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COMPARISON OF APPLICATION OF RESPONSE SURFACE METHODOLOGY AND TAGUCHI METHOD, IN THE OPTIMISATION OF EXTRACTION PARAMETERS, FOR PRODUCTION OF BIOPIGMENT FROM A NEW ISOLATE OF DISTILLERY EFFLUENT

MABEL RACHEL JACOB[1], VARSHAKUMARI[2] & ELIZABETH AMUDHINI STEPHEN[3]

[1 & 2] Research Scholar, Programme of Food Processing & Engineering, Department of Biosciences & Technology, Karunya University, Coimbatore, Tamil Nadu, India

[3]Professor, Department of Mathematics, Karunya University, Coimbatore, Tamil Nadu, India

ABSTRACT

This study was done to determine the apt optimization tool, for a particular experiment that is to find out the effects of factors like temperature, solvent type and concentration and extraction time on pigment obtained from *Planococcusmaritimus*AHJ_2. As per single factor experiments and L16 orthogonal multiple factors design of experiment, the optimum conditions were identified with respect to maximum pigment yield obtained. Methanol was identified as the suitable solvent at 80% concentration and optimum temperature is 80˚C, with an optimum extraction time of 10 minutes while according to response surface method, optimum extraction would happen with methanol at 55% concentration, in 9.06 minutes at 75˚C. But, the bio pigment yield that would be obtained according to response surface methodology is lower than that according to taguchi method.

KEYWORDS: *Planococcusmaritimus*ahj_2, Response Surface Methodology, Single Factor Experiments & Orthogonal Multiple Factors Design of Experiment

INTRODUCTION

Since ancient times, colorants obtained from natural sources, ores, insects, plants, animals and microorganisms were in use. But, due to the difficulty and expensive methods involved in its production, the place of natural pigments was slowly being overtaken by the synthetic pigments, which are mostly produced by chemical methods which are being proved to be harmful in recent studies (Koes et al., 1994). Advances in organic chemistry have enabled easy and cheap production of synthetic dyes, and current applications of such dyes are in textile industry, leather industry, food technology, cosmetics, paper production etc. (Chidambaram et al., 2013). Recently, awareness about harmful effects of synthetic dyes is being spread among the consumers and natural pigments are gaining back their importance, in today's world (Krishnamurthy et al., 2002). Many natural pigments have a wide range of pharmacological effects, while most of the synthetic dyes are being identified to have carcinogenic effects. Also, bio pigments are found to bear many biological activities, such as antioxidant activities. Thus, bio pigments prove to be best and further development and optimisation of technology will definitely help in better, easier and cheaper production of these pigments.

Bio pigments are sourced from bacteria, algae, fungi, plants and animals. Microbial cultures are identified to bear a huge potential for improving the production of bio pigments such as carotenoids, flavonoids, quinones, rubramines etc. Pigmented bacteria can be identified and isolated from various environmental sources. But, the synthetic medium to be used for microbial pigment production may be costly. Agro-industrial residues are found to be an effective substrate for bio pigment production at low cost. Then, the pigment may be extracted by solvent extraction and then characterized by instrumental-based analytical techniques such as UV-Vis, FTIR etc. (Chidambaram et al.,2013). Also, biosynthetic pathways of pigments can be manipulated to develop the desired molecular structure of pigment and consequently its color. For example, *Streptomyces galilaeus*ATCC 31133 and ATCC 31671 which are used for production of anthracyclinesaclacinomycin A and 2-hydroxyaklavinone, produced an anthraquinone, aleosaponarin 2, when it was genetically cloned using the DNA of species *Streptomyces coelicolor (*Bartel PL et al., 1990). So, the advantages of microbial production of bio pigments are huge. Food grade pigments obtained from microbial sources are available in the market now, such as color from *Monascus sp.* Astaxanthin from *Xanthophyllomycesdendrorhous*etc. (Kim et al., 1997)

Optimisation of various parameters for production of pigments will help in reducing the cost and also aid in enhancing the pigment yield (Parekh S et al., 2000). So, single factor experiments are done to identify the optimum parameters and multiple factors design of experiments are also employed. Medium optimization is one important process to for getting maximum yield which includes various factors such as medium composition, pH, temperature, acidity etc. One factor at a time optimization will be time consuming because it involves many experimental runs but will be used for obtaining accurate results. Response surface methodology (RSM) and taguchi method are two important tools, used for medium and process optimization.

