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Novel membrane sensor for determination of lamotrigine in pharmaceuticals and urine

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A B S T R A C T

Lamotrigine (LMT), chemically known as [6-(2,3-dichlorophenyl)-1,2,4-triazine-3,5-diamine], is a broad spectrum antiepileptic drug, used as monotherapy and as an adjunct with other antiepileptic drugs for treatment of partial and generalized toxic-clonic seizures. It is used to treat neurological lesions and as a tranquilizer. A selective electrochemical membrane sensor has been developed and validated for determination of LMT. The membrane constructed using LMT and molybdophosphoric acid in THF and PVC is applicable for the detection of 5×10^{-4} to 9×10^{-3} M LMT in the pH range between 4.6 and 5.8 with the Nernstian slope of 57.14±1 mV/decade. The regression coefficient value of 0.9932 showed a good linear correlation between the concentrations of LMT and measured cell potentials. The limits of detection (LOD) and quantification (LOQ) values for the fabricated sensor were 1.3×10^{-5} and 4×10^{-5} M LMT, respectively. Various experimental conditions were optimized to reach the effective performance characteristics of the sensor. The effect of various cations, anions and organic species on the performance of sensor was studied by following standard-addition procedure. The results revealed no such variations due to presence of foreign ions or species. The fabricated sensor was subjected to validation to check accuracy, precision, robustness and ruggedness. The mean accuracy for determination of LMT was found to be 99.16%. The developed sensor was successfully used to determine LMT in tablets and in spiked human urine.

1. Introduction

Lamotrigine (LMT) is chemically known as 3,5-diamino-6-(2,3-dichlorophenyl)-as-triazine (**Fig. 1**), is an anticonvulsant drug used in the treatment of epilepsy and bipolar disorder. It is also used off-label as an adjunct in treating clinical depression.

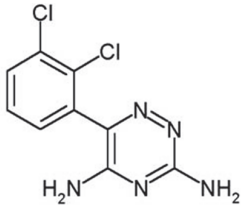

Fig. 1. Chemical structure of LMT

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As an antiepileptic drug LMT is attracted by many analysts. The monograph in the United States Pharmacoepia (USP)[1] described a chromatographic procedure for determination of LMT using monobasic potassium phosphate buffer, triethylamine and acetonitrile as mobile phase. The drug LMT in body-fluids was analysed by various researchers utilizing variety of chromatographic techniques[2-17]. Various methods have been reported for its determination in pharmaceuticals and they are titrimetry with acetous perchloric acid in anhydrous acetic acid medium[18], visible spectrophotometry[19-21], UV-spectrophotometry[22], planar chromatography[2], thin layer chromatography[23], high performance liquid chromatography[24-25], ultra performance liquid chromatography[26] and adsorptive stripping voltammetry[27]. Two types of LMT ion-selective electrode is reported by Gupta et al. [28]. These electrodes are based on PVC membranes doped with LMT-tetraphenyl borate (TPB) or LMT-phosphotungstic acid (PT) ion-pair complexes as molecular recognition materials. The electrodes are used for determination of LMT in urine and plasma. There is also a report for LMT determination using molecularly imprinted polymers[29] using methacrylic acid as the functional monomer, ethylene glycol dimethacrylate as crosslinker, 2,2'-azo-bis-iso-butyronitrile as the initiator in 4:3 (v/v) tetrahydrofuran and acetonitrile. The method is applicable to determine LMT over the concentration range from 1×10^{-6} to 1×10^{-3} M in its pure state and in real samples. Though the reported procedures are specific to determine LMT, they failed to present outcomes of detailed validation aspects.

Research in the field of development of potentiometric sensors is gaining more and more attention and a number of potentiometric sensors have been developed for the determination of species in the areas of chemical, pharmaceutical and biomedical analyses[30-41]. Potentiometric sensors have extensive applications to quantify the compounds since they neither require sophisticated instrument nor relying on stringent experimental conditions. Therefore, an attempt is made to develop a novel potentiometric membrane sensor for the determination of LMT in pharmaceuticals and spiked human urine. The membrane sensor is fabricated by preparing the ion pair complex of LMT with molybdophosphoric acid and its membrane with polyvinyl chloride in THF. Different parameters are optimized to improve the selectivity of membrane for accurate and precise determination of LMT. The fabricated sensor is used to determine LMT in pharmaceuticals and spiked human urine.

