TEXTILE DYING USING RED PIGMENT PRODUCED BY FUNGI *MONASCUS RUBER* TIEGHEM IOC 2225

Diana Castro Diaz[1], Jesus Calderon Gamero[1] & Ruly Terán Hilares[1]*

[1] *Bioprocess Laboratory, Faculty of Pharmaceutical, Biochemical, and Biotechnological Sciences, Catholic University of Santa María (UCSM), Arequipa, Peru*
** Corresponding author's email address: rteran@ucsm.edu.pe*

ABSTRACT

The textile industry is known for its high levels of pollution, primarily due to the extensive use of synthetic pigments for dyeing fabrics. To mitigate the environmental impact of these dyes, sustainable alternatives are being explored, including natural pigments produced by microorganisms. One promising microorganism is the fungus *Monascus ruber*, which produces a variety of colors through its secondary metabolism. In this study, the production of red pigment by *Monascus ruber* was evaluated for its application in the textile industry. The pigment produced enabled efficient dyeing of acetate and polyester fibers. Thus, this study highlights a sustainable alternative to reduce the environmental impact of synthetic dyes widely used in the textile industry.

Keywords: Biotechnology, *Monascus ruber*, Fiber Dyeing, Environmental Sustainability.

1 INTRODUCTION

In the textile industry, synthetic pigments are extensively employed for dyeing various natural and synthetic fabrics. As one of the major consumers of artificial dyes worldwide [1], this industry is notorious for its high pollution levels, generating substantial amounts of liquid waste and requiring significant volumes of water, energy, and chemicals in its daily operations [2]. However, the continued use of these colorants and the challenges associated with their removal result in significant environmental impacts. Consequently, there has been a concerted effort to seek sustainable alternatives derived from natural sources, which offer comparable applicability with reduced environmental consequences [3]. In this way, natural pigments production by microorganisms (yeast, fungi, bacteria, and microalgae) is an attractive process due to the fast growing of microorganisms, high pigment productivity and possibility of using cheap substrates as carbon source. Moreover, microorganism cultivation is non-dependent on weather conditions as vegetable sources. Among the various alternatives [4], fungi emerge as promising producers of natural pigments. For instance, *Monascus ruber* can yield a diverse range of hues derived from various chemical structures generated by its secondary metabolism [5]. Thus, in this study, red pigment was produced in glucose based medium, recovered using solvent, concentrated by filtration, and subsequently utilized for textile dyeing.

2 MATERIAL & METHODS

Microorganisms, medium and spore production

In this study, a *Monascus ruber* Tieghem strain IOC 2225 was used. The microorganism was stored on PDA agar until needed. For spore production, the petri dishes were incubated at 28.5°C for 7 days. To recover the spores, 10 ml of sterilized distilled water was added to the agar plates. Subsequently, scraping was conducted across the entire plate, and the solution containing the spores was collected and utilized in the fermentation process.

Red pigment production

Fermentation was conducted using a medium consisting of glucose (20 g/L), peptone (2.5 g/L), $CaCl_2$ (0.1 g/L), and KH_2PO_4 (5g/L). Each fermentation was carried out in 100 ml of medium contained in 125 ml Erlenmeyer flasks. The medium was sterilized in an autoclave at 121°C, 15 psi, for 15 minutes. Subsequently, the medium was inoculated with spores (1 mL) to achieve a concentration of 3×10^6 spores/mL. The flasks were then placed in incubators set to specific conditions: 30°C temperature, agitation at 150 rpm, pH maintained at 5, and left for 7-15 days. Samples were collected periodically to analyze sugar by DNS method and pigment production by spectrophotometer at 495 nm.

Concentration of pigment using polymeric membranes

After the fermentation process (15 days), the liquid fraction was recovered by filtration using Whatman No. 4 filter paper. The filter paper was weighed, folded, and placed in a funnel to slowly add the culture medium. Once the liquid culture medium was filtered, the filter paper with the recovered biomass was removed and dried at 50°C overnight. The filtered liquid containing pigment was then subjected to concentration via nanofiltration using a Sterlitech HP4750 High Pressure Stirred Cell (USA). A TriSet flat sheet membrane with a molecular cut-off weight of 150 Dalton was utilized for the concentration of pigment.

Pigment Extraction and textile dying

Once the pigment is filtered and concentrated, it can be extracted successively with ethyl acetate in a 1:1 v/v ratio. This extraction process is carried out in a separation funnel to separate the mixture into two phases. The ethyl acetate layer containing the pigments is then recovered and used for dyeing various textiles. To dye, the purified pigment extracted with ethyl acetate is employed. A certain volume of the pigment is placed in a beaker, where a sample of Multifiber 10 (Acetate, cotton, nylon, polyester, acrylic, wool) with dimensions of 2 x 2 fabric is submerged during 15 min at 100 °C. Subsequently, the fabric is washed four times with distilled water, vigorously agitating and changing the water after each wash.

3 RESULTS & DISCUSSION

The typical curve of substrate consumption and pigment production is shown in Figure 1A. As depicted in the figure, glucose consumption extends over a long period, with approximately 40% of the glucose being utilized. The microorganism can completely consume the carbon source depending on the type of sugar (fructose, maltose, lactose, and galactose) and the cultivation conditions [7]. The produced pigment exhibits an absorbance peak at 495 nm (Figure 1B), consistent with findings from other researchers [6]. Regarding the production of red pigment, over approximately 15 days, a production level of 8.5 AU was observed. The fermentation broth containing the red pigment was subjected to membrane concentration through ultrafiltration, aiming to increase the concentration for subsequent liquid extraction using solvent and application to textile dyeing, resulting in 972 AU at 493 nm.

