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CHARACTERIZATION OF INDICATOR ELECTRODE AS A CHOLESTEROL SENSOR MADE FROM CONDUCTIVE POLYMER PANI-P-TOLUENE SULFONATE

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ABSTRACT

A study has been conducted on the modification of indicator electrode membranes as urea sensors in four membrane layers. The first layer is a urease enzyme in a PVA polymer, the second layer is glutaraldehyde GA as a cross-linker between the first layer, and the third layer is a conductive polymer, PANI-p-toluene sulfonate or PANI-benzene sulfonate. The fourth layer is a 61% PVC-KTp-ClPB-O-NPOE polymer. These layers are sequentially coated from the tungsten indicator electrode. Then, continued on the cholesterol sensor by dissolving 6 mg of ChOx enzyme (50% water: 50% alcohol) in 5 mL, which is dropped one drop into a solution of 0.0350 g PVA, 10 mL of warm water that has been cooled. The purpose of this study was to determine the confidence level R^2, sensitivity, detection range, and detection limit. These indicator electrodes were analysed using potentiometric cells from each indicator electrode whose third layer is made of PANI-p-toluene sulfonate or PANI-benzene sulfonate and a reference electrode. The confidence level R^2 = 99.49%, sensitivity 20.82 mV/div, detection range 10^{-7} – 10^{-2} M and detection limit 10^{-7} M of the indicator electrode made from PANI-p-toluene sulfonate conductive polymer. The confidence level R^2 = 99.12%, sensitivity 38.84 mV/div, detection range 10^{-7} – 10^{-4} M and detection limit 10^{-7} M of the PANI-benzene sulfonate conductive polymer. According to the confidence level and detection limit, the best results were obtained on the tungsten indicator electrode made from PANI-p-toluene sulfonate.

Keywords: Coating; Sensitivity; Tungsten; PANI-benzenesulfonic Acid; PANI-p-toluenesulfonic Acid.
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INTRODUCTION

The characterisation of the cholesterol analyte indicator electrode was carried out based on research on the urea analyte indicator electrode, namely, on the electrode membrane consisting of four layers. The layers use PVA polymer in the first layer, glutaraldehyde (GA) in the second layer, PANI conductive polymer with p-toluene sulfonic acid or benzene sulfonic acid in the third layer, and PVC polymer with KTp-ClPB-o-NPOE in the fourth layer. Each material is placed according to its function: PVA as an encapsulation and entrapment of the cholesterol enzyme, GA as a cross-link between PVA cholesterol enzyme ChOx with PANI p-toluene sulfonate acid or PANI benzene sulfonate acid, PVC with KTp-ClPB-o-NPOE as the active substance, KTp-ClPB forms an anion, which is a negatively charged ion, while o-NPOE 61% as a PVC plasticiser. In the urea analyte indicator electrode as a urea sensor, an increase in sensitivity, detection range and detection limit from the same coating pattern was continued on the cholesterol analyte indicator electrode as a cholesterol sensor. To increase sensitivity, detection range and detection limit, a third layer of PANI acid p-toluene sulfonate or PANI acid benzene sulfonate was used. The method used at this stage is still the potentiometric biosensor method; after obtaining good results, the voltammetric method was continued. The steps to the potentiometric biosensor method were first carried out by synthesising and characterising the cholesterol ChOx enzyme with UV-vis[1], characterising the pattern of four layers of indicator electrodes (tungsten) with SEM-EDX, and the pattern of four layers of membranes on glass with FTIR. Next, characterisation and analysis were carried out with potentiometric cells on each of the 0.001M KH_2PO_4 + KCl phosphate buffer solutions[2] and the cholesterol ChOx enzyme in the range of 10^{-7}, 10^{-6}, 10^{-5}, 10^{-4}, 10^{-3}, 10^{-2} and 10^{-1} M. The KH_2PO_4 phosphate buffer was used because it has a high ionic strength and is low-cost.[3] There are two indicator electrodes analysed, namely (a)PVA-ChOx/GA/PANI-p-toluene sulfonic acid/PVC-KTpClPB-o-NPOE, denoted by PANI-p-

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toluene sulfonic acid, (b)PVA-ChOx/GA/PANI-benzene sulfonic acid/PVC-KTpClPB-o-NPOE, denoted by PANI-benzene sulfonic acid.

