



Anticorrosive Performance of Bacterial Eumelanin Polymer as a Novel Corrosion Inhibitor Doped into Hybrid Sol-Gel Matrix

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Abstract

Melanins are a group of organic polymer that widely distributed in nature from microorganisms to human. These ubiquitous pigments have many different properties such as antioxidant, antimicrobial, antivenin, anti-inflammatory, radioprotective, photoprotective etc. In this present study, it was aimed to determine the anticorrosion performance of eumelanin polymer purified from *Streptomyces parvus* BSB49 strain. For this purpose, hybrid sol-gel matrix was synthesized using tetraethyl orthosilicate and 3-aminopropyltriethoxysilane in order to incorporate eumelanin as a doping agent. In this process, mild steel plates with a composition of Si \leq 0.16%, Mn \leq 0.15%, Mg \leq 0.03%, Al \leq 0.06%, P \leq 0.01%, Na \leq 0.02%, and Ca \leq 0.01% were employed as the substrate to apply the coatings. A three-electrode cell setup loaded with 0.5 M HCl corrosion medium was used to investigate the corrosion behaviour of the coated samples. The corrosion rate reduced with the increase in doping concentration of eumelanin. Thus, the obtained results through electrochemical impedance spectroscopy and potentiodynamic polarization measurements revealed that the eumelanin pigment produced using *Streptomyces parvus* BSB49 strain has anticorrosive properties.

Keywords: Eumelanin, Biosynthesis, Anticorrosion, Impedance, Polarization.

Hibrit Sol-Jel Matrisine Katkılı Yeni Bir Korozyon İnhibitörü Olarak Bakteriyel Eumelanin Polimerinin Antikorozyon Performansı

Öz

Melaninler, doğada mikroorganizmalardan insanlara kadar çok çeşitli canlı gruplarında yaygın olarak dağılmış bir organik polimer grubudur. Neredeyse tüm canlı gruplarında bulunan bu pigmentler, antioksidan, antimikrobiyal, antivenin, antiinflamatuvar, radyoprotektif, fotokoruyucu vb. birçok farklı özelliğe sahiptir. Bu çalışmada, *Streptomyces parvus* BSB49 suşundan saflaştırılan eumelanin polimerinin korozyon önleyici performansının belirlenmesi amaçlanmıştır. Bu amaçla, eumelanini bir doping ajanı olarak dahil etmek için tetraetil ortosilikat ve 3-aminopropiltrioksilan kullanılarak hibrit sol-jel matrisi sentezlenmiştir. Bu işlemde Si \leq %0,16, Mn \leq %0,15, Mg \leq %0,03, Al \leq %0,06, P \leq %0,01, Na \leq %0,02 ve Ca \leq %0,01 bileşimine sahip yumuşak kaplamaları uygulamak

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için çelik levhalar kullanılmıştır. Substrat kaplanmış numunelerin korozyon davranışını araştırmak için 0,5 M HCl korozyon ortamı ile yüklenen üç elektrotlu bir hücre düzeni kullanılmıştır. Ömelaninin doping konsantrasyonundaki artışla korozyon hızı azalmıştır. Böylece elektrokimyasal empedans spektroskopisi ve potansiyodinamik polarizasyon ölçümleri ile elde edilen sonuçlar, *Streptomyces parvus* BSB49 suşu kullanılarak üretilen eumelanin pigmentinin antikorozyf özelliklere sahip olduğunu ortaya koymuştur.

Anahtar Kelimeler: Eumelanin, Biyosentez, Antikorozyon, Empedans, Polarizasyon.

1. Introduction

Melanins are a group of organic polymers that composed of phenolic and/or indolic compounds. These natural pigments are characteristically hydrophobic, negatively charged and high molecular weight. Melanins are divided into five different groups as eumelanin, pheomelanin, neuromelanin, allomelanin and pyomelanin [1, 2]. The most common groups of melanins in humans are black - brownish eumelanin and yellow - reddish pheomelanin pigments and these pigment groups are the determining factors in hair, eyes, and skin color [3, 4]. L-tyrosine amino acid is used as the precursor molecule in the synthesis of eumelanin in humans. This amino acid is first converted to L-3,4-dihydroxyphenylalanine (L-dopa) by the enzyme tyrosinase. In the next step, the resulting L-dopa molecule is converted to the highly reactive dopaquinone molecule. This biotransformation series then continues as cyclodopa, dopachrome, and dihydroxyindoles. The resulting dihydroxyindoles molecules polymerize to form the eumelanin polymer [5, 8]. In the pheomelanin pigment synthesis, the initial stages are similar to the of eumelanin synthesis. However, during this process, 2-S-cysteinyl-dopa is obtained by cysteinylolation of the dopaquinone molecule, and in the next process, 2-S-cysteinyl-dopa molecules are transformed into benzothiazine intermediate molecules. Finally, benzothiazine intermediate polymerizes to form pheomelanin [9, 10].