Figure 3 provides a summary diagram of all the activities described. During the dyeing stage, it was found that the pigment was highly effective for dyeing acetate fiber (red) and acrylic (dark red), with a lighter red tone observed in wool. However, as seen in Figure 2G, the pigment is not suitable for dyeing textiles such as nylon and acrylic. Specific colors can only be achieved on certain fabrics without tints.

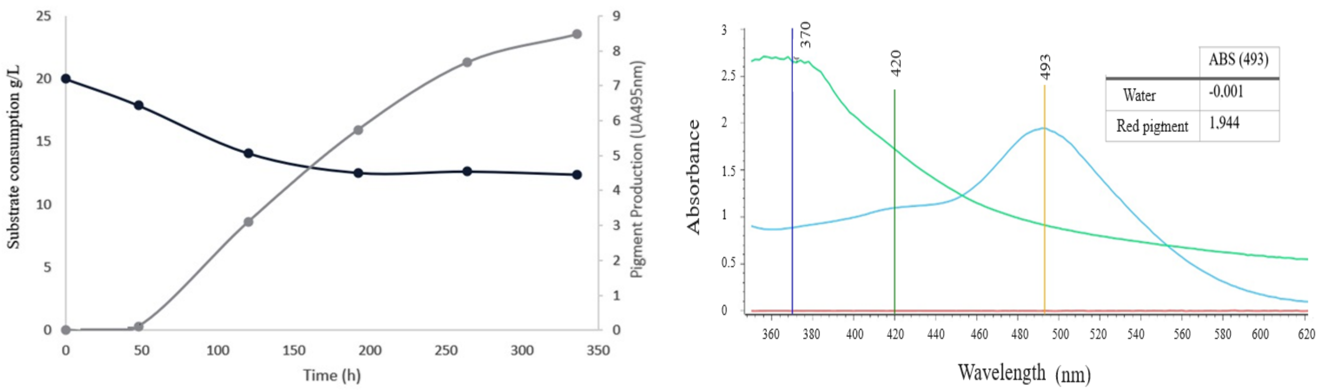

Figure 1 A) Substrate consumption and red pigment production and **B)** Absorption spectrum of red pigment in spectrophotometer.

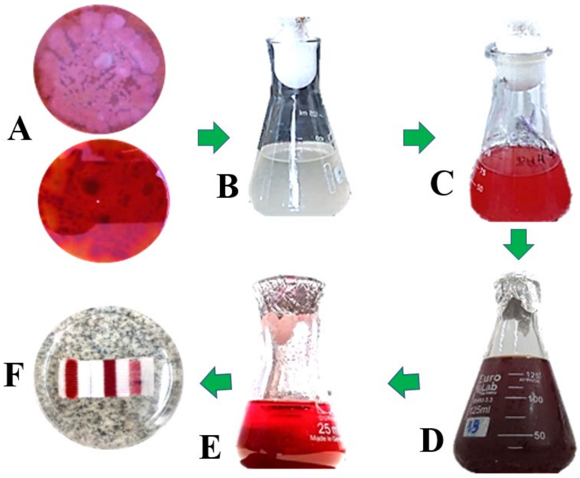

Figure 2 Scheme of the experimental process for pigment extraction. A. - Petri dish of *Monascus ruber* showing both sides; B. - Liquid culture medium inoculated with *Monascus ruber*; C. - Culture medium after 15 days of incubation at 30°C; D. - Filtered and concentrated culture medium; E. - Pigment extraction using ethyl acetate; F.- Dyeing of different fiber fabrics such as acetate, cotton, nylon, polyester, acrylic, wool.

4 CONCLUSION

The fungus produced a red pigment that can be used to dye synthetic fibers. Additionally, with the use of suitable mordants, it may be possible to use this pigment for dyeing natural fibers. The production of microbial pigments and their application in the textile industry can significantly contribute to reducing the environmental impact of synthetic dyes.

REFERENCES

1 ZARUMA, P. PROAL.J. HERNÁNDEZ, IC. SALAS, HI. RFCQ. 2018 RFCQ. 34 (19). 38–47.

2 KHELIFI, E. GANNOUN, H. TOUHAMI, Y. BOUALLAGUI, H. HAMDI, M. 2008 J Hazard Mater 152 (2) 683–9.

3 VELMURUGAN, P. KIM, M-J. PARK, J-S. KARTHIKEYAN, K. LAKSHMANAPERUMALSAMY, P. LEE, K-J. et al. 2010 Fiber Polym.11 (4) .598–605

4 ORONA-NAVAR, A. AGUILAR-HERNÁNDEZ, I. NIGAM, KDP. CERDÁN-PASARÁN, A. ORNELAS-SOTO, N. 2021. J Biotechnol. 332 (0168-1656) 29–53.

5 AGBOYIBOR, C. KONG, W-B. CHEN, D. ZHANG, A-M. NIU, S-Q. Biocatal Agric Biotechnol 16. (1878-8181).433–47

6 LIU, J. REN, Y. YAO, S. 2010. Chin J Chem Eng .18 (6). 1013–7.

7 ABDEL-RAHEAM, HEF. ALRUMMAN, SA. GADOW, SI. EL-SAYED, MH. HIKAL, DM. HESHAM, AE-L. et al. Front Microbiol. 2022.13(1664-302x)

8 ASHIS, KS. AWWAD, N. ALGARNI, HM. 2020. A practical guideline of few standardized ready-made shades of natural dye textiles. Chemistry and Technology of natural and Synthetic In: Dyes and pigments. ASHIS, KS. AWWAD, N. ALGARNI, HM. (ed). 1st ed. IntechOpen. Londen

ACKNOWLEDGEMENTS

This research was funded by the Universidad Católica de Santa María (UCSM) of Peru -Grant N° 7797-CU-2021.