EXPERIMENTAL

Material and Methods

The materials used were 1.0 mm diameter tungsten wire, p-toluene sulfonic acid, benzene sulfonic acid, Polyvinyl alcohol (PVA), Polyaniline (PANI), Polyvinyl chloride (PVC), ChOx cholesterol enzyme, all obtained from Sigma-Aldrich.

General Procedure

SEM-EDX (Bruker-129 eV Zeiss) is a tool used for morphological analysis and mass and atomic composition of an element, UV-Vis (Rayleigh UV-1601) for analysis of UV-A, UV-B and UV-C absorbance spectrum patterns, XRD-6100 (Shimazu) for analysis of XRD spectrum pattern materials, and FTIR for analysis of transmittance spectrum patterns.

Detection Method

Membrane matrix with W_{PVA}: W_{PVC} ratio is 1:1[4]; PVC: active substance-1:2[5], as the active substance is $KTpCl_2PB$, o-NPOE plasticiser used 61%, namely 0.0647 mg, cholesterol enzyme ChOx with 1: 1, namely 2.5 mL alcohol, water 2.5 mL alcohol. PVA 0.0350 g was dissolved in 10 mL of hot water until completely dissolved, then cooled. Then, one drop of ChOx enzyme was dropped into the cooled PVA solution, as a PVA-ChOx solution. Glutaraldehyde 2.9 g was dissolved in 100 mL of water, as a GA solution. PANI-p-toluene sulfonate and PANI-benzene sulfonate, each at 2 M, and PVC-$KTpCl_2PB$-o-NPOE in 10 ml THF with a composition of 0.0350 g PVC, 0.0700 g $KTpCl_2PB$, and 0.0647 mg of 61% o-NPOE. Four solutions were used, each representing the order of membrane coating on tungsten for SEM-EDX analysis and on glass for XRD and FTIR analysis. The first layer was PVA-ChOx, the second layer was 2.9% GA, the third layer was PANI-p-toluene sulfonic acid or PANI-benzene sulfonic acid, and the fourth layer was PVC-$KTpCl_2PB$-o-NPOE.

RESULTS AND DISCUSSION

The morphology of the indicator electrode membrane is of two types, namely (1) PVA-ChOx1/GA/PANI-p-toluene sulfonic acid/PVC-KTp-CLPB-o-NPOE, denoted as PANI-p-toluene sulfonic acid; (2) PVA-ChOx-1/GA/PANI-benzene sulfonic acid/PVC-KTp-CLPB-o-NPOE, denoted as PANI-benzene sulfonic acid, as can be seen in Fig.-1, with magnifications of 3000x and 5000x with a voltage of 10 kV. More pores appear in the PANI-benzene sulfonic acid sample compared to PANI-p-toluene sulfonic acid. Similarly, PANI-benzene sulfonic acid with an intensity of 32000 (a.u) has a small energy of 2 kV and PANI-p-toluene sulfonic acid with an intensity of 11000 (a.u) has an energy greater than 2 kV, Fig.-2.

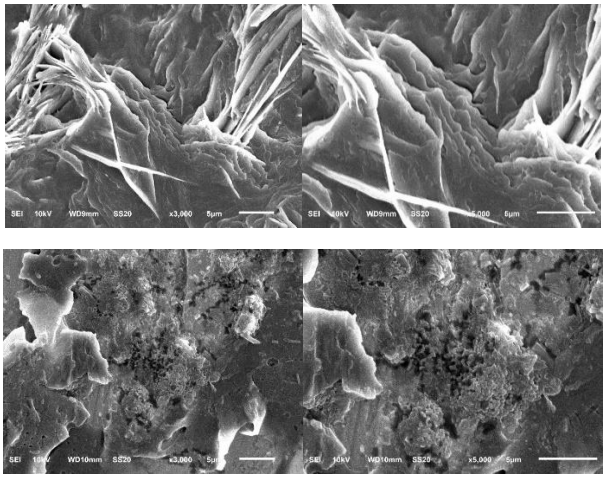

Fig.-1: SEM Morphology of (a) PANI-p-Toluene Sulfonic Acid, (b) PANI-Benzene Sulfonic Acid