A third type of melanin found in humans is neuromelanin. Similar to other melanin groups, neuromelanins are hydrophobic and insoluble in water and it is known that the amount of neuromelanin in the substantia nigra in the human brain increases with age. In addition, it is considered that, thanks to its heavy metal chelating feature, it binds heavy metals reaching the central nervous system and thus indirectly assumes a preventive role against cytotoxic and genotoxic damage. However, contrary to this information, it is also stated that neuromelanin pigment accumulating in the substantia nigra with aging causes neurodegeneration and Parkinson's disease [11, 12].

Another group of melanin is allomelanin. Allomelanins are a nitrogen-free and highly complex structured member of the melanin family. It is stated that this pigment is generally produced by fungi by polymerization of different molecules such as dihydrofolate, homogentisic acid and catechols [13, 14]. The last member of the melanin family is pyomelanin, and this pigment is formed as a result of a series of enzymatic reactions after the breakdown of tyrosine or phenylalanine amino acids. In the process of synthesis of pyomelanin pigment by microorganisms, these amino acids are first broken down to acetoacetate and fumarate. 4-hydroxyphenylpyruvic acid dioxygenase (4-HPPD) and homogentisic acid oxidase (HGA-oxidase) enzymes are responsible for these degradation processes. In this process, in case of overproduction of homogentisic acid, accumulated homogentisic acid molecules outside the cell form pyomelanin polymers by autopolymerization [15-17].

Melanin pigments have many different bioactive properties. Thanks to their antioxidant, radioprotective and photoprotective

properties, melanins protect hereditary materials in living things against mutation and thus provide an advantage against adverse environmental conditions [18, 19]. The antimicrobial [20, 21], antiviral [22], antivenin [23], anti-inflammatory [24], liver protective [25], immunomodulatory [26], antiproliferative [27], thermoregulative [28] and chemoprotective effects [29] of these heterogeneous copolymer pigments are also given in the literature.

Due to these unique properties, melanins have the potential to be used in many industrial areas such as medicine, pharmacology, textile and packaging industry [13, 30-33]. In addition to these usage areas, melanins are an ideal natural product for the cosmetics industry and for the production of sunscreens, thanks to their ability to absorb UV rays and X-rays to a large extent. Lastly, due to being an organic semiconductor, melanin polymers will undoubtedly be one of the most important raw materials in the production of organic electronic materials in the future [34].

There are virtually little or no reports in literature on the utilization of eumelanin as a corrosion inhibitor for the corrosion protection of mild steel. In this study, it was aimed to determine the anticorrosive performance of eumelanin pigment produced by *Streptomyces parvus* BSB49 strain as a potent corrosion inhibitor through the doping into hybrid sol-gel matrix. Electrochemical impedance and potentiodynamic polarization measurements were employed to investigate the anticorrosion behaviour of eumelanin pigment.

2. Material and Method

2.1. Isolation of *Streptomyces* Strains and Determination of Melanin Pigment Production Properties

For the isolation of *Streptomyces* strains, soil samples taken from the rhizosphere were transferred to sterile 50 mL Falcon tubes and brought to the laboratory. Soil samples, which were left to dry for 48 hours at room temperature, were mixed with CaCO₃ (at a rate of 10:1 w/w) and kept in a humid atmosphere at 37°C for one week. After these pre-treatments, the soil samples were diluted with physiological saline. And then, 1 mL of the prepared dilutions was taken and transferred to ISP2 and ISP4 media [35]. At the end of these procedures, these diluted samples spread on the surface of the ISP4 medium with a drigalski loop. These prepared petri dishes were incubated at 25°C for two weeks. Different colonies observed on the 2nd, 4th, 7th and 14th days of the incubation period were transferred to petri dishes for isolation. These isolated *Streptomyces* strains were inoculated to a 1 cm² area in a petri dish and incubated at 36°C for 72 hours. Microorganisms that formed a dark black zone around the colony were considered positive in terms of melanin pigment production [27, 36].