2. Experimental

2.1. Apparatus

A digital dual channel potentiometer (PICO Ltd., Chennai, India), Ag/AgCl reference electrode and copper wire were used for potential measurements.

2.2. Materials and methods

The chemicals and reagents used were of analytical grade. Distilled water was used throughout the work. The pure LMT (99.8%) was kindly provided by Torrent Pharmaceuticals Ltd (Mumbai, India). Lamitor-DT tablets (100mg LMT/tablet) (Indrad-382721, Mehsana, India) were purchased from local commercial sources. Dodeca-molybdophosphoric acid (PMA), tetrahydrofuran (THF) and polyvinyl chloride (PVC) were supplied by S. D. Fine Chem Ltd, Mumbai, India. Concentrated sulphuric acid (H_2SO_4) (98% v/v, Sp. gr. 1.84) was supplied by Merck, Mumbai, India. Urine sample was collected from a 21 year male healthy volunteer, it was filtered and diluted ten times with water before use.

A 0.1 M solution of H_2SO_4 was prepared by diluting suitable volume of concentrated H_2SO_4 with water. A 0.01 M PMA solution was prepared by dissolving calculated amount the compound in distilled water. Solutions of 1mM each of Na_2CO_3, $NaHCO_3$, NaOH, CH_3COOH, CH_3COONH_4, KSCN, sucrose, fructose, glucose, maltose, starch, lactose, glycine, sodium fluoride, calcium chloride, nickel chloride, potassium chloride, ammonium chloride, cadmium chloride and cobalt chloride were prepared

by dissolving required weight of the respective compound (all from S.D. Fine Chem Ltd., Mumbai, India) in distilled water.

2.3. Preparation standard LMT solution

A standard solution of 0.01M LMT was prepared by accurately dissolving calculated quantity of pure drug in 100 mL of 0.1M H_2SO_4 in a volumetric flask.

2.4. PROCEDURE

2.4.1. Fabrication of the sensor

A mixture of 20 mL of each of 0.01M solutions of LMT and PMA was stirred for 20 minutes and the resulted precipitate was collected on Whatmann No. 41 filter paper by filtration. The precipitate was dried overnight at room temperature. A 20 mg of dried precipitate was taken in a Petri Dish of 4 cm width, about 0.1g of PVC and 10ml of THF were added. The content after mixing was allowed to evaporate under room temperature for 24 hours. The dried membrane was fused to one end of non-conducting glass tube with the aid of THF. The dried tube was filled by 3-5 mL internal solution of 0.01M LMT. A pure copper wire of 2.0 mm diameter and 15 cm length was tightly insulated leaving 1.0 cm at one end and 0.5 cm at other end for connection. One terminal of the wire was inserted into internal solution and the other terminal was connected to the potentiometer. The sensor was conditioned by soaking in analyte solution for 6 hours.

2.4.2. Preparation of calibration curve

Into a series of 10.0ml volumetric flasks varying aliquots (0.0, 0.5, 1.0, 1.5, 3.0, 4.5, 6.0, 7.5 and 9.0 ml) of 0.01M standard LMT solutions were placed with the help of a microburet. The volume of each flask was adjusted to 10 mL with water. The potential of each solution was measured by using LMT-PMA sensor *versus* Ag/AgCl reference electrode.

The calibration graph of measured potential *versus –log* [LMT] was prepared. The concentration of the unknown was found by using calibration graph or regression equation derived using potential and *–log* [LMT] data.

2.4.3. Procedure for interference study

In a 10 ml volumetric flask, 2 ml of 0.01M drug solution and 2ml of 1mM solution of interferent were taken. The solution after adjusting to pH 5 and diluting to the mark, the potentials of each were measured using the electrochemical cell assembled for preparation of calibration curve.

