ACADEMIA ROMÂNĂ
Revue Roumaine de Chimie
http://web.icf.ro/rrch/

Rev. Roum. Chim.,
2020, *65*(3), 247-254
DOI: 10.33224/rrch.2020.65.3.04

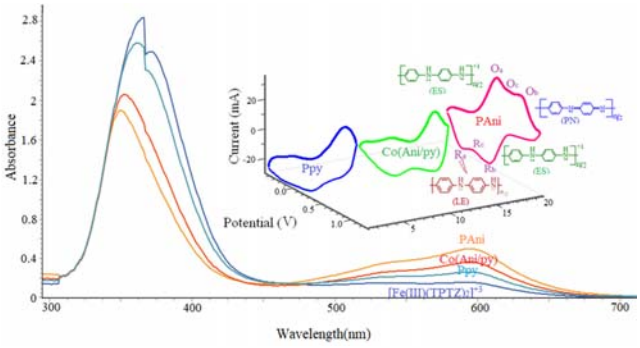
IRON (III) REDUCTION AS A MEASURE OF "ANTIOXIDANT POWER" USING HOMO AND COPOLYMER OF PYRROLE AND ANILINE ELECTROSYNTHESIZED

Nafiseh HAJIABDOLAH,[a] Ali PARSA,[a,b,*] Hossein ANARAKI-ARDAKANI[a,*] and Hossein JALALI- JAHROMI[a]

[a] Department of Chemistry, Mahshahr Branch, Islamic Azad University, Mahshahr, Iran
[b] Department of Chemistry, College of Science, Yadegar -e- Imam Khomeini (RAH) Shahre Rey Branch, Islamic Azad University, Tehran, Iran

Received November 10, 2019

Ferric Reducing Antioxidant Power (FRAP) assay is a measure of assessment of the total antioxidant power of compounds. In this study, the ability of homo and copolymer of Aniline (Ani) and Pyrrole (Py) to act as an antioxidant on ferric ion reduction is investigated. The oxidative homo and copolymerization on composite 2B pencil graphite were accomplished in citric acid medium containing calcium chloride ($CaCl_2$) as the supporting electrolyte by cyclic voltammetry method. The morphology and structural analysis of homo and copolymer was confirmed by SEM and FTIR. The FTIR and UV-visible spectra indicated that the increase of aniline monomer in the copolymer chain enhances the ferric ion reduction ability. The electrochemical impedance spectroscopy (EIS) is used for further investigation. It was revealed that the increase of aniline ring in polymeric chain decreased the charge transfer resistance (*R*ct) and thus increases the reduction effect and improved the free radical scavenging activity of copolymers.



INTRODUCTION

Nowadays, conductive polymers are widely used in the manufacture of sensors, catalysts, electrolyte fluids, as well as in biomedicine for drug delivery, protein purification, tissue engineering, neural interactions, and activators.[1-3] Various types of conductive polymers with antioxidant properties can be synthesized, such as; polyaniline and polypyrrole and the other polyphenols.[4-6] The role and beneficial effects of antioxidants against many human diseases due to oxidative corruption have attracted much attention in recent years as antioxidants prevent the attack of reactive oxygen species and end radical reactions.[7] Antioxidants reduce the risk of cardiovascular disease and stroke by neutralizing free radicals on the one hand and preventing the progression of cancers.[8] The antioxidant capacity can be determined by Ferric Reducing Antioxidant Power (FRAP) method.[9, 10] In this method, the ability of antioxidants to reduce ferric ions in the Fe^{3+}-tripyridyltriazine [(Fe (III) –TPTZ)] complex is measured.[11] By reducing ferric ions and converting them to ferrous ions in acidic medium and in the presence of the triperidyltriazine (TPTZ) ligand, the Fe^{2+}-tripyridyltriazine [(Fe (II) –TPTZ)] complex is formed, which is blue in color. Then, optical sensing is measured at 593 nm.[12-14]

* Corresponding author: Hosseinanaraki@yahoo.com; Tel.: 09125190488 or iausr.parsa@yahoo.com; Tel.: 09122581176

In this study, electrosynthesized homo and copolymer aniline/pyrrole in 1 M citric acid containing calcium chloride ($CaCl_2$) has been studied. Then, using the UV-Vis apparatus, the antioxidant effect of homo and copolymer on ferric ion reduction at 593 nm was investigated. Also, electrochemical impedance spectroscopy (EIS) was used to quantitatively and qualitatively study electrochemical processes in conductive polymer films and to justify their antioxidant properties.