2.2. Bioproduction and purification of eumelanin polymer

Eumelanin polymer biosynthesis was carried out as described by Bayram et al. (2020). Nutrient broth (NB, Merck, Germany) was used for the production of melanin pigment. For this purpose, 150 mL volume of NB in 250 mL flasks were sterilized in autoclave (Hirayama HV-50, Japan) at 121 °C for 15 min. After the sterilized NB was allowed to cool at room temperature, *Streptomyces parvus* BSB49 strain was inoculated. After the inoculation process, the samples were incubated for one week in an orbital shaker (Heidolph Unimax 1010, Schwabach, Germany) set at 35 °C and 200 rpm. At the end of the one-week incubation period, the dark black colored media were transferred to 50 mL Falcon tubes and centrifuged (Beckman Coulter Allegra X-30R, Brea, California, USA) at 10,000 rpm for 10 min. With these first centrifugation processes, the bacterial biomass was largely separated from the medium and the same processes were repeated three times in order to avoid residue. Bacterial biomass-free supernatant samples were adjusted to pH 2 using 6 M HCL (Merck, Germany) and allowed to polymerize at room temperature for 24 h. At the end of the 24 h polymerization period, the samples were centrifuged again at 10,000 rpm for 10 min. After centrifugation, the supernatant was carefully poured and the remaining melanin pellets at the bottom of the Falcon tubes were left to dry at 55 °C for 24 h [27, 36].

2.3. Synthesis of hybrid sol-gel matrix

Hybrid sol-gel coatings engaging APTES and TEOS alkoxy silane precursors were prepared according to the method adopted by Wu and co-workers (2017) with modifications [37]. Analytical grade ethanol functioned as the solvent to obtain a homogenized solution of alkoxy silane precursors. Diluted silane precursors and ethanol were mixed in a ratio of 3:2 (v/v), respectively. Upon combining the silane precursors and ethanol, the mixture was incorporated with 100 ppm and 1000 ppm concentrations of eumelanin. The pH of the solution was held at 3.0 to improve the rate of hydrolysis and condensation reactions. Next, the silanol matrix was constantly stirred at 30 ± 1 °C for 24 h to yield a translucent solution. Afterwards, the sol-gel formulation was subjected to aging process, which improves the coating properties [38].

2.4. Application of coatings

Mild steel plates with a composition of Si \leq 0.16%, Mn \leq 0.15%, Mg \leq 0.03%, Al \leq 0.06%, P \leq 0.01%, Na \leq 0.02%, and Ca \leq 0.01% was employed as the substrate to apply the coatings. Before the coating assembly, the plates were abraded with different grades of SiC papers (200, 400, 600 and 1000-grit). Specimens were degreased with acetone to eliminate surface residues. Dip-coating technique was applied to coat the sol-gel matrix on mild steel surface by dipping upright for 12 h to permit a homogeneous coating. After withdrawing the coated samples, the samples were dried in the air and subsequently in an oven at 105 °C for 15 min to warrant a thorough coating assembly. The average layer thickness of the assembled coatings averaged 3-5 μ m.

2.5. Analysis of corrosion performance

A three-electrode cell setup loaded with 0.5 M HCl corrosion medium was used to investigate the corrosion behaviour of the coated samples. Corrosion performance was examined by a Gamry Instruments Reference 600 Potentiostat/Galvanostat. Initially, the open circuit potential stabilization was accomplished

for 10 min to attain a steady-state potential. The cell setup was maintained at 30 °C throughout the corrosion study. Corrosion parameters were obtained by Gamry Echem Analyst software. Each experiment was triplicated to attain data with high accuracy and reproducibility.

2.6. Potentiodynamic polarization (PD) analysis

After each coated metal substrate has achieved a steady-state, a potential limit of ± 0.25 V vs. E_{ocp} was applied at a scan rate of 0.5 mV s^{-1} . Corrosion inhibition efficiencies (IE%) were determined based on Eq. (1) [39].

$$IE\% = \left(\frac{i_{corr} - i_{corr'}}{i_{corr}} \right) \times 100 \quad (1)$$

where i_{corr} and i_{corr}' refer to corrosion current densities (in mA cm^{-2}) with and without an inhibitor, respectively.

2.7. Electrochemical impedance spectroscopy (EIS) analysis

EIS analysis was conducted at a frequency range of 10^4 Hz to 10^{-1} Hz under AC excitation of a sinusoidal wave of 10 mV amplitude. The impedance data were acquired in the form of Nyquist plots. IE% values were determined with the use of R_{ct} values according to Eq. (2) [40].

$$IE\% = \left(1 - \frac{R_{ct}}{R_{ct}'} \right) \times 100 \quad (2)$$

where R_{ct} and R_{ct}' represent charge transfer resistance (in $\Omega \text{ cm}^2$) with and without an inhibitor, respectively.

3. Results and Discussion

3.1. PD analysis

Potentiodynamic polarization measurement reveals the electrochemical kinetics of the corrosion process. Fig. 1 represents the Tafel diagrams obtained during the PD study. Corrosion potential data were determined through the intersection of cathodic and anodic branches of the polarization curves using Tafel extrapolation method. In addition, table 1 outlines the polarization parameters of the bare mild steel and coated metal substrates.