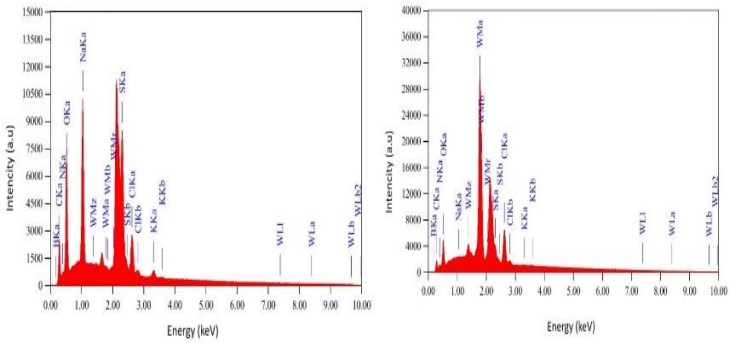

Fig.-2: EDX Spectrum Patterns of Indicator Electrodes (a) PANI-p-Toluene Sulfonic Acid, (b) PANI-Benzene Sulfonic Acid

Table-1: Mass and Atomic Composition of edx for PANI-Benzene Sulfonic Acid

Element	Mass Composition (%)	Atomic Composition (%)
B	00.04	00.24
C	0.17	0,84
N	01.09	05.06
O	07.29	29.57
Na	00.05	00.13
Cl	09.48	17.36
K	00.17	00.28
W	78.23.00	27.62

The largest mass and atomic composition in the electrodes (a) PANI-benzene sulfonate acid, (b) PANI-p-toluene sulfonate on each tungsten element in Table-1 and Table-2. This is supported by the morphology of PANI-p-toluene sulfonate, which has more pore flowers that can be seen from the magnification of 3000x and 5000x. Likewise, the EDX intensity spectrum pattern has more peaks in the PANI-p-toluene sulfonate electrode, even though the intensity is smaller than that of the PANI-benzene sulfonic acid electrode. While the peak intensity of the PANI-benzene sulfonate acid electrode is only one, which is very high compared to that of the PANI-benzene sulfonate acid electrode.

Table-2: Mass and Atomic Composition of edx for PANI-p-Toluene Sulfonate Acid

Element	Mass Composition (%)	Atomic Composition (%)
B	0,40	0,64
C	0,90	1,26
N	0,09	0,09
O	1,20	1,26
Na	0,59	0,42
S	0,35	0,19
Cl	0,35	0,19
K	0,11	0,04
W	96,00	95,9

FTIR Characterization

There are differences in the FTIR spectral patterns caused by the two types of acids in the PANI conductive polymer. In the PANI-p-toluene sulfonic acid electrode membrane, the % transmittance spectrum pattern increases, while in the PANI-benzene sulfonic acid electrode membrane, the % transmittance decreases, as can be seen in Fig.-3.

Pure PVA exhibits strong band formation at 3430 – 3244 cm^{-1}, which is associated with O-H stretching vibration. Asymmetric and symmetric stretching vibration of the C–H bond at 2941 and 2855 cm^{-1}. The doping level depends on the position and height of certain IR absorption peaks to identify the structural modification of the PVA main chain.[6]

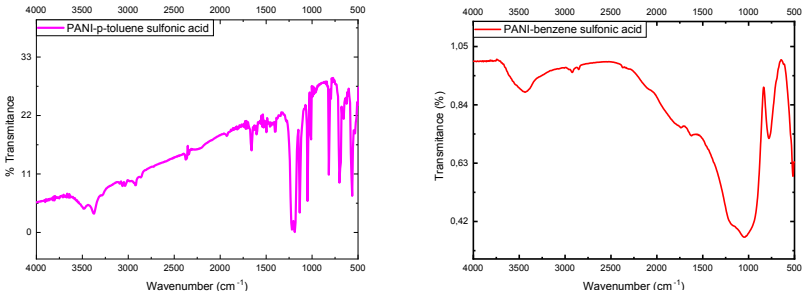

Fig.-3: FTIR Spectral Patterns of (a) PANI-p-Toluene Sulfonic Acid, (b) PANI-Benzene Sulfonic Acid

