

