088	
Residual	47320.49	16	2957.53			
Lack of Fit	46034.69	9	5114.97	27.85	0.0001	significant
Pure Error	1285.80	7	183.69			
Total	1.643E+005	30				

Significant lack of fit is bad -- we want the model to fit.

Std. dev	54.38	R-squared	0.8119
Mean	72.53	Adj R-Squared	0.4599
C.V. %	74.98	Pred R-Square	0.1774
PRESS	1.351E+005	Adeq Precision	2.912

The "Pred R-Squared" of 0.1774 is not as close to the "Adj R-Squared" of 0.4599 as one might most expected. It may indicate a large block effect or a possible problem with your model and/or data. Things to consider are model reduction, response transformation, outliers, *etc*. "Adeq Precision" measures the signal to noise ratio. A ratio of 2.91 indicates an inadequate signal. We should not use this model to navigate the design space.

Fig. 1 showed that observed that the surface tension of biosurfactant decreased when the temperature increased from low to a high level stating that 20°C is sufficient for optimum productivity, whereas the productivity increased when the concentration of *Achromobacter xylos* increased from low to high level because intermolecular interaction is very high [8-15,22-30].

Fig. 2 showed that biosurfactant surface tension was decreased when the impeller speed (rpm) increased from low to high whereas static condition is prevailed in temperature indicating the contribution for biosurfactant surface tension by temperature is minimum. It is showed that the surface tension of biosurfactant decreased when the temperature and pH of biosurfactant increased from low to high (Fig. 3).

Fig. 4 showed that the biosurfactant surface tension was decreased when concentration of biosurfactant increased from low to a high. Whereas static condition is prevailed in rpm indicating the contribution for biosurfactant surface tension by concentration is minimum. It is observed that the surface tension of biosurfactant decreased when the concentration and pH of biosurfactant increased from low to high (Fig. 5).

$$\text{Surface Tension} = +4.64393\ \text{X Temperature} - 142.59204\ \text{X Concentration} + 0.39536\ \text{XRPM} + 13.27741\ \text{X pH} - 0.15390\ \text{X Temperature X Concentration} - 0.015931\ \text{X Temperature X RPM} + 0.056945\ \text{X Temperature X pH} + 0.31302\ \text{X Concentration X RPM} + 17.70483\ \text{X Concentration X pH} - 0.069394\text{X RPM X pH} - 0.028214\ \text{X Temperature}^2 + 4.46937\ \text{X concentration}^2 + 1.41678\text{E} - 003 * \text{RPM}^2 - 2.82865 * \text{pH}^2 \quad (3)$$

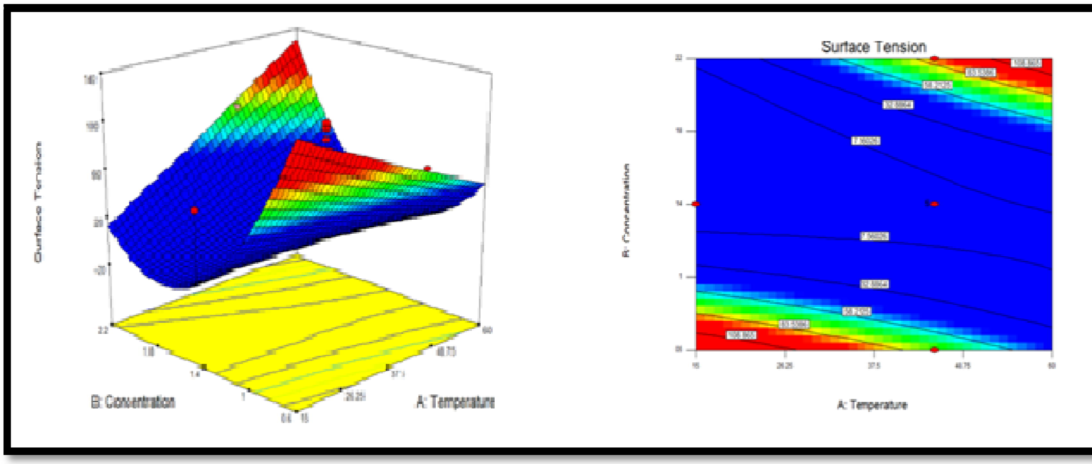

Fig 1. 3D and contour surface plots showing the mutual effect between pair of variables temperature (A) and Concentration (B) on biosurfactant surface tension