In the present study, it was observed that E_{corr} values showed 33-37 mV increment for the eumelanin-doped sol-gel coated substrates over the bare mild steel. The shifting of E_{corr} of coated specimens to more positive values suggest improved corrosion resistance due to the inhibition of metal dissolution, i.e., the anodic reaction [41]. The corrosion rate (CR) of each electrode was determined based on Eq. (3) [42]. Hence, 1000 ppm eumelanin doped sol-gel coating recorded the lowest CR with 80.62% of inhibition efficiency.

$$\text{Corrosion rate (mpy)} = \frac{k \times i_{corr} \times M_{ew}}{\rho \times SA} \quad (3)$$

where i_{corr} expresses the current density in $\mu\text{A cm}^{-2}$, k represents a constant defining the units of the corrosion rate (0.1288 for m/yr), M_{ew} is the equivalent weight (g/equivalent), SA indicates the exposed surface area of mild steel in cm^2 and ρ indicates the

density of the metal in g cm^{-3} the incorporation of different concentrations of eumelanin at 30°C .

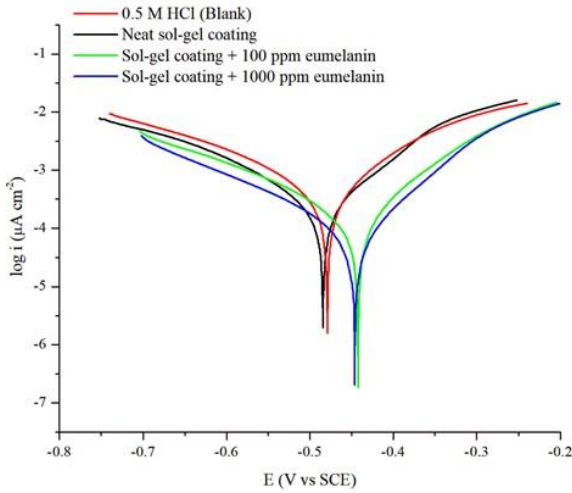


Figure 1. Tafel curves of coated mild steel specimens in 0.5 M HCl solution without and with the incorporation of different concentrations of eumelanin at 30°C .

3.2. EIS analysis

The electrochemical impedance spectroscopy analysis was carried out to assess the impact of eumelanin on the corrosion behaviour of hybrid sol-gel coated mild steel in 0.5 M HCl solution. Fig. 2 depicts the corresponding Nyquist plots obtained through the EIS experiments. The semi-circular shaped Nyquist diagrams acquired through EIS analysis denote the nature of the corrosion process on mild steel is charge transfer controlled.

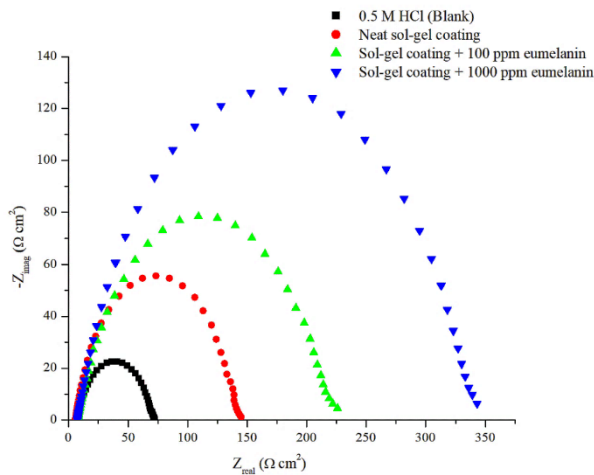


Figure 2. Nyquist plots of coated mild steel specimens in 0.5 M HCl solution without and with the incorporation of different concentrations of eumelanin at 30°C .

The electrical equivalent circuit (Fig. 3b) adopted to fit the electrochemical impedance spectroscopy data follows previous studies for sol-gel coated Fe metal/HCl solution interface [43-46]. R_s is the solution resistance, R_{ct} represents the charge transfer resistance, whereas R_{coat} stands for the resistance of inhibitive coating. CPE_{dl} and CPE_{coat} symbolize the constant phase elements, where the deviation parameters for the CPE elements

are n and m , respectively. Table 2 represents the electrochemical impedance parameters established from the electrical equivalent circuits. The reducing trend of CPE_{dl} values implies to increased electrical double layer thickness and/or reduced local dielectric constant as a result of the formation of corrosion inhibitive film [47-48].