Response Surface Methodology

It is a collection of statistical techniques for designing experiments, evaluating the effect of various factors and finding optimum conditions (Xiong et al., 2004). In this method the relation of various factors is shown by a quadratic equation. RSM would solve multivariate data which may be found from appropriately designed experiments and solve multivariate equation (Gharibzahedi SMT et al; 2012). Canthaxanthin production by *D. natronolimnaea*from cheese whey was optimized using statistical experimental methods (Khodaiyan et al., 2008).

Taguchi Method

Taguchi method is employed for the determination of optimum factors for pigment production from microbial sources. Optimization of conditions for submerged culture fermentation of *Monascus spp.* to produce a high yield of monacolin K, was done using taguchi method. By using orthogonal arrays, it helps in reducing number of experiment trials in an effective way. It helps to optimize various processes that involve multiple factors (C. C Chung et al., 2006).

This study is to determine the effective tool among Response surface method and taguchi method, to optimize the production of bio pigment from *Planococcusmaritimus*AHJ_2. Taguchi method (L16 orthogonal multiple factor experiments) following single factor experiments were utilised by Varsha M. Chaudhari., 2013, to identify the optimum conditions for the extraction of pigment. A comparison of the results obtained using response surface methodology is done against the optimum conditions, which are identified by taguchi method, in this particular study.

METHODOLOGY

Method of Extraction and Statistical Method for Optimization

Extraction of pigment from *Planococcusmaritimus*AHJ_2 was done by submerged fermentation. Single factor experiments were done to identify the conditions at which maximum pigment yield could be obtained followed by multiple factor L16 orthogonal design of experiment to determine the optimum conditions (Varsha M. Choudhary., 2013).

Varsha.,2013 determined that maximum pigment yield was obtained when methanol was used as the solvent at concentration of 80% in an extraction time of 10 minutes at 80˚C using single factor experiment and then, by L16 orthogonal method and range analysis.

Change of Statistical Tool

The results obtained from single factor experiments were evaluated using central composite design combined with response surface analysis to identify which tool is effective in determining the optimum conditions for maximum pigment production. Solvent type chosen is methanol. The analysis was done with following conditions:

Sl. No.	PARAMETER	UNIT	RANGE
1.	Temperature	degree C	60-90
2.	Extraction time	Minutes	5-20
3.	Solvent concentration	%	60-90

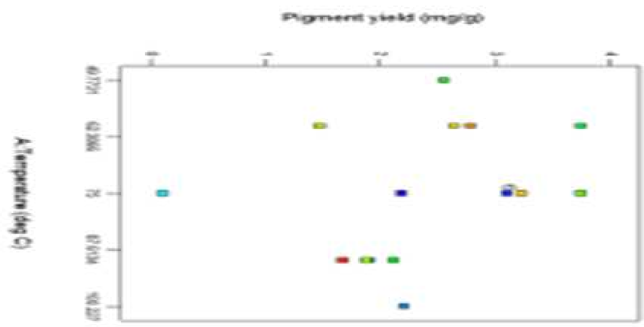
RESULTS & DISCUSSIONS

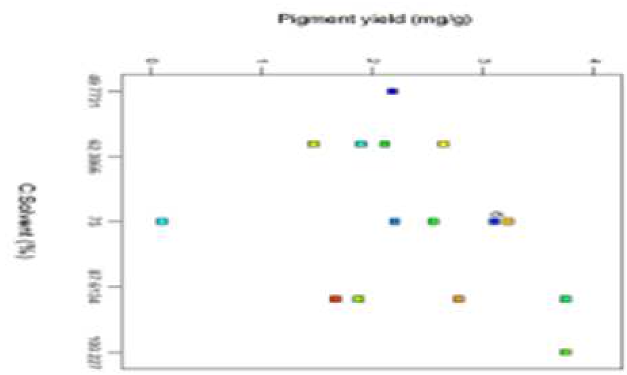
According to single factor experiment and L16 Orthogonal experiments, the maximum pigment yield obtained was 5.04 mg/g with methanol being used as the solvent at a concentration of 80% and extraction being done at 80˚C, in 10 minutes (Varsha M. Choudhary., 2013).