O-H stretching vibration peaks at 3435cm^{-1} and 3310cm^{-1}, hydroxyl groups at 3450cm^{-1}, free hydroxyl groups of PVA at 3570cm^{-1} [7]. GA pattern peaks at 3297.5cm^{-1} (N-H) stretching group, at 1158.7cm^{-1} (C–O) stretching group, (C–H) band peak at 1425cm^{-1}, (N–H) bending peak at 1506cm^{-1}, O-H group at 3359cm^{-1}.[8] -OH and $-CH_3$ stretching groups were observed at 3450 and 2850cm^{-1}, at 1377 and 1416cm^{-1} represented C=O and C–O stretching vibrations of the group ionized carboxyl and at 1066 and 1127cm^{-1} the C-Ostretching vibration of the hydroxyl group together with the S=O stretching vibration at1239cm^{-1}.[9] PANI chain at % transmission peak at wave number 1299 cm^{-1}, % transmission peak at wave number 500, 621 and 1032 cm^{-1} indicates the presence of sulfonate group in PANI from p-toluene sulfonate, % transmission peak at 735, 707 and 757 cm^{-1} indicates the spectrum of benzene ring.[10,11,12] Benzene ring at 1164, 690 cm^{-1} [13], C-H stretching band peak at 742 cm^{-1} is the characteristic peak of PANI benzene ring, characteristic peak at 1686 cm^{-1} stretching -C=C- benzene ring, at 1570 cm^{-1} N–H bond stretching vibration. FTIR spectrum of PVC 646, 815, 1335 and 1390 cm^{-1},[14], peak at 1735 cm^{-1}, peak at 1710 cm^{-1}. C-Cl stretching peak at 835 cm^{-1}.[15]

Characterisation of Potentiometer Cells

The FTIR spectrum pattern in Fig.-3 is similar to the buffer analysis at various concentrations of 10^{-7}, 10^{-6}, 10^{-5}, 10^{-4}, 10^{-3}, 10^{-2} and 10^{-1} M against time in Fig.-4, both for the PANI-p-toluene sulfonic acid and PANI-benzene sulfonic acid polymer membranes. The potentiometer voltage increases when the potentiometer cell contains a solution with a KH_2PO_4 buffer concentration of 10^{-7} -10^{-2} M for the PANI-p-toluene sulfonic acid membrane (Fig.-4a); while for the PANI-benzene sulfonic acid membrane, it decreases from a buffer concentration of 10^{-7} - 10^{-4} M (Fig.-4b). The voltage response over time of the solution at buffer molarity conditions (biosensor interference) is very influential, as seen in Fig.-4a and 4b. The real-time response to concentration variations greatly affects sensitivity.[16]

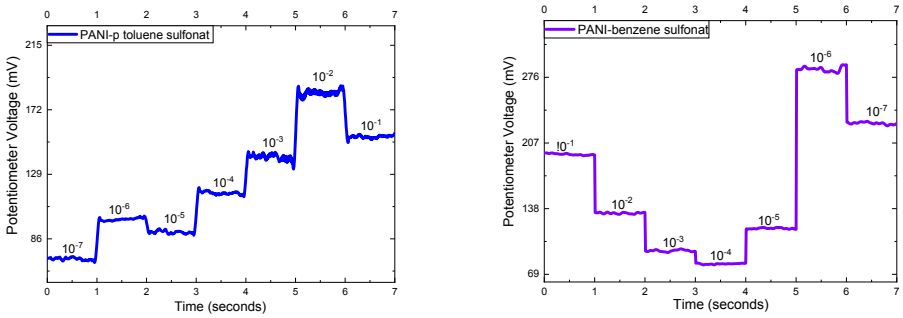

Fig.-4: Effect of Buffer at Concentrations of 10^{-7}, 10^{-6}, 10^{-5}, 10^{-4}, 10^{-3}, 10^{-2} and 10^{-1} M in a Time Span of One Second For Indicator Electrodes (a) PANI-p-Toluene Sulfonic Acid, (b) PANI-Benzene Sulfonic Acid