2.4.4. Procedure for tablets

Twenty tablets were weighed and transferred in to a clean dry mortar and powdered. Portion of the tablet powder equivalent to 64.02 mg of LMT was transferred in to a 25 ml volumetric flask and shaken with 20 ml of 0.1M H_2SO_4 for 20 minutes. The content was diluted to the mark with the same solvent, mixed well and filtered through Whatmann No. 41 filter paper. A suitable aliquot was taken and its potential measured by following the procedure described for preparation of calibration curve. The concentration of LMT was calculated using the calibration curve or regression data.

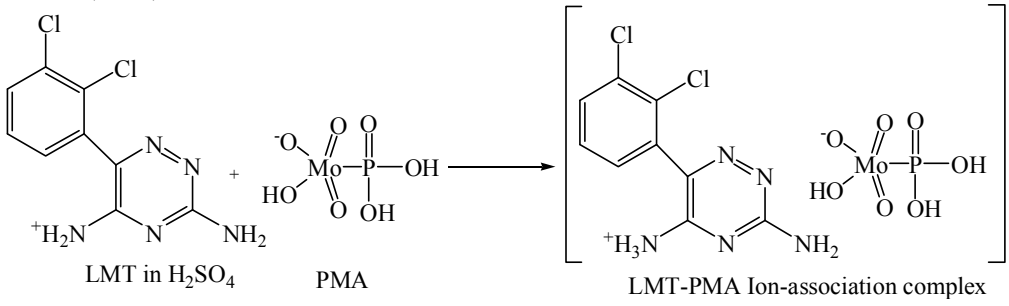
2.4.5. Procedure for spiked human urine

In a 10ml volumetric flask 1ml of 1:10 urine and 2ml of 0.01M LMT solution were taken. The volume was brought to the mark with water and mixed well. After bringing the solution to the optimum pH of 5 the potential of the solution was measured using LMT-PMA sensor and Ag-AgCl reference electrode. The concentration of LMT in the solution was calculated using the calibration curve or regression data.

3. Results and discussion

The development and validation of ion-selective electrodes using membranes is of great interest for pharmaceutical analysis because they offer the advantages of simplicity of fabrication and operation, rapid response time, fair detection limits, acceptable selectivity, accuracy and precision, applicable to the detection of wider concentration range of species in coloured and turbid solutions, and probability to automate and computerize.

The membrane was prepared based on the reaction between aqueous cationic LMT with the solution of dodeca-molybdophosphate (PMA) to form a stable 1:1 water insoluble ion association complex, with low solubility product and suitable grain size precipitate. The probable reaction scheme for the formation of LMT-PMA ion-association complex is given in scheme 1. The formed ion-associate of LMT-PMA was used to fabricate the membrane consisting with poly-vinyl chloride (PVC) using tetrahydrofuran (THF).



Scheme 1. Reaction pathway for formation of LMT- PMA ion-pair complex

The following systematic representation is depicted for the electrochemical cell assembly:

AgCl Reference electrode ‖ LMT-PMA Sensor | 0.01M LMT solution | Cu-Wire

3.1 Optimization of variables

Different experimental variables such as pH, soaking time, response time, stability and effect of interferents were optimized by measuring the potential of the LMT solution of known concentration using the developed sensor.

The optimum pH range of the sensor was found to be from 4.6 to 5.8 and between which the potential measured for each solution of LMT of any concentration within the linear range were almost constant. There were lower potential values observed at pH lesser than 4.6 and 5.8 (**Fig. 2**).

The soaking time was examined by immersing the sensor into a solution of LMT of known concentration for different time periods. From a series of investigations it was found that the average soaking time for the conditioning of the electrode is 6 hours (**Fig. 3**). Therefore, it was found necessary for the sensor to soak in standard analytic solution at least for 6 hours prior to its use for analyses.