EXPERIMENTAL

Materials

Anilline (sigma chemicals, USA) was purified by distillation under a nitrogen atmosphere at reduced pressure. The resulting colorless liquid was kept in the dark at 5 C°. Pyrrole (py) (Merk, Germany) was vacuum distilled before use calcium chloride (CaCl2), citric acid (C6H8O7) and 2, 4, 6- tripyridyl – s– triazine) (TPTZ) were of analytical grade and used as received. Potassium ferricyanide ($K_3[Fe(CN)_6]$) and ferrocyanide ($K_4[Fe(CN)_6]$) (Shanghani No.1 Reagent Factory , China) were used as received. All aqueous soloution were freshly prepared using ultra – pure water from Milli – Q plus (Millipore Corp., USA).

Equipment

The Potentiostat and Galvanostat (Ivium Technologies, Netherlands), made by the Netherlands, has been used to electrosynthesis and characterization electrochemical impedance spectroscopy (EIS) measurements. Electrochemical studies were performed on a conventional three-electrode custom-made cell. The three-electrode cell consists of working electrode (2B pencil composite graphite, 1.8 mm diameter) (Staedtler Lumograph, Germany) and Pt electrode as counter electrode against Ag /AgCl electrode as reference electrode have been used in electrochemical synthesis. The structural analysis and characterization purposes were determined by fourier transform infrared (FTIR) spectroscopy system 2000 (TENSOR 27, Bruker) and UV–visible spectrophotometer V-500 (JASCO, Japan). China's SEM (Scanning Electron Microscopy) model KYKY-EM3200 was used to elucidate the morphology of the homo and copolymer of Ani and Py.

Procedure

The oxidative homo and copolymerization of Ani and Py on the surface of composite 2B pencil graphite was performed using 10 ml solution containing 50 mM monomers, 1 M citric acid and 0.5 M calcium chloride by sequentially sweeping the potential between − 0.4 and +1.2 V vs., scan rate 100 mV/s^{-1}, under OFN atmosphere and at 25 ± 2 °C.

The FRAP fresh working solution was prepared by mixing 25 mL acetate buffer (300 mM acetate buffer at pH 3.6), 2.5 mL TPTZ solution (10 mM TPTZ solution in 40 mM HCl) and 2.5 mL $FeCl_3.6H_2O$ solution (20 mM) and then warmed at 37 °C before using. 10 mg of homo and copolymer were allowed to react with 5 mL of the FRAP solution for 30 min in the dark condition. Reading the colored solution (ferrous tripyridyltriazine complex) was then taken at 593 nm.

The EIS of homo and copolymer films in 1 M citric acid containing 0.001 M $K_4[Fe(CN)_6]$ and $K_3[Fe(CN)_6]$ was performed using the following parameters: amplitude of 0.01 V; frequency range of 100 Hz to100000 Hz, potential range: 0.5 V.

RESULTS AND DISCUSSION

Electrochemical synthesis of homo and Copolymer of Aniline and Pyrrole

Fig. 1a shows cyclic voltammogram of 50 mM aniline in 1 molar citric acid containing 0.5 M calcium chloride on the working electrode (composite 2B pencil graphite) at the potential range of -0.4 to 1.2 V and scan rate of 100 mV/s. Three pairs of redox peaks were observed at a potential range of -0.2 to 1 V. The first pair of redox peak in the range of -0.2 to 0.3 V (O_a/R_a) is related to conversion of leucoemeraldine (LE) to emeraldine salt (ES). The second pair above of the potential range of 0.6 V (O_b/R_b) is due to ES oxidation to PN (pernigraniline). Pair of redox peaks in the range of 0.3 to 0.6 V (O_c/R_c) is attribute to the degradation of hydroquinone products to quinones.[15-19]

Fig. 1b shows an electrosynthesis cyclic voltammogram of 50 mM Py in 1 M citric acid containing 0.5 M calcium chloride on the working electrode at the potential range of -0.4 to 1.2 V and a scan rate of 100 mV/s. Due to the easier oxidation of the pyrrole monomer and the easier formation of its cation radical, the PPy synthesis peak shifts to more negative potentials.[19,20] In the PPy synthesis voltammogram, in the forward scan, a configuration state was observed which disappeared on the reverse scan; it was reported that the peak is related to the impurities of the composite bed.[21, 22]

Fig. 1c shows electro-copolymerization cyclic voltammogram of 50 mM Py and Ani in 1 M citric acid containing 0.5 M calcium chloride on the working electrode at the potential range of -0.4 to 1.2 V and scan rate of 100 mV/s. In the first step, the relative rate of copolymer formation depends on the type and potential of oxidation of each monomer, material, and appearance of the electrode. In the next step, the reaction of the radical cations binding to each other occurs in order to form a copolymer.[23-25]