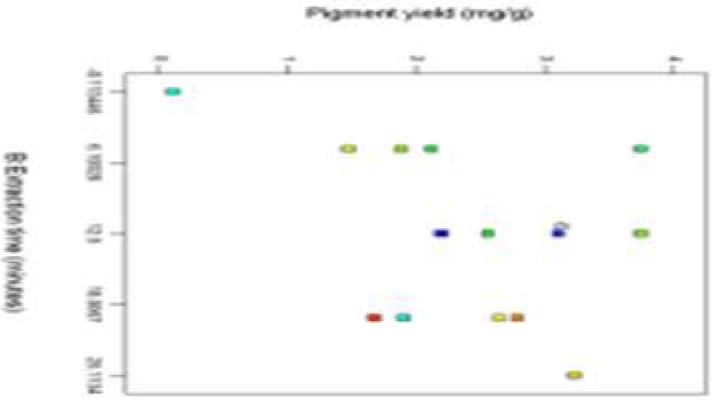
According to central composite design combined with response surface analysis, the relation of parameters and the multivariate data is given by the following equation:

Pigment yield = $+3.10-0.27*A+0.37*B+0.33*C-0.079*AB-0.36*AC-0.27*BC-0.26*A^2-0.51*B^2-0.052*C^2$

Where, A-Temperature, B – Extraction time, C – Solvent concentration.







According to central composite design combined with response surface analysis, the maximum yield that could be obtained is 2.79 mg/g at 75˚C, in an extraction time of 9.06 minutes with solvent concentration being 55%.

CONCLUSIONS

Both response surface method and taguchi method are often used, for optimization of production of pigments. Response surface helps to identify the optimum conditions using multivariate data and taguchi method identifies the optimum conditions, by considering factors over certain levels. Single factor experiments are necessary to identify the effect of each factor on the pigment yield. L16 orthogonal method is found to be more effective than central composite design, combined with response surface analysis for this particular experiment.

REFERENCES

1. Bartel PL, Zhu CB, Lampel JS, Dosch DC, Connors NC, Strohl WR, Beale JM Jr, Floss HG, Bio-synthesis of anthraquinones by interspecies cloning of actinorhodin biosynthesis genes in Streptomyces: clarification of actinorhodin gene functions. J Bacteriol, 1990; 172(9): 4816-26.
2. Chidambaram KulandaisamyVenil, ZainulAkmarZakaria, Wan Azlina Ahmad, Bacterial pigments and their applications. Process Biochemistry, 2013; 48:1065-1079.
3. C. C Chung, H. H Chen, P. C Hsieh, Application of the taguchi method to optimize *Monascus spp.* Culture. Journal of Food Process Engineering, 2007; 30(2):241-254.
4. Parekh S, Vinci V, Strobel RJ. Improvement of microbial strains and fermen-tation processes. ApplMicrobiolBiotechnol. 2000; 54:287–301.
5. Gharibzahedi SMT, Mousavi SM, Hamedi M, Khodaiyan F, Razavi SH. Devel-opment of an optimal formulation for oxidative stability of walnut-beverage emulsions based on gum arabic and xanthan gum using response surfacemethodology. CarbohydrPolym.2012; 87:1611–9.
6. Khodaiyan F, Razavi SH, Mousavi SM. Optimization of canthaxanthin pro-duction by Dietzianatronolimnaea HS-1 from cheese whey using statisticalexperimental methods. BiochemEng J. 2008;40:415–22.
7. Kim SW, Seo WT, Park YH, Enhanced production of carotene from *Blakesleatrispora*with span 20. Biotechnol. Lett., 1997; 19: 561-562.

8. Koes REK, Quattrocchio F, Mol JNM. The flavonoid biosynthetic pathway inplants: function and evolution. Bioassays, 1994; 16:123–32.

9. Krishnamurthy KV, Siva R, Senthil TK. Natural dye-yielding plants of Sher-varoy Hills of Eastern Ghats. In: Proceedings of National Seminar on theConservation of the Eastern Ghats. 2002; 151–3.

10. Varsha M. Choudhary. Optimization of the extraction parameters for the production of biopigment from the new isolate of distillery effluent. Journal of Scientific and Innovative Research, 2013; 2(6): 1044-1051.

11. Xiong, Y. H., Liu, J. Z., Song, H. Y., and Ji, L. N. Enhanced production of extracellular ribonuclease from Aspergillus niger by optimization of culture conditions using response surface methodology. Biochemical Engineering Journal, 2004; 21: 27–32.