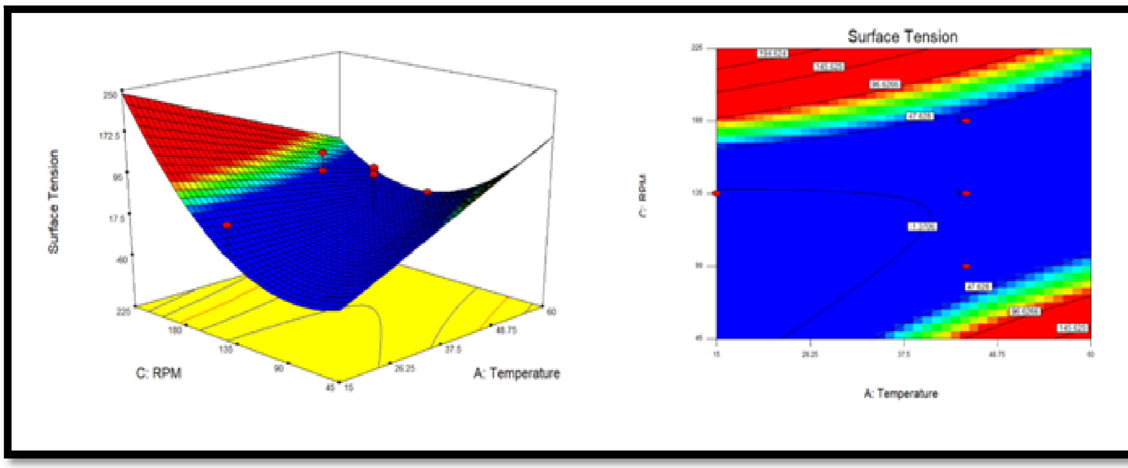

Fig 2. 3D and contour surface plots showing the mutual effect between pair of variables temperature (A) and RPM (C) on biosurfactant surface tension

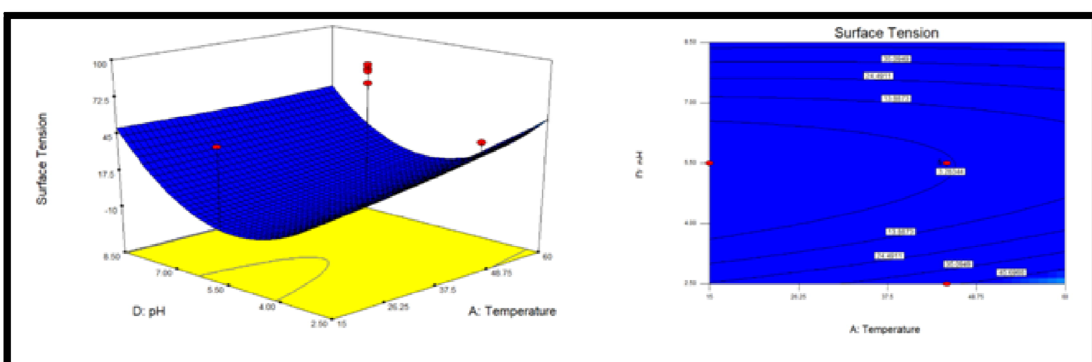

Fig. 3. 3D and contour surface plots showing the mutual effect between pair of variables temperature (A) and pH (D) on biosurfactant surface tension

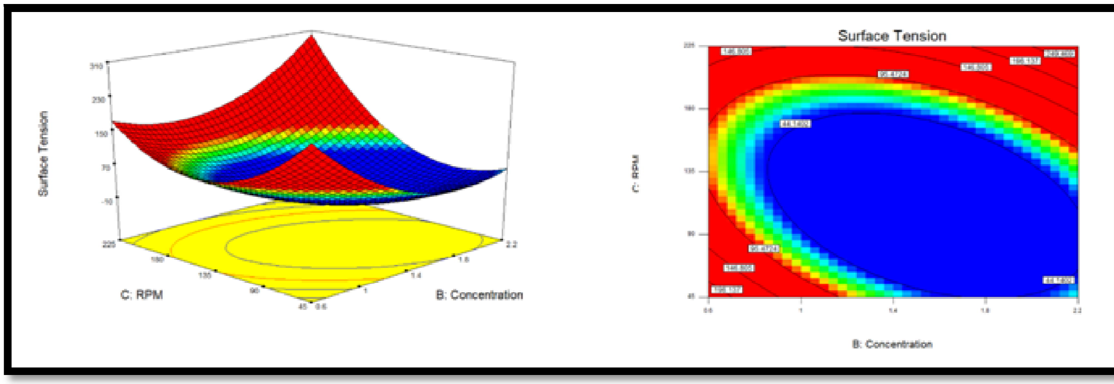

Fig. 4. 3D and contour surface plots showing the mutual effect between pair of variables concentration (B) and rpm (C) on biosurfactant surface tension