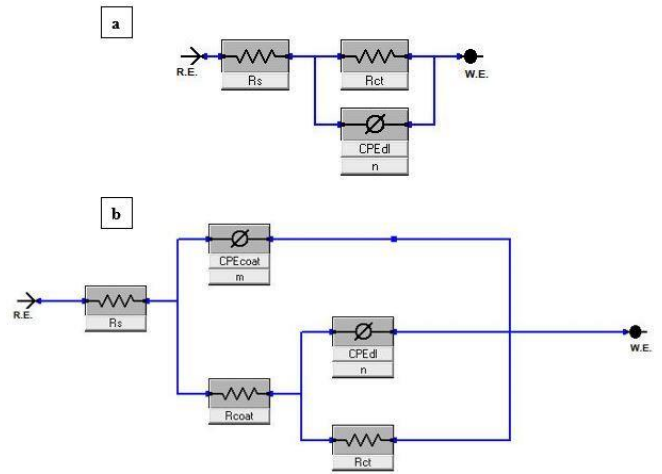


Figure 3. Electrical equivalent circuit for a) blank mild steel and b) coated mild steel in 0.5 M HCl in the absence and presence of different concentrations of eumelanin

Based on Fig. 1, bare mild steel substrate exposed to the electrolyte demonstrated the highest i_{corr} , which means the most susceptible to undergo corrosion. On the other hand, the application of neat APTES-TEOS hybrid sol-gel coating was able to retard the metal dissolution with 43.98% inhibition efficiency, acting as a physical hindrance between the metal and the corrosive electrolyte. Table 1 outlines the polarization parameters of the bare mild steel and coated metal substrates. In the presence of increasing eumelanin concentration, corrosion current density reduced significantly and the corrosion potential became nobler. Thus, the barrier effect improved in the presence of eumelanin to mitigate the contact of corrosive ions and the metal. Even with the application of coatings, corrosion phenomenon is inevitable due to the structural imperfections of the coatings, including cracks and pores [49, 50].

Electrochemical impedance spectroscopy analysis was performed to evaluate the corrosion behavior of hybrid sol-gel coated mild steel in 0.5 M HCl solution of eumelanin polymer. As can be visualized from Fig. 2, Nyquist plots were attained in the form of depressed semicircles (at high frequency region), attributing to inhomogeneity (interfacial irregularities) and surface roughness of the metal substrate [47, 48, 51]. With the incorporation of eumelanin into the synthesized APTES-TEOS hybrid sol-gel matrix, the diameter of the semicircle increased with increased doping concentration, signifying the corrosion protective ability of eumelanin.

Table 1. Potentiodynamic polarization parameters of coated mild steel specimens in 0.5 M HCl without and with the incorporation of different concentrations of eumelanin.

Sample	E_{corr} (V vs SCE)	j_{corr} ($\mu\text{A cm}^{-2}$)	β_a (V dec ⁻¹)	$-\beta_c$ (V dec ⁻¹)	Corrosion rate (mpy)	IE%
0.5 M HCl (Blank)	-0.479	364.2	0.1022	0.1442	166.0	-
Neat sol-gel coating	-0.484	204.0	0.0717	0.1237	92.8	43.98
Sol-gel coating + 100 ppm eumelanin	-0.442	127.5	0.0794	0.1722	58.4	64.99
Sol-gel coating + 1000 ppm eumelanin	-0.446	70.6	0.0890	0.1233	32.3	80.62

Table 2. Electrochemical impedance parameters of coated mild steel specimens in 0.5 M HCl without and with the incorporation of different concentrations of eumelanin.

Sample	R_s ($\Omega \text{ cm}^2$)	R_{coat} ($\Omega \text{ cm}^2$)	CPE_{coat} ($\mu\text{F cm}^2$)	m	R_{ct} ($\Omega \text{ cm}^2$)	CPE_{dl} ($\mu\text{F cm}^2$)	n	IE%
0.5 M HCl (Blank)	7.66	-	-	-	63.53	850.8	0.7949	-
Neat sol-gel coating	6.02	24.78	179.6	0.8745	115.49	631.6	0.9034	44.99
Sol-gel coating + 100 ppm eumelanin	7.92	40.94	460.3	0.8625	194.48	455.2	0.7944	67.33
Sol-gel coating + 1000 ppm eumelanin	7.51	59.85	342.2	0.8759	336.22	251.2	0.8149	81.10

EIS study revealed that in the presence of 1000 ppm eumelanin, which rendered the largest semi-circular plot (i.e., highest coating resistance), the inhibition efficiency of the coating reached up to 81.10%, affirming the potential of eumelanin as an effective corrosion inhibitor. Thus, Nyquist diagrams exemplify that the fabricated coatings with the doping of eumelanin have effective adhesion and inhibitive properties with mild steel surface, implying reduced infiltration of the corrosive ions to mild steel surface. The diameter of Nyquist plots achieved for the bare mild steel and hybrid sol-gel coated mild steel in 0.5 M HCl embodies the capacitive loops associated with the charge transfer that is linked with the corrosion process [48, 52]. Coated samples in the presence of eumelanin obtained the least χ^2 (chi-squared) values in the range of 10^{-3} .