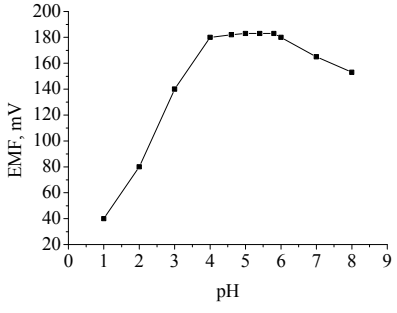


Fig. 2. Effect of pH on EMF (1.0×10^{-3} M LMT)

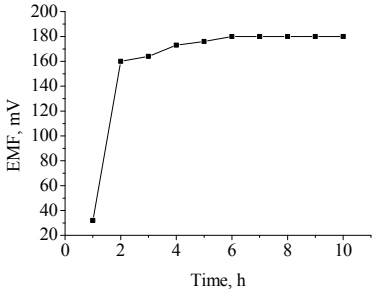


Fig. 3. Effect of soaking time on EMF (1.0×10^{-3} M LMT)

After immersing the LMT-PMA sensor along with the reference electrode into the solution of analyte or containing analyte reproducible and constant potential readings were observed in less than 5 seconds (**Fig. 4**). Therefore, the response time of the sensor was found as 5 seconds.

The developed sensor was subjected to measure the potential of LMT solution in the presence of various organic and inorganic compounds, cations and anions as interferents. The study was undertaken by separately spiking the solutions of 1mM each of intereferent into a 1.0×10^{-3} M LMT solution. This was done in accordance to the IUPAC guidelines[42, 43]. None of the added species affected the potential. This confirmed that the sensor is selective for the determination of LMT in the presence of such charged or neutral species.

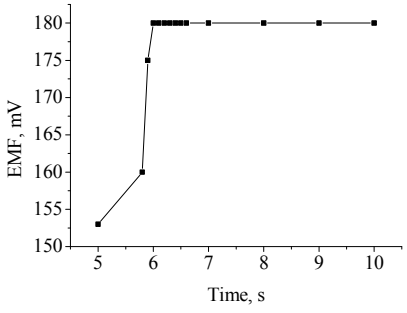


Fig. 4. Study of response time (1.0×10^{-3} M LMT)

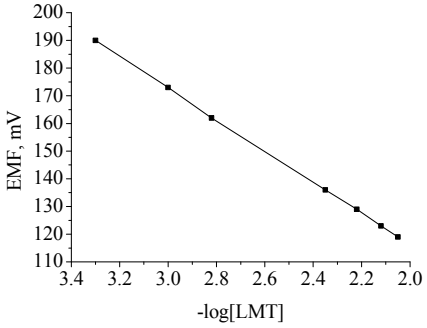


Fig. 5. Calibration graph

3.2. Method validation

The electrochemical response parameters of developed LMT-PMA sensor were evaluated according to IUPAC recommendations[42, 43]. The data obtained are summarized in table 1. The results showed that the sensor provides rapid, stable and linear response for the LMT concentration ranged from 5×10^{-4} to 9×10^{-3} M. The calibration graph (**Fig. 5**) obtained was linear with Nernestian slope of 57.14±1 mV/decade. Stable potentiometric readings were obtained with variations within ±1 mV for the span period of more than a month. The lower limit of detection, calculated from the intercept of the two lines of the calibration graph, is 1.3×10^{-5} M. The other electrochemical features of the sensor have also been presented in Table 1.

Table 1. Electrochemical characteristics of the LMT-PMA sensor

Parameters	**Values**
Linear range, M	5×10^{-4} - 9×10^{-3}
Limit of detection (LOD), M	1.3×10^{-5}
Limit of quantification (LOQ), M	4.0×10^{-5}
Slope (m), mV/decade	57.14±1
Intercept (b), mV	1.7550
Correlation coefficient (r)	0.9932
Response time, s	<5
Working pH range	4.6-5.8
Life span of sensor, days	>30

3.2.1 Accuracy and precision

Accuracy and intra- and inter-day precision were evaluated by analysing pure LMT solutions at three different concentrations in seven replicates during the same day and five replicates during different days. The amount of LMT found was calculated for each measurement. The RE (%) and the RSD (%) values were calculated. The percent relative error which is an index of accuracy is ≤4.50 and

is indicative of acceptable accuracy. The obtained RSD values ranged between 2.11 and 4.34% indicated that the results are prcise enough. The results of this study are presented in Table 2.