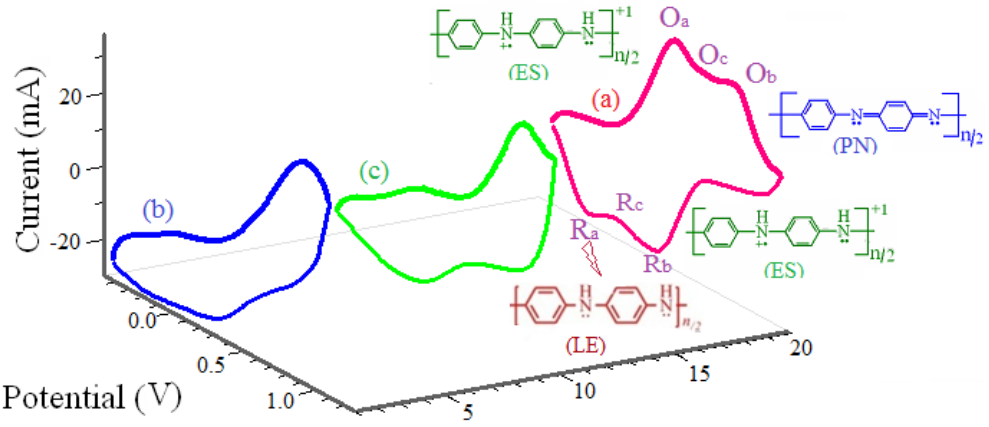


Fig. 1 – The schematic of cyclic voltammograms obtained during electrosynthesis of a) PAni, b) PPy, c) Poly(Ani-*co*-Py) in 1 M citric acid containing 0.5 M $CaCl_2$. The potential is cycled between -0.4 to 1.2 V at a scan rate of 100 mV/s.

Structural analysis

Fig. 2a shows baseline corrected FTIR spectra of PAni electrosynthesized. The presence of two bands in the range of 1654 and 1567 cm^{-1} are related to C-C stretching vibration of quinoid (Q) and benzenoid (B) rings, respectively, while the absorption band at 1496 cm^{-1} attribute to C-N stretching vibration of ES configurations state of PAni. The in-of-plane and out-of-plane CH bending vibration band of the aromatic rings appear in the absorption band of 1154 to 1009 cm^{-1}, respectively.[26]

Fig. 2b shows the FT-IR spectrum of PPy electrosynthesized. The stretching vibration band C-C and C-N in the PPy ring structure appear at the range of 1532 cm^{-1} and 1407 cm^{-1}, respectively. The emergence of absorption band in the range of 1203 cm^{-1} is attributed to the vibration deformation band of in-plane C-H and N-H. The absorption bands at around 1100 to 1254 cm^{-1} is related to the high vibration of the PPy ring. The absorption bands in the range of 1038 cm^{-1}, 820 cm^{-1}, and 601 cm^{-1} are related to out-of-plane of, C–H bending vibrations of the aromatic rings, C-H ring deformation and C-H vibrations of PPy ring, respectively. In areas of 678 cm^{-1}, it was observed that absorption band is related to flexural C-C deformation of PPy ring.[20]

Fig. 2c shows the FT-IR spectrum of Poly(Ani-*co*-Py) electrosynthesized. The spectrum of copolymer indicates that presence of monomers of Ani and Py in its formation. The absorption bands at 1206 and 1299 cm^{-1} indicate that on the presence of PAni, and the absorption bands bands in the range of 1544 cm^{-1} and 1034 cm^{-1} are related to stretching vibrational bands of C = C and C-H that demonstrate the presence of PPy. The band at 912 cm^{-1} can be attributed to out-of plane C–H of the PPy aromatic ring. The C-N stretching band is observed at 1250 cm^{-1} wavelength, although in this research it's observed in a lower wavelength of 1206 cm^{-1}.[24]

Morphology analysis

The SEM images of homo and copolymer Ani and Py electrosynthesized in the presence of calcium chloride as supporting electrolyte are different to each other.[27] Fig. 3 shows the SEM image of the PAni electrosynthesized from 50 mM Ani in 1 M citric acid containing 0.5 M calcium chloride. The SEM image obtained from PAni electrosynthesized on the working electrode shows a dense mass-shaped morphology.