4. Conclusions and Recommendations

In conclusion, this work explored the prospect of using eumelanin pigment as an effective corrosion inhibitor over conventional organic materials. In this study, eumelanin polymer was produced using *Streptomyces parvus* BSB49 strain and the anticorrosive performance of this polymer was investigated. For this purpose, hybrid sol-gel matrix was synthesized using tetraethyl orthosilicate and 3-aminopropyltriethoxysilane. Based on the obtained results, it was affirmed that the eumelanin polymeric pigment has anticorrosion properties, where the incorporation into fabricated silanol coatings offered significant corrosion mitigation over the neat coating.

5. Acknowledge

Ethics Committee Approval:

This study does not contain any studies with human participants or animals.

Author Contributions:

This study was designed by SB and MHH. MÖ were responsible for eumelanin biopolymer production and purification with SB. MHH and TSH were responsible for synthesis of hybrid sol-gel matrix, application of coatings, analysis of corrosion performance, potentiodynamic polarization analysis and electrochemical impedance spectroscopy analysis. SB and MHH were responsible for the writing of the manuscript, final editing and submission.

Conflict of Interest:

The authors declare that they have no conflict interests.

References

[1] Banerjee A., Supakar S., Banerjee R. Melanin from the nitrogen fixing bacterium *Azotobacter chroococcum*: a spectroscopic characterization. PLoS One. 2014;9(1):e84574. <https://doi.org/10.1371/journal.pone.0084574>

[2] Ryu I.Y., Choi I., Ullah S., Choi H., Chun P., Moon H.R. Tyrosinase Inhibitory Effects of Derivatives of (E)-2-(Substituted Benzylidene)-3, 4-Dihydronaphthalen-1 (2 H)-One. Bulletin of the Korean Chemical Society. 2020;41(12):1134-1139. <https://doi.org/10.1002/bkcs.12122>

[3] Dwyer T., Muller H.K., Blizzard L., Ashbolt R., Phillips G. The use of spectrophotometry to estimate melanin density in Caucasians. Cancer Epidemiology and Prevention Biomarkers. 1998;7(3):203-206.

[4] d'Ischia M., Wakamatsu K., Cicoira F., Di Mauro E., Garcia-Borron J.C., Commo S., Ito, S. Melanins and melanogenesis: from pigment cells to human health and technological applications. Pigment cell & melanoma research. 2015;28(5):520-544.

[5] Riley P.A. Melanin. International Journal of Biochemistry & Cell Biology. 1997;29:1235-1239

[6] Land E.J., Ramsden C.A., Riley P.A. Quinone chemistry and melanogenesis. Methods in Enzymology. 2004;378:88-109

[7] Eisenman H.C., Casadevall A. Synthesis and assembly of fungal melanin. Applied Microbiology and Biotechnology. 2012;93(3):931-940.

[8] Langfelder K., Streibel M., Jahn B., Haase G., Brakhage A.A. Biosynthesis of fungal melanins and their importance for human pathogenic fungi. Fungal Genetics and Biology. 2003;38:143-158

[9] Prota G. Recent advances in the chemistry of melanogenesis in mammals. Journal of Investigative Dermatology. 1980;75(1):122-127.

[10] Ito S. A chemist's view of melanogenesis. Pigment cell research. 2003;16(3):230-236.

[11] Fedorow H., Tribl F., Halliday G., Gerlach M., Riederer P., Double K.L. Neuromelanin in human dopamine neurons: comparison with peripheral melanins and relevance to Parkinson's disease. Progress in Neurobiology. 2005;75(2):109-124. <https://doi.org/10.1016/j.pneurobio.2005.02.001>

[12] Zucca F.A., Segura-Aguilar J., Ferrari E., Muñoz P., Paris I., Sulzer D., Zecca L. Interactions of iron, dopamine and neuromelanin pathways in brain aging and Parkinson's disease. Progress in Neurobiology. 2017;155:96-119. <https://doi.org/10.1016/j.pneurobio.2015.09.012>

[13] Płonka P., Grabacka M. Melanin synthesis in microorganisms: biotechnological and medical aspects. Acta Biochimica Polonica. 2006;53(3):429-443

[14] McCallum N.C., Son F.A., Clemons T.D., Weigand S.J., Gnanasekaran K., Battistella C., Gianneschi N.C. Allomelanin: A biopolymer of intrinsic microporosity. Journal of the American Chemical Society. 2021;143(10):4005-4016. <https://doi.org/10.1021/jacs.1c00748>

[15] Ruzafa C., Solano F., Sanchez-Amat A. The protein encoded by the *Shewanella colwelliana* melaA gene is a p-hydroxyphenylpyruvate dioxygenase. FEMS microbiology letters. 1994;124(2):179-184.