Table 2. Results of accuracy and precision study

LMT taken, mmol L^{-1}	Intra-day (n = 7)			Inter-day (n = 5)		
	LMT found, mmol L^{-1}	% RE	% RSD	LMT found, mmol L^{-1}	% RE	% RSD
3.00	3.05	1.67	2.11	3.03	1.00	2.45
6.00	5.73	4.50	2.42	5.84	2.67	4.34
9.00	9.24	2.67	2.39	8.76	2.67	2.36

RE: Relative error; RSD: Relative standard deviation.

3.2.2 Robustness and ruggedness

The robustness of an analytical procedure is a measure of its capacity to remain unaffected by small, but deliberate variations in experimental parameters and provides an indication of its reliability for normal usage. Under deliberately varied experimental conditions [pH: 4.6(±2) – 5.8(±2) and temperature: 25±2 ^{O}C] the %RSD values ranged from 1.11 to 2.12% revealed robustness. In method ruggedness, the analyses with different potentiometers, on different days by different analysts were performed. Such variations did not yield any appreciable changes in the measurement. The inter-instrumental and inter-analysts RSD values of <3.4% declared that the proposed potentiometric sensor is robust enough. The results of robustness and ruggedness studies are presented in Table 3.

Table 3. Results of method robustness and ruggedness study, expressed as %RSD

LMT taken, M	Robustness		Ruggedness	
	Alteration of pH	Alteration of Temperature	Inter-analysts' (n=3)	Inter-instruments' (n=3)
3.00	2.12	1.92	1.89	1.32
6.00	1.11	1.99	1.62	1.87
9.00	1.94	1.20	1.90	1.63

3.2.3 Application to tablets

A 5 mL of 0.01M LMT solution of tablets extract prepared under '*procedure for tablets'* was subjected to analysis by the optimized procedure. The mean measured potential for the tablets extract was found as same as that obtained for the pure drug solution. The results in this study were compared with those of a reference method[1]. The reference method is a chromatographic method and in which LMT is quantified using a mixture of monobasic potassium phosphate buffer, triethylamine and acetonitrile as mobile phase. The accuracy and precision were evaluated by applying Student's *t*- test and variance ratio *F*- test, respectively. The calculated *t*- and *F*- values at 95% confidence level did not exceed the tabulated values and this confirmed insignificant difference between the results of reference and proposed methods. The mean percent recovery of LMT from tablets was found to be 98.9 with RSD value of 3.2%. These results are presented in Table 4.

Table 4. Results of analysis of tablets by the proposed method and statistical comparison of the results with the reference method

Tablet analyzed	Label claim, mg/tablet[a]	Found[b] (Percent of label claim ±SD)	
		Reference method	Proposed method
Lamitor-DT	100	99.67±1.76	98.90±1.56 ***t*** = 0.73 ***F*** = 1.27

[a]Amount in mg per tablet; [b]mean value of 5 determinations.

3.2.4 Recovery study

A standard addition procedure was followed to further assure the accuracy of the sensor. The solutions were prepared by spiking pure drug into a pre-analyzed tablet extract at three different levels and potentials measured using the sensor. To a 2 mL of 0.01M LMT tablets extract, 1, 2 and 3 mL of 0.01 M pure LMT drug solutions were spiked (five replicates), and pH was adjusted. After diluting the solutions to 10 mL, the potentials of each were measured and the amounts of LMT calculated. The recovery of pure LMT was computed. The percentage recovery of LMT from tablets, presented in Table 5, ranged from 98.33 to 102.4% revealed that the sensor is selective to give satisfactory in the presence of excipients.