Fig. 4 shows the SEM image of the PPy electrosynthesized from 50 mM Py in 1 M citric acid containing 0.5 M calcium chloride. The SEM image of the PPy shows cluster morphology. The EDX analysis results indicate that amount of carbon (C) and nitrogen (N) are 55.08 and 13.17%, respectively. These results are in good agreement with the calculated value in polymer, which is (C) 61.35% and (N) 15.90%.[28]

Fig. 5 shows the SEM image of Poly(Ani-*co*-Py) electrosynthesized from 50 mM Ani and 50 mM Py in 1 M citric acid containing 0.5 M calcium chloride. The SEM image of Poly(Ani-*co*-Py) electrosynthesized on the working electrode shows a granular and rough morphology. The results of EDX analysis from Poly(Ani-*co*-Py) electrosynthesized in the presence of $CaCl_2$ for carbon (C) and nitrogen (N) are 90.16 and 1.33%, respectively. These results are in good agreement with the calculated value in polymer, which are 93.66% (C) and 1.76% (N), respectively.[28]

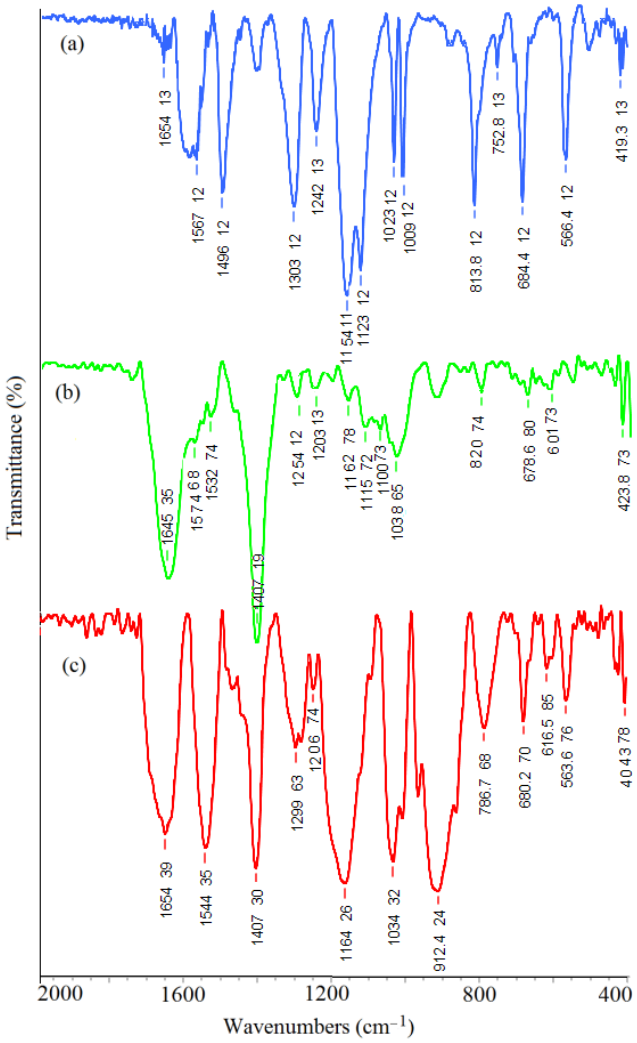


Fig. 2 – Baseline corrected FTIR spectra (2000-400 cm^{-1}) of a) PAni, b) PPy, c) Poly(Ani-*co*-Py) electropolymerized in 1 M citric acid containing 0.5 M $CaCl_2$.

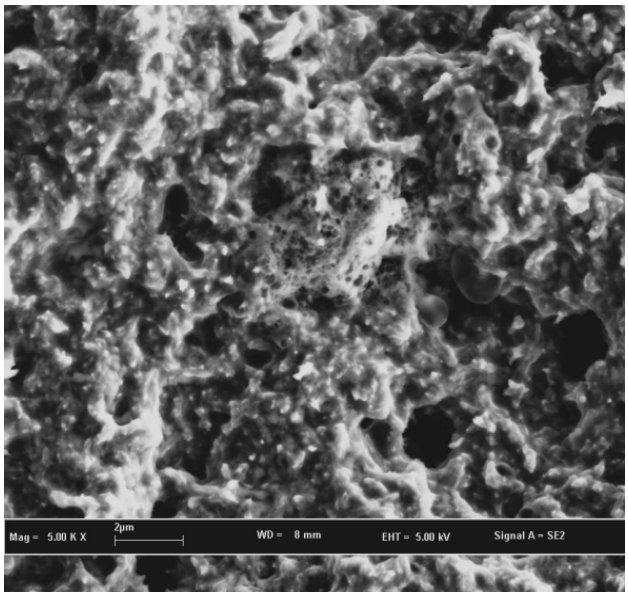


Fig. 3 – The SEM image of electropolymerization of 50 mM Ani solution in 1 M citric acid containing 0.5 M calcium chloride on composite graphite.