The voltage versus time graph patterns in Fig.-4a and 4b were converted to voltage versus -log buffer concentration graph patterns, aiming to obtain a linear curve. The linear curve is used to determine sensitivity, detection range, detection limit, and confidence level R^2. Figure-5a shows that for the PANI-p-toluene sulfonate acid indicator electrode, the sensitivity is 20.82 mV/div, the detection range is 10^{-7} – 10^{-2} M, the detection limit is 10^{-7} M, and the confidence level is 99.67%. Figure-5b shows that for the PANI-benzene sulfonate acid indicator electrode, the sensitivity is 36.84 mV/div, the detection range is

10^{-7} – 10^{-4} M, the detection limit is 10^{-7} M, and the confidence level is 99.12%. Thus, the best results were obtained on the PANI-p-toluene sulfonate acid indicator electrode.

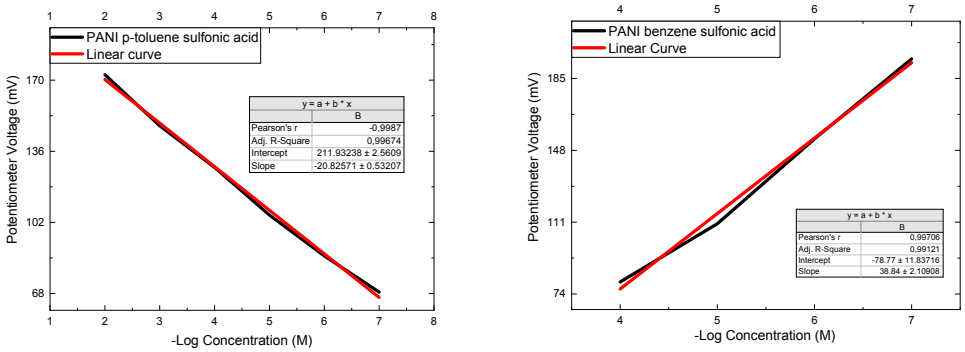

Fig.-5: Linear Curves of Potentiometer Cell Analysis Indicator Electrodes for (a) PANI-p-Toluene Sulfonic Acid, (b) PANI-Benzene Sulfonic Acid

According to 17, those with a low detection range have high sensitivity. Phosphate buffer plays a role in maintaining pH balance in the body, while cholesterol is a fat that has an important role in various cellular functions, such as building cell membranes and producing hormones. Potentiometer cell analysis using phosphate buffer is appropriate, only need to increase the sensitivity of the conductive polymer parameters and the type of acid used. PANI conductivity 10 Scm^{-1}, Polythiophene (PTH) conductivity 100 Scm^{-1} (Siemens per centimetre), PANI conductivity 0.0045 Scm^{-1} [18]. SWCNT/PANI composite super high conductivity of 4000 Scm^{-1}.[19]

CONCLUSION

Based on SEM-edx analysis, the mass and atomic composition are greater in the PANI-p-toluene sulfonic acid element, as well as in (a) % Transmittance increases linearly, (b) the effect of buffer at concentrations of 10^{-7}, 10^{-6}, 10^{-5}, 10^{-4}, 10^{-3}, 10^{-2} and 10^{-1} M in a time span of 1 second for the indicator electrode, and (c) Linear curve of the potentiometer cell analysis indicator electrode. The best results were obtained on the PANI-p-toluene sulfonic acid indicator electrode with a sensitivity of 20.82 mV/div, a detection range of 10^{-7} – 10^{-2} M, a detection limit of 10^{-7} M and a confidence level of 99.67%.

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CONFLICT OF INTERESTS

The authors have no conflict of interest to declare.