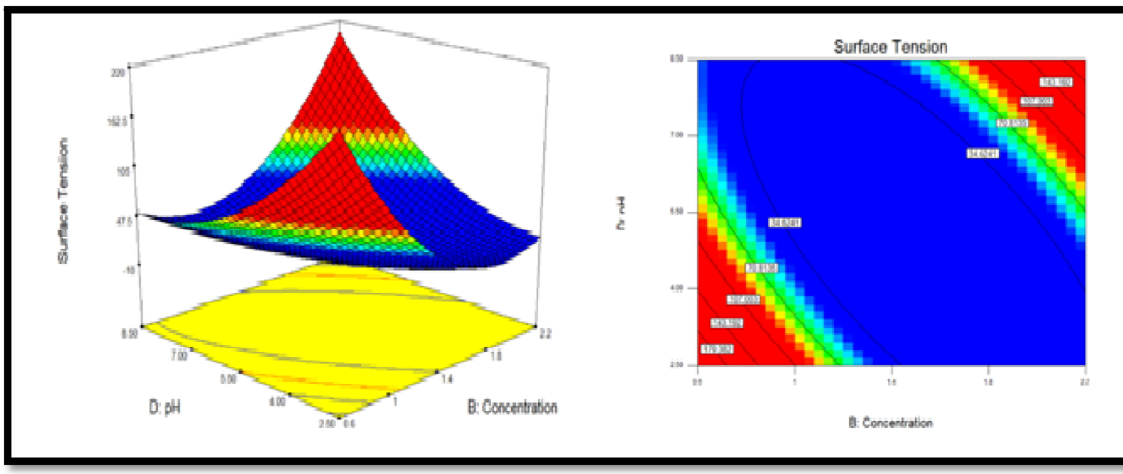

Fig. 5. 3D and contour surface plots showing the mutual effect between pair of variables concentration (B) and pH (D) on biosurfactant surface tension

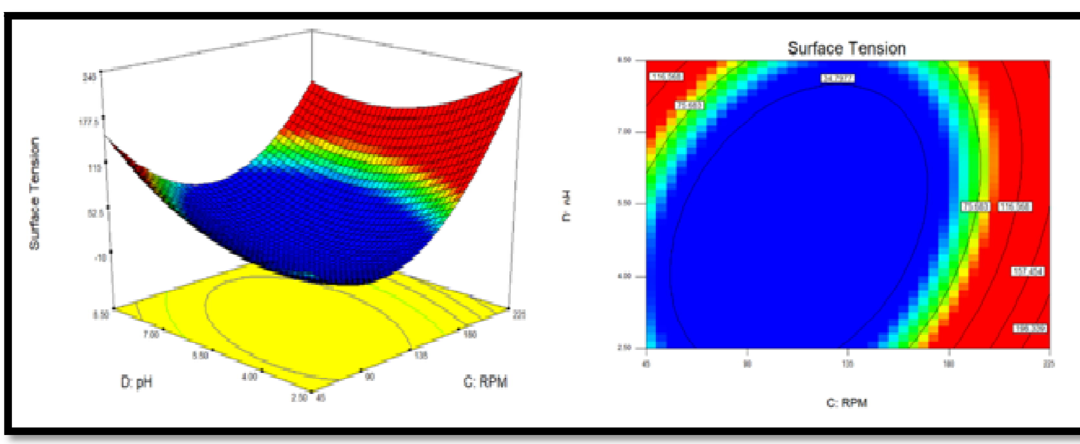

Fig. 6. 3D and contour surface plots showing the mutual effect between pair of variables rpm (C) and pH (D) on biosurfactant surface tension

Fig. 6 showed that the surface tension of biosurfactant decreased when the rpm increased from low to a high level stating that 45 is sufficient for optimum productivity, whereas the surface tension decreased when the concentration of *achromobacter xylos* increased from low to high level.

4. CONCLUSION

Response surface methodology successfully applied to optimize the four factors to enhance the biosurfactant surface tension. Temperature, concentration, rpm, pH were optimized, according to central composite design of RSM. The surface plots and the optimized values obtained. The minimum surface tension of biosurfactant was temperature-45^{o}C, concentration-1.8g/l, RPM-180 rev/mint and pH-4. The model was well fitted with the experimental results. Application of RSM defined as the optimal levels for enhanced production of biosurfactant with less experimental runs and interaction effects of the variables.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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