Table 5. Results of accuracy assessment by recovery test for Lamitor-DT tablets

LMT in tablet, mmol L^{-1}	Pure LMT added, mmol L^{-1}	Total found, mmol L^{-1}	Pure LMT recovered (Percent±SD*)
3.01	1.50	4.54	102.0±1.23
3.01	3.00	5.96	98.33±0.66
3.01	4.50	7.62	102.4±1.18

*Mean value of three measurements

3.2.5 Spiked human urine analysis

From the analysis of spiked human urine sample the percent recovery of LMT were ranged from 93.6 to 98.6% with RSD of <5% indicated that the endogenous substances did not interfere to the assay and hence the sensor is suitable for its use in physiotherapeutic administration of LMT.

4. Conclusions

This is the first paper describing the fabrication of membrane sensor using phosphomolybdic acid and its application to determine lamotragine in pharmaceuticals and spiked human urine. The sensor provides fast and linear Nernestian response over a wide range of lamotragine concentration. The sensor has been successfully used to determine drug content in pure state, tablets and spiked human urine with acceptable recovery. The results obtained were accurate and precise with good agreement to consider the sensor for its use as a tool to determine lamotragine in quality control laboratories. The electrochemical cell's assembly is a simple, low cost and selective tool for direct determination of lamotragine in aqueous media without involving any tedious extraction step.

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References

1. The United States Pharmacopoeia, XXIV rev, National Formulary XIX Rockville, USP Convention, USA, 2000.
2. Dreassi, E., Corbini, G.P., Corti, M., Ulivelli, Rocchi R. **(1996).** Quantitative analysis of lamotrigine in plasma and tablets by planar chromatography and comparison with liquid chromatography and UV spectrophotometry. *J. AOAC. Intl.,* 79(6), 1277.

3. Contin, M., Balboni, M., Callegati, E., Candela, C., Albani, F., Riva, R., Baruzzi, A. **(2005).** Simultaneous liquid chromatographic determination of lamotrigine, oxcarbazepine monohydroxy derivative and felbamate in plasma of patients with epilepsy. *J. Chromatogr. B.,* 828(1-2), 113.
4. Sean, R., Scheuer., Mark L. **(1998).** Determination of lamotrigine in biologic materials by a simple and rapid liquid chromatographic method. *Ther. Drug. Monit.,* 20(2), 209.
5. Matar, K.M., Nicholls, P.J., Bawazir, S.A., Al-Hassan, M.I., Tekle, A. (**1999**). Liquid chromatographic determination of six antiepileptic drugs and two metabolites in microsamples of human plasma. *Ther. Drug. Monit.,* 21(5), 559.
6. Cheng, C.L., Chou, C.H., Hu, O.Y. **(2005).** Determination of lamotrigine in small volumes of plasma by high-performance liquid chromatography. *J. Chromatogr. B.,* , 817(2), 199-206.