AUTHOR CONTRIBUTIONS

The present work is related to *SDG-9: Industry, Innovation, and Infrastructure*. All the authors contributed significantly to this manuscript, participated in reviewing/editing and approved the final draft for publication. The research profile of the authors can be verified from their ORCID ids, given below:

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REFERENCES

1. S. Abd Hakim, E. M. Ginting, A. M. Siregar, Rugaya, and A. Rais, *Rasayan Journal of Chemistry*, **18(2)**, 882(2025), http://doi.org/10.31788/RJC.2025.1829099
2. S. Brudar and B. Hribar-Lee, *Biomolecules*, **9(2)**, 65(2019), https://doi.org/10.3390/biom9020065
3. Y. H. Cheng, C. Chande, Z. Li, N. Haridas Menon, S. Kaaliveetil, and S. Basuray, *Biosensors*, **13(12)**, 999(2023), https://doi.org/10.3390/bios13120999
4. M. A. Rahman, P. Kumar, D. S. Park, and Y. B. Shim, *Sensors*, **8(1)**, 118(2008), https://doi.org/10.3390/s8010118
5. S. Abd Hakim, M. Rianna, and A. Rais, Mater. Sci. Energy Technol., *Materials Science for Energy Technologies* **6**, 89(2023), https://doi.org/10.1016/j.mset.2022.11.010
6. S. S. Nafee, T. A. Hamdalla, and S. A. Shaheen, *Phase Transitions, Research Article*, **90(5)**, 439(2017), https://doi.org/10.1080/01411594.2016.1260722
7. Y. T. Wang, Y. Wang, J. Guo, and Q. X. Zhou, *Journal of Macromolecular Science, Part B Physics* **59(1)**, 49(2020), https://doi.org/10.1080/00222348.2019.1687137
8. A. Sethi, M. Ahmad, T. Huma, and W. Ahmad, *Drug Delivery*, **28(1)**, 1569(2021), https://doi.org/10.1080/10717544.2021.1944398
9. S. Akhshabi, E. Biazar, V. Singh, S. H. Keshel, and N. Geetha, *International Journal of Nanomedicine*, **13,** 4405(2018), https://doi.org/10.2147/IJN.S165739
10. M. Bounedjar, N. Naar, and A. Mekki, *Journal of Macromolecular Science, Part B, Physics,* **60(3)**, 190(2021), https://doi.org/10.1080/00222348.2020.1843849
11. M. Beygisangchin, S. A. Rashid, S. Shafie, and A. R. Sadrolhosseini, *Materials*, **14(23)**, 7382(2021), https://doi.org/10.3390/ma14237382
12. A. M. Herrera González, M. Caldera Villalobos, J. Garciá-Serrano, and A. A. Pelaéz Cid, Des. *Designed Monomers and Polymers*, **19(4)**, 330(2016), https://doi.org/10.1080/15685551.2016.1152543
13. P. Chaubisa, D. Dharmendra, Y. Vyas, P. Chundawat, N. K. Jangid, and C. Ameta, *Polymer Bulletin*, **81**, 3333(2024), https://doi.org/10.1007/s00289-023-04873-8
14. A. H. Alshammari, M. Alshammari, M. Ibrahim, K. Alshammari, and T. A. M. Taha, *Optics & Laser Technology*, **168**, 109833(2024), https://doi.org/10.1016/j.optlastec.2023.109833
15. M. M. Ghobashy et al., *Scientific Reports*, **13**,7514(2023), https://doi.org/10.1038/s41598-023-34583-0
16. F. Khosravi, S. M. Loeian, and B. Panchapakesan, *Biosensors*, **7(2)**, 17(2017), https://doi.org/10.3390/bios7020017
17. H. Alrabiah, E. A. Ali, R. A. Alsalahi, M. W. Attwa, and G. A. E. Mostafa, *Polymers*, **15(13)**, 2808(2023), https://doi.org/10.3390/polym15132808
18. M. E. Martínez-Cartagena *et al.*, *Polymers*, **14(18)**, 3913(2022), https://doi.org/10.3390/polym14183913
19. R. Wu, H. Yuan, C. Liu, J. Le Lan, X. Yang, and Y. H. Lin, *RSC Advances*, **8**, 26011(2018), https://doi.org/10.1039/C8RA04863K

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