[16] Turick C.E., Knox A.S., Becnel J.M., Ekechukwu A.A., Milliken C.E. Properties and function of pyomelanin. Biopolymers. 2010;449:72

[17] Bayram S. Production, purification, and characterization of *Streptomyces* sp. Strain MPPS2 extracellular pyomelanin pigment. Archives of Microbiology. 2021;203:4419-4426 <https://doi.org/10.1007/s00203-021-02437-w>

[18] Solano F. Melanin and melanin-related polymers as materials with biomedical and biotechnological applications—cuttlefish ink and mussel foot proteins as inspired biomolecules. International journal of molecular sciences. 2017;18(7):1561.

[19] Venil C.K., Zakaria Z.A., Ahmad W.A. Bacterial pigments and their applications. Process Biochemistry.

- 2013;48(7):1065–1079. <https://doi.org/10.1016/j.procbio.2013.06.006>
- [20] Zerrad A., Anissi J., Ghanam J., Sendide K., El Hassouni M. Antioxidant and antimicrobial activities of melanin produced by a *Pseudomonas balearica* strain. *Journal of Biotechnology Letters*. 2014;5(1):87-94.
- [21] Łopusiewicz L. The isolation, purification and analysis of the melanin pigment extracted from *Armillaria mellea* rhizomorphs. *World Scientific News*. 2018;100:135-153.
- [22] Teplyakova T., Kosogova T. Fungal bioactive compounds with antiviral effect. *Journal of Pharmacy and Pharmacology*. 2015;3(8):357.
- [23] Hung Y.C., Sava V., Hong M.Y., Huang G. S. Inhibitory effects on phospholipase A2 and antivenin activity of melanin extracted from *Thea sinensis* Linn. *Life sciences*. 2004;74(16):2037-2047.
- [24] Page S., Chandhoke V., Baranova A. Melanin and melanogenesis in adipose tissue: possible mechanisms for abating oxidative stress and inflammation?. *Obesity reviews*. 2011;12(5):e21-e31.
- [25] Sava V., Hung Y., Blagodarsky V., Hong M.Y., Huang, G. The liverprotecting activity of melanin-like pigment derived from black tea. *Food Research International*. 2003;36(5):505–511. [https://doi.org/10.1016/S0963-9969\(02\)00199-0](https://doi.org/10.1016/S0963-9969(02)00199-0)
- [26] Sava V.M., Galkin B.N., Hong M.Y., Yang P.C., Huang G.S. A novel melanin-like pigment derived from black tea leaves with immunestimulating activity. *Food Research International*. 2001;34:337–43.
- [27] Bayram S., Dengiz C., Gerçek Y.C., Cetin I., Topcul M.R. Bioproduction, structure elucidation and in vitro antiproliferative effect of eumelanin pigment from *Streptomyces parvus* BSB49. *Archives of Microbiology*. 2020;202(9):2401-2409. <https://doi.org/10.1007/s00203-020-01956-2>
- [28] Margalida A., Negro J.J., Galván I. Melanin-based color variation in the bearded vulture suggests a thermoregulatory function. *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology*. 2008;149(1):87-91.
- [29] Tarangini K., Mishra S. Production of melanin by soil microbial isolate on fruit waste extract: two step optimization of key parameters. *Biotechnology Reports*, 2014;4:39-146.
- [30] Fan Q., Cheng K., Hu X., Ma X., Zhang R., Yang M., Cheng Z. Transferring biomarker into molecular probe: melanin nanoparticle as a naturally active platform for multimodality imaging. *Journal of the American Chemical Society*. 2014;136(43):15185-15194.
- [31] Gogurla N., Roy B., Min K., Park J.Y., Kim S. A Skin-Inspired, Interactive, and Flexible Optoelectronic Device with Hydrated Melanin Nanoparticles in a Protein Hydrogel–Elastomer Hybrid. *Advanced Materials Technologies*, 2020;5(4):1900936.
- [32] Schweitzer A.D., Revskaya E., Chu P., Pazo V., Friedman M., Nosanchuk J.D., Dadachova E. Melanin-covered nanoparticles for protection of bone marrow during radiation therapy of cancer. *International Journal of Radiation Oncology* Biology* Physics*. 2010;78(5):1494-1502.
- [33] Araújo M., Viveiros R., Correia T.R., Correia I.J., Bonifácio V.D., Casimiro T., Aguiar-Ricardo A. Natural melanin: A potential pH-responsive drug release device. *International journal of pharmaceutics*. 2014;469(1):140-145.
- [34] Di Mauro E., Rho D., Santato C. Biodegradation of bio-sourced and synthetic organic electronic materials towards green organic electronics. *Nature communications*. 2021;12:1-10. <https://doi.org/10.1038/s41467-021-23227-4>