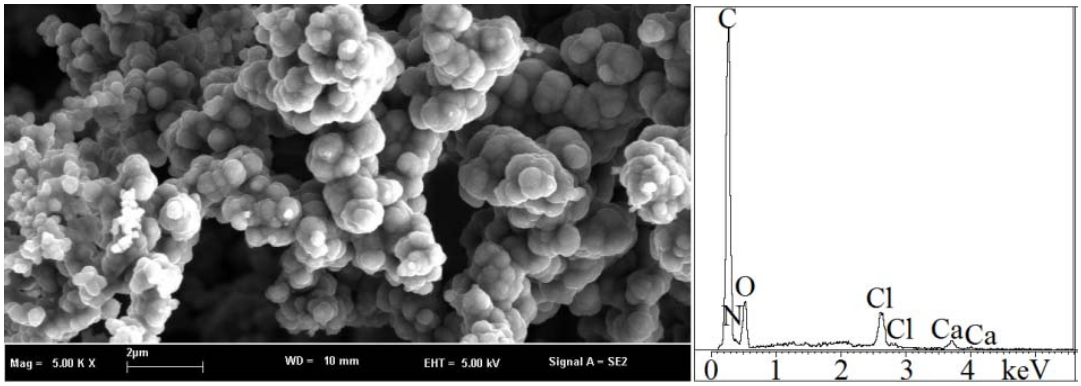


Fig. 4 – The SEM image of electropolymerization of 50 mM PPy solution in 1 citric acid containing 0.5 M calcium chloride on composite graphite.

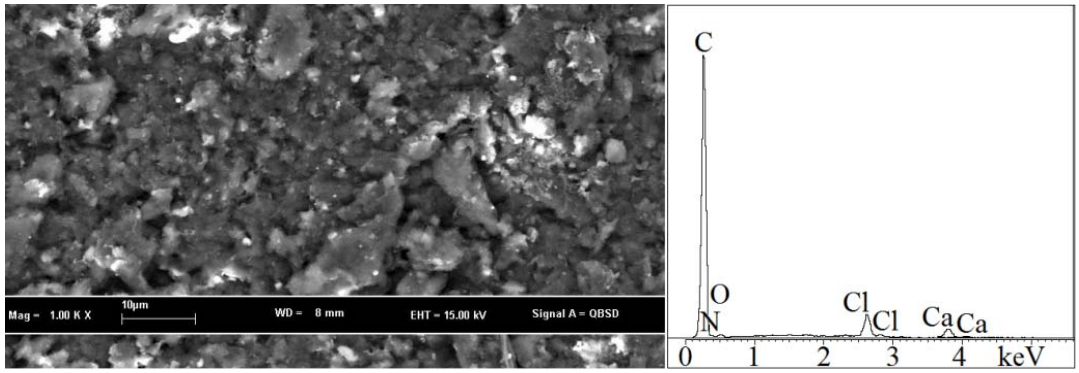


Fig. 5 – The SEM images of copolymerization of 50 mM Ani solution and 50 mM Py solution in 1 M citric acid containing 0.5 M calcium chloride on composite graphite.

Ferric ion reduction

In this research, spectrophotometry has been used to measure fast iron ions. Since Fe ion absorption is very small in spectroscopy, a colored complex is made of it that has a significant absorption in the UV-visible range compared to Fe ion and has high sensitivity in measuring iron ions.[29]

Fig. 6 shows the UV-visible spectrum of 10 ml of the 0.001 molar Fe (III)-TPTZ, exposed to 0.01 g of (a) PAni, (b) PPy and (c) Poly(Ani-*co*-Py) for 30 min. The difference in absorption intensity indicates the ability of the polymer to transfer electrons to the Fe^{+3}-TPTZ complex, which indicates the more ability of the polymer to transfer electrons and consequently the formation of a Fe^{+2}-TPTZ blue complex with higher absorption intensity. In other words, the concentration of the Fe^{+2}-TPTZ complex increases and as a result, the absorption intensity increases, and thus the concentration of the Fe^{+3}-TPTZ complex decreases.[7,30] Fig. 6d shows the UV-visible spectrum of the Fe^{+3}-TPTZ complex without being exposed to homo and copolymer. The absorption intensity, due to the transferring power of electron homo and copolymer Ani and Py electrosensitized increases by converting Fe (III) to Fe (II). Spectra: (a) The greater effect of PAni on electron release with the highest absorbance (0.54); (b) the less effective impact of Poly(Ani-*co*-Py) on release of electron as compared to PAni and the greater effect of copolymer on release of electron in comparison with PPy; and c) the lesser effect of a PPy on release of electrons with a minimum absorption intensity (0.33) in a 593 nm wavelength. These results indicate the ability of the polymer to reduce Fe (III) and the formation of a complex of Fe (II) with the TPTZ ligand.

Electrochemical impedance spectroscopy

The Nyquist spectroscopic impedance curve of modified electrode with: (a) PAni, (b) PPy; and c) Poly(Ani-*co*-Py) in 0.001 molar ($Fe(CN)_6]^{-3/-4}$) solution at E_{app} 0.5 V and frequency range used was from 100 to 100,000 Hz are shown in Fig. 7.