7. Matar, K.M., Nicholls, P.J., Bawazir, S.A., Al-Hassan, M.I., Tekle, A. **(1998).** A rapid liquid chromatographic method for the determination of lamotrigine in plasma. *J. Pharm. Biomed. Anal.,* 17(3), 525.
8. Londero, D., Lo Greco. **(1997).** New micromethod for the determination of lamotrigine in human plasma by high-performance liquid chromatography. *J. Chromatogr. B. Biomed. Sci. Appl.,* 691(1), 139.
9. Elizabeth, G.S., Lower, D.R., Virji, M.A., Krasowski, M.D. **(2007).** Simultaneous determination of lamotrigine, zonisamide, and carbamazepine in human plasma by high-performance liquid chromatography. *Biomed. Chromatogr.,* 21(3), 225.
10. Patil, K.M., Bodhankar, S.L. (**2005**). Simultaneous determination of lamotrigine, phenobarbitone, carbamazepine and phenytoin in human serum by high-performance liquid chromatography. *J. Pharm. Biomed. Anal.,* 39(1-2), 181.
11. Saracino, A.P., Maria, A.M., Koukopoulos, G.M., Sani, M.M.A., Raggi, M.A. **(2007).** Simultaneous high-performance liquid chromatographic determination of olanzapine and lamotrigine in plasma of bipolar patients. *Ther. Drug. Monit.,* 29(6), 773.
12. Bartoli, A., Marchiselli, R., Gatti, G. (**1997**). A rapid and specific assay for the determination of lamotrigine in human plasma by normal-phase HPLC. *Ther. Drug. Monit.*, 19(1), 100.
13. Cociglio, M., Alric, R., Bouvier, O. (**1991**). Performance analysis of a reversed-phase liquid chromatographic assay of lamotrigine in plasma using solvent-demixing extraction. *J. Chromatogr.B: Biomed. Sci. Appl.,* 110 (1), 269.
14. Lensmeyer, G.L., Gidal, B.E., Wiebe, D.A. (**1997**). Optimized high-performance liquid chromatographic method for determination of lamotrigine in serum with concomitant determination of phenytoin, carbamazepine, and carbamazepine epoxide. *Ther. Drug Monit.,* 19(1), 292.
15. Angelis-Stoforidis, P., Morgan, D.J., O'Brien, T.J., Vajda, F.J.E. (**1999**). Determination of lamotrigine in human plasma by high-performance liquid chromatography. *J. Chromatogr. B. Biomed. Sci. Appl.,* , 727(1), 113.
16. Croci, D., Salmaggi, A., de Grazia, U., Bernardi, G. (**2001**). New high-performance liquid chromatographic method for plasma/serum analysis of lamotrigine. *Ther. Drug. Monit.,* , 23(6), 665.
17. Castel-BrancoAlmeida, A.M., Falcao, A.C., Macedo, T.A., Caramona, M.M., Lopez, F.G. (**2001**). Lamotrigine analysis in blood and brain by high-performance liquid chromatography. *J. Chromatogr. B. Biomed. Appl.,* 755(1), 119.
18. Rajendra Prasad, N., Basavaiah, K., Vinay, K.B. (**2009**). Determination of drug content of pharmaceuticals containing Lamotrigine by titrimetry in non-aqueous medium. *Proc. Indian Nat. Sci. Acad.,* 75(3), 131.
19. Rajendra Prasad, N., Basavaiah, K., Vinay, K.B. (**2010**). Sensitive spectrophotometric determination of lamotrigine in bulk drug and pharmaceutical formulations using bromocresol green. *Ecl. Quim.,* 35(1), 55.