- [35] Şahin N., Uğur A. Investigation of the antimicrobial activity of some *Streptomyces* isolates. *Turkish Journal of Biology*. 2003;27(2):79-84.
- [36] El-Naggar N.A., El-Ewasy S.M. Bioproduction, characterization, anticancer and antioxidant activities of extracellular melanin pigment produced by newly isolated microbial cell factories *Streptomyces glaucescens* NEAE-H. *Scientific Reports* 2017;7:42129. <https://doi.org/10.1038/srep42129>
- [37] Wu Y., Du Z., Wang H., Cheng X. Synthesis of aqueous highly branched silica sol as underlying crosslinker for corrosion protection. *Progress in Organic Coatings*. 2017;111:381-388.
- [38] Hamidon T.S., Ishak, N.A., Hussin, M.H. Enhanced corrosion inhibition of low carbon steel in aqueous sodium chloride employing sol–gel-based hybrid silanol coatings. *Journal of Sol-Gel Science and Technology*. 2021;97(3):556-571.
- [39] Fayomi O.S.I., Akande I.G., Popoola A.P.I., Molifi H. Potentiodynamic polarization studies of Cefadroxil and Dicloxacillin drugs on the corrosion susceptibility of aluminium AA6063 in 0.5 M nitric acid. *Journal of Materials Research and Technology*, 2019;8(3):3088-3096.
- [40] Tasić Ž.Z., Mihajlović M.B.P., Radovanović M.B., Antonijević M.M. Electrochemical investigations of copper corrosion inhibition by azithromycin in 0.9% NaCl. *Journal of Molecular Liquids*. 2018;265:687-692.
- [41] Kirtay S. Preparation of hybrid silica sol–gel coatings on mild steel surfaces and evaluation of their corrosion resistance. *Progress in Organic Coatings*. 2014;77(11):1861-1866.
- [42] Sharma P., Pandey P M. Corrosion rate modelling of biodegradable porous iron scaffold considering the effect of porosity and pore morphology. *Materials Science and Engineering: C*. 2019;103:109776.
- [43] Ishak N.A., Hamidon T.S., Zi-Hui T., Hussin M.H. Extracts of curcumin-incorporated hybrid sol–gel coatings for the corrosion mitigation of mild steel in 0.5 M HCl. *Journal of Coatings Technology and Research*, 2020;17(6):1515-1535.
- [44] Ali S.M., Emran K.M., Messali M. Improved protection performance of modified sol-gel coatings with pyridinium-based ionic liquid for cast iron corrosion in 0.5 M HCl solution. *Progress in Organic Coatings*. 2019;130:226-234.
- [45] Ćurković L., Ćurković H.O., Salopek S., Renjo M.M., Šegota S. Enhancement of corrosion protection of AISI 304 stainless steel by nanostructured sol–gel TiO₂ films. *Corrosion Science*. 2013;77:176-184.
- [46] Ateş S., Baran-Aydın E., Yazıcı B. The corrosion behavior of the SnO₂-coated mild steel in HCl solution at different temperature. *Journal of Adhesion Science and Technology*. 2021;35(4):419-435.
- [47] Hamidon T.S., Hussin M.H. Susceptibility of hybrid sol-gel (TEOS-APTES) doped with caffeine as potent corrosion protective coatings for mild steel in 3.5 wt.% NaCl. *Progress in Organic Coatings*. 2020;140:105478.
- [48] Prasad A.R., Shamsheera K.O., Joseph A. Electrochemical and surface characterization of mild steel with corrosion resistant zirconia network fabricated by aqueous sol-gel technique. *Journal of the Indian Chemical Society*. 2021;98(4):100052.

- [49] Pourhashem S., Afshar A. Double layer bioglass-silica coatings on 316L stainless steel by sol-gel method. *Ceramics International*. 2014;40(1):993-1000.
- [50] John B., Paulraj S., Mathew J. The role of shielding gas on mechanical, metallurgical and corrosion properties of corten steel welded joints of Railway Coaches using GMAW. *Advances in Science and Technology. Research Journal*. 2016;10(32):156-168
- [51] Shaw P., Obot I.B., Yadav M. Functionalized 2-hydrazinobenzothiazole with carbohydrates as a corrosion inhibitor: electrochemical, XPS, DFT and Monte Carlo simulation studies. *Materials Chemistry Frontiers*. 2019;3(5):931-940.
- [52] Abd El-Lateef H.M., Khalaf M.M. Corrosion resistance of ZrO₂-TiO₂ nanocomposite multilayer thin films coated on carbon steel in hydrochloric acid solution. *Materials Characterization*. 2015;108:29-41.