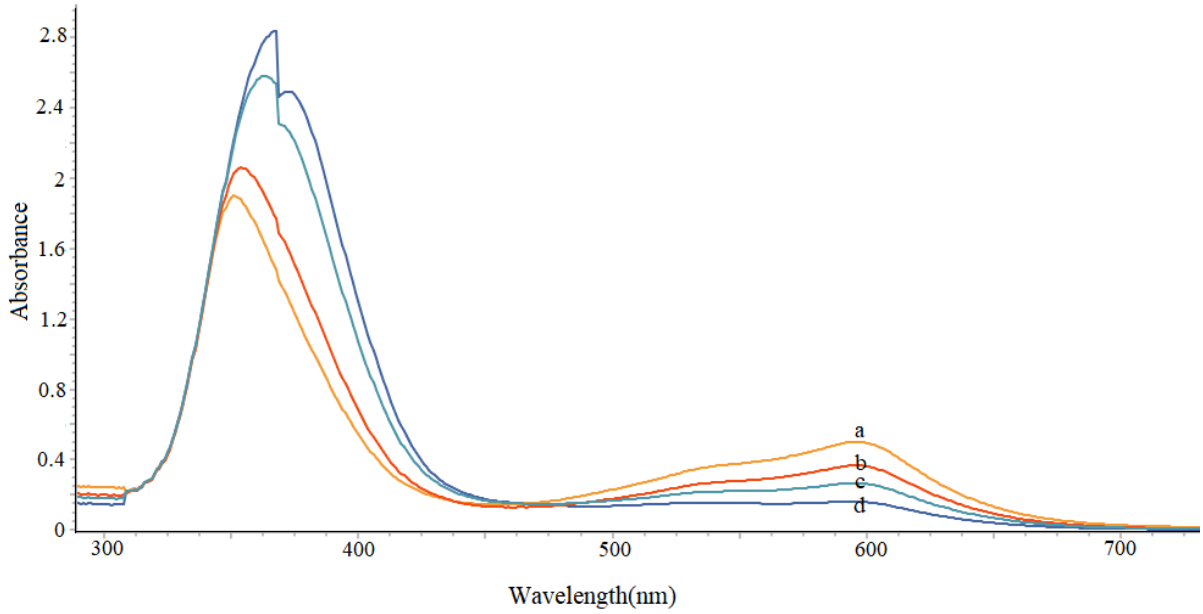


Fig. 6 – UV-vis spectra of 0.001 M complex Fe^{+3} in 1 M citric acid after 30 min exposure to 0.01 g from: a) PAni, b) Poly(Ani-*co*-Py), c) PPy, electropolymerized. d) Absorbance of Fe^{+3} Ion in the absence of homo and copolymer is 0.16.

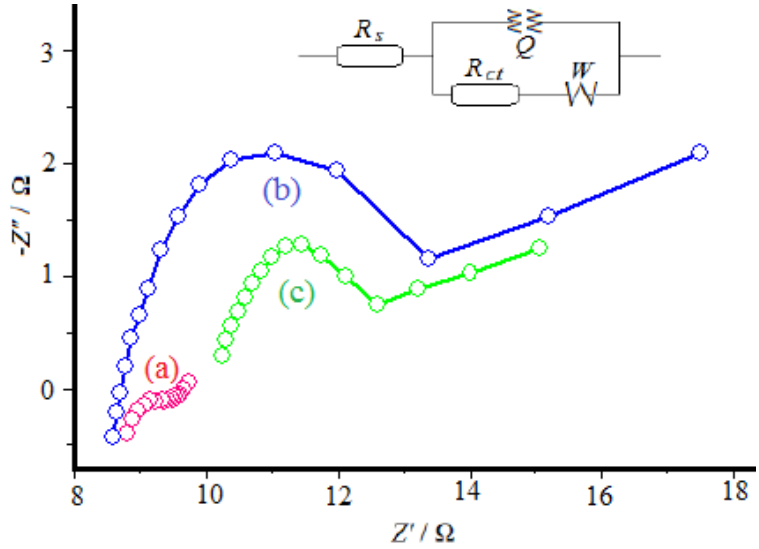


Fig. 7 – Nyquist plots of modified electrode with: a) PAni, b) PPy, c) Poly(Ani-*co*-Py) in 0.001 M potassium ferric and Ferro cyanide ($[Fe(CN)_6]^{-3/-4}$) solution at E_{app} 0.5 V. The ac potential amplitude was 10 mV and frequency range used was from 100 KHZ to 100Hz.

As shown in Fig. 7a and b, the further increase of the observed current in the graphite electrode modified with PAni reduces the charge transfer resistance (11 ohms), and the decrease in the observed current in the graphite electrode modified with PPy increases the charge transfer resistance (15.6 ohms). This is due to the fact that the lesser the charge transfer resistance for the polymer, the higher the charge transfer, resulting in easier redox transfers for the conductive polymer. As the Warburg impedance (Zw) is low at the surface of the double layer, the penetration phenomenon is better at the electrode surface and there could be a better charge transfer.[31, 32] Thus, the Warburg impedance obtained in the test for a graphite electrode modified with PAni 56 [1/Ω sqrt (Hz)], PPy 99 [1/Ω sqrt (Hz)] and Poly(Ani-*co*-Py) 75 [1/Ω sqrt (Hz)], confirms the better penetration of Fe ions into the surface of the electrode modified with PAni, as compared to that modified with PPy. N represents the degree of surface roughness of the modified electrode, where the more porous and thicker it is, the lesser the electron transfer from the polymer surface. The numbers obtained in the table refer to the smoother surface of the PAni and hence the better electron transfer.[33, 34]

Table 1

The best fitted values of Randles' equivalent circulate elements simulated from impedance data for homo and copolymer of Ani and py films

Element	$R_{(s)}$ (Ω)	$R_{(ct)}$ (Ω)	W(1/Ω sqrt (Hz))	Q (F)	N
PAni	8.4	1.1×10^{1}	5.6×10^{1}	2.1×10^{-3}	3.5×10^{-1}
PPy	8.3	1.56×10^{1}	9.9×10^{1}	9.2×10^{-6}	6.1×10^{-1}
Poly Ani/py	10.1	1.38×10^{1}	7.5×10^{1}	8.2×10^{-6}	5.1×10^{-1}

Another important parameter in EIS is the double layer capacity that is formed on the electrode/electrolyte surface and has an electron exchange feature. According to the calculations, the double layer (Q) capacity of graphite electrode modified with PAni is more electrically charged with more electron exchange and the double layer capacity of the graphite electrode modified with PPy is the least and has less electron exchange.[25, 34, 35]

CONCLUSION

Result has shown that homo and copolymer of aniline and pyrrole electrosynthesized in citric acid in the presence of $CaCl_2$ have significant antioxidant activity via ferric ion reduction. The structural analysis by FT-IR confirms the formation of homo and copolymer of aniline and pyrrole. The UV-vis spectra demonstrate the antioxidant properties of copolymer whenever the intense blue-colored is developed at the absorption maximum of 593 nm by reduction of [Fe^{+3}-TPTZ] complex to [Fe^{+2}-TPTZ]. In addition, biologists have used Fe^{+3}-TPTZ complex as a measure of the antioxidant effect of compounds known as ferric reducing antioxidant power (FRAP). The more absorbance amount indicate that on the greater the antioxidant effect. Reduction of Fe (III) to Fe (II) using homo and copolymer aniline and pyrrole was not only investigated, but, according to biotechnology researchers, it can be concluded that homo and copolymer aniline and pyrrole have a good antioxidant effect. Additionally, EIS studies show the polymer's ability to charge transfer. The results showed a high sensitivity of electrosynthesized homo and copolymer of aniline and pyrrole proportional to Fe content.

Acknowledgment: The authors of the paper thank direct financial and spiritual support provided by the head of the University and deputy of research and technology of Mahshahr Branch and Yadegar Imam Khomeini Shahre Rey branch of Islamic Azad University, sincerely.

REFERENCES

1. G. Anantha-Iyengar, K. Shanmugasundaram, M. Nallal, K.-P. Lee, M. J. Whitcombe, D. Lakshmi and G. Sai-Anand, *Prog. Polym. Sci.*, **2019**, *88*, 1-129.
2. G. Wang, Y. Yu, H. Liu, C. Gong, S. Wen, X. Wang and Z. Tu, *Fuel Process. Technol.*, **2018**, *179*, 203-228.
3. A. A. Mohamad, *Solar Energy*, **2019**, *190*, 434-452.
4. P. A. Kilmartin, M. Gizdavic-Nikolaidis, Z. Zujovic, J. Travas-Sejdic, G. A. Bowmaker and R. P. Cooney, *Synth. Met.*, **2005**, *153*, 153-156.
5. D. Granato, F. Shahidi, R. Wrolstad, P. Kilmartin, L. D. Melton, F. J. Hidalgo, K. Miyashita, J. v. Camp, C. Alasalvar, A. B. Ismail, S. Elmore, G. G. Birch, D. Charalampopoulos, S. B. Astley, R. Pegg, P. Zhou and P. Finglas, *Food Chem.*, **2018**, *264*, 471-475.
6. A. V. Nand, S. Swift, B. Uy and P. A. Kilmartin, *J. Food Eng.*, **2013**, *116*, 422-429.
7. K. I. Berker, K. Güçlü, İ. Tor and R. Apak, *Talanta*, **2007**, *72*, 1157-1165.
8. A. M. Jones, A. S. Kinsela, R. N. Collins and T. D. Waite, *J. Hazard. Mater.*, **2016**, *320*, 143-149.
9. K. Lokesh, M. Venkataranganna, G. G. Raj, H .Patil and H. Dave, *J. Trace Elem. Med Biol.*, **2018**, *45*, 114-124.
10. K. M. Naji, F. H. Thamer, A. A. Numan, E. M. Dauqan, Y. M. Alshaibi and M. R. D'Souza, *Heliyon*, **2020**, *6*, e03162.
11. A. Jones, S. Pravadali-Cekic, G. R. Dennis, R. Bashir, P. J. Mahon and R. A. Shalliker, *Anal. Chim. Acta*, **2017**, *967*, 93-101.
12. I. F. Benzie and J. J. Strain, *Anal. Biochem.*, **1996**, *239*, 70-76.
13. T. Suktham, A. Jones, A. Soliven, G. R. Dennis and R. A. Shalliker, *Microchem. J.*, **2019**, *149*, 104046.
14. B. L. Sprague and J .G. McNally, *Trends Cell Biol.*, **2005**, *15*, 84-91.
15. H. Baniasadi, A. Ramazani S.A, S. Mashayekhan and F. Ghaderinezhad, *Synth. Met.*, **2014**, *196*, 199-205.
16. C. Basavaraja, W. J. Kim, Y. D. Kim and D. S. Huh, *Mater. Lett.*, **2011**, *65*, 3120-3123.
17. N. Sahiner and S. Demirci, *React. Funct. Polym.*, **2016**, *105*, 60-65.
18. A. Parsa and S. A. Salout, *J. Electroanal. Chem.*, **2016**, *760*, 113-118.
19. K. M. Govindaraju and V. C. A. Prakash, *Colloid Surface A*, **2015**, *465*, 11-19.
20. L. Hostert, G. de Alvarenga, M. Vidotti and L. F. Marchesi, *J. Electroanal. Chem.*, **2016**, *774*, 31-35.
21. H. Yuvaraj, M. H. Woo, E. J. Park, Y. T. Jeong and K. T. Lim, *Eur. Polym. J.*, **2008**, *44*, 637-644.
22. S. Radhakrishnan and S. Paul, *Sensors and Actuators B: Chemical*, **2007**, *125*, 60-65.

23. A. Parsa, M. Sadeghi, M. Maleki, S. Parhizkar and S. Ab Ghani, *Electrochim. Acta*, **2014**, *127*, 34-38.
24. A. Parsa and S. Ab Ghani, *J. Electrochem. Soc.*, **2009**, *156*, E105-E111.
25. A. Parsa, M. Sadeghi, Z. Parsa, A. Shakeri, M. Tehrani and S. Ab Ghani, *J. Iran Chem. Soc.*, **2015**, *12*, 889-895.
26. A. A. Rogachev, M. A. Yarmolenko, A. V. Rogachev, J. Xiaohong, H. Cao, E. N. Lysenko and A. P. Surzhikov, *Appl. Surf. Sci.*, **2019**, *483*, 19-25.
27. S. J. Choi and S. M. Park, *J. Electrochem. Soc.*, **2002**, *149*, E26-E34.
28. P. Xu, X. J. Han, C. Wang, B. Zhang and H. L. Wang, *Synth. Met.*, **2009**, *159*, 430-434.
29. G. Yue and E. Asselin, *Electrochim. Acta*, **2014**, *146*, 307-321.
30. S. Dudonné, X. Vitrac, P. Coutiere, M. Woillez and J.-M. Mérillon, *J. Agric. Food. Chem.*, **2009**-1768, 57, 1774
31. M. Ates, *Prog. Org. Coat.*, **2011**, *71*, 1-10.
32. H. Nara, D. Mukoyama, R. Shimizu, T. Momma and T. Osaka, *J. Power Sources*, **2019**, *409*, 139-147.
33. J. Stojadinović, M. Fan, A. Battistel and F. LaMantia, *ChemElectroChem*, **2015**, *2*, 1031-1035.
34. L. F. Macía, M. Petrova and A. Hubin, *J. Electroanal. Chem.*, **2015**, *737*, 46-53.
35. C. T. P. da Silva, V. L. Kupfer, G. R. da Silva, M. Pereira and A. W. Rinaldi, *Int. J. Electrochem. Sci*, **2016**, *11*, 5380-5394.