20. Rajendra Prasad, N., Basavaiah, K., Vinay, K.B. (**2010**). Micro and nanogram determination of lamotrigine in pharmaceuticals by visible spectrophotometry using bromophenol blue. *Indian J. Chem. Tech.,* 17(5), 220.
21. Alizadeh, N., Khakinahad, R., Jabbari, A. (**2008**). Spectrophotometric determination of lamotrigine in pharmaceutical preparations and urine by charge-transfer complexation. *Pharmazie.,* 63(11), 791.
22. Talekar, R.S., Dhake, A.S., Sonaje, D.B., Mourya, V.K. (**2000**). Spectrophotometric determination of lamotrigine. *Indian J. Pharm. Sci.,* 62(1), 51.
23. Youssef ,N.F., Taha, E.A. (**2007**). Development and validation of spectrophotometric, TLC and HPLC methods for the determination of lamotrigine in presence of its impurity. *Chem. Pharm. Bull.,* 55(4), 541.
24. Elizabeth, G-S., Giannoutsos, S., Lower, D.R., Virji, M.A., Krasowski, M.D. (**2007**). Drug monitoring: simultaneous analysis of lamotrigine, oxcarbazepine, 10-hydroxycarbazepine, and zonisamide by HPLC-UV and a rapid GC method using a nitrogen-phosphorus detector for levetiracetam. *J. Chromatogr. Sci.,* 45(9), 616.
25. Anantha Kumar, D., Venkata Kumar, C., Seetharamaiah, P., Sheshagiri Rao, J.V.L.N. (**2010**). Estimation of lamotrigine by RP-HPLC method. *J. Chem.,* 7(S1), S203.
26. Vinay, K. B., Revanasiddappa, H. D., Cijo1, M. X., Ramesh, J., Swamy, N., Rajendraprasad, N. (**2015**). Stability indicating ultra performance liquid chromatographic assay of lamotrigine in pharmaceuticals. *Trakia J. Sci.,* 13(1), 1.
27. Olga, D.R., Calvo, M.E., Acros-Martinez, M.J. (**2008**). Determination of lamotrigine in pharmaceutical preparations by adsorptive stripping voltammetry using screen printed electrodes. *Sensors*, 8(12), 4201.
28. Gupta, V. K., Singh, A. K., Gupta, B. (**2007**). Development of membrane electrodes for selective determination of some antiepileptic drugs in pharmaceuticals, plasma and urine. *Anal. Bioanal. Chem.,* 389, 2019.
29. Hayedeh B.S., Soltan A. E., Atefeh T., Fatemeh B., Hadi A., Nasim A., Somayeh M. (**2011**). Potentiometric Sensing of Lamotrigine Based on Molecularly Imprinted Polymers. Electroanal. 23(11), 2716 .
30. Ensafi, A., Allafchian, A. R. (**2008**). Potentiometric sensor for betahistine determination in pharmaceuticals, urine and blood serum. *J. Pharm. Biomed. Anal.,* 47(4-5), 802.
31. Kanberoglu, G. S., Coldur, F., Topcu, C., Cubuk, O. (**2015**). PVC-Membrane Potentiometric Sensor for the Determination of Tamoxifen in Pharmaceutical Formulations. *IEEE Sensors J.,* 15(11), 6199.
32. AlRabiah, H., Al-Majed, A., Abounassif, M., Mostaf, G.A.E. (**2016**). Two novel potentiometric sensors for determination of clonidine in some pharmaceutical formulation. *Int. J. Electrochem. Sci.,* 11, 6761.
33. Ganjali, M.R., Karimi, S., Shahtaheri, S.J., Norouzi, (**2013**). Determination of clonidine by potentiometry using PVC membrane electrode. *Int. J. Electrochem. Sci.,* 8, 1999.
34. Del Valle, E.M. (**2004**). Cyclodextrins and their uses: a review. *Process Biochem.,* 39, 1033.
35. Hedges, A.R. (**1998**). Industrial Applications of Cyclodextrins. *Chem. Rev.,* 98, 2035.
36. Yang, R., Li, K.A., Wang, K., Zhao, F., Li, N., Liu, F. (**2003**). Porphyrin assembly on β-cyclodextrin for selective sensing and detection of a zinc ion based on the dual emission fluorescence ratio. *Anal. Chem.,* 75, 612.
37. El-Kosasy, A.M., Nebsen, M., El-Rahman, M.K.A., Salem, M.Y., El-Bardicy, M.G. (**2011**). Comparative study of 2-hydroxy propyl beta cyclodextrin and calixarene as ionophores in potentiometric ion-selective electrodes for neostigmine bromide. *Talanta,* 85, 913.
38. Trojanowicz, M. (**2014**). Enantioselective electrochemical sensors and biosensors: A mini-review. *Electrochem. Comm.,* 38, 47.
39. Patil, S.R., Turmine, M., Peyre, V., Durand, G., Pucci, B. (**2007**). Study of β-cyclodextrin/fluorinated trimethyl ammonium bromide surfactant inclusion complex by fluorinated surfactant ion selective electrode. *Talanta,* 74, 72.

40. El-Kosasy, A.M. (**2003**). Determination of hydroxyurea in capsules and biological fluids by ion-selective potentiometry and fluorimetry. *J. AOAC Intl.,* 86, 15.
41. Ozoemena, K.I., Stefan, R-I. (**2005**). Enantioselective potentiometric electrodes based on alpha-, beta-, and gamma-cyclodextrins as chiral selectors for the assay of l-proline. *Talanta,* 66, 501.
42. IUPAC Analytical Chemistry Division, Recommendation for Nomenclature of Ion Selective Electrode. *Pure Appl. Chem.,* **1994**, 66, 2527.
43. IUPAC Analytical Chemistry Division, Potentiometric selectivity coefficients of ion selective electrodes. Pure Appl. Chem., **2000**, 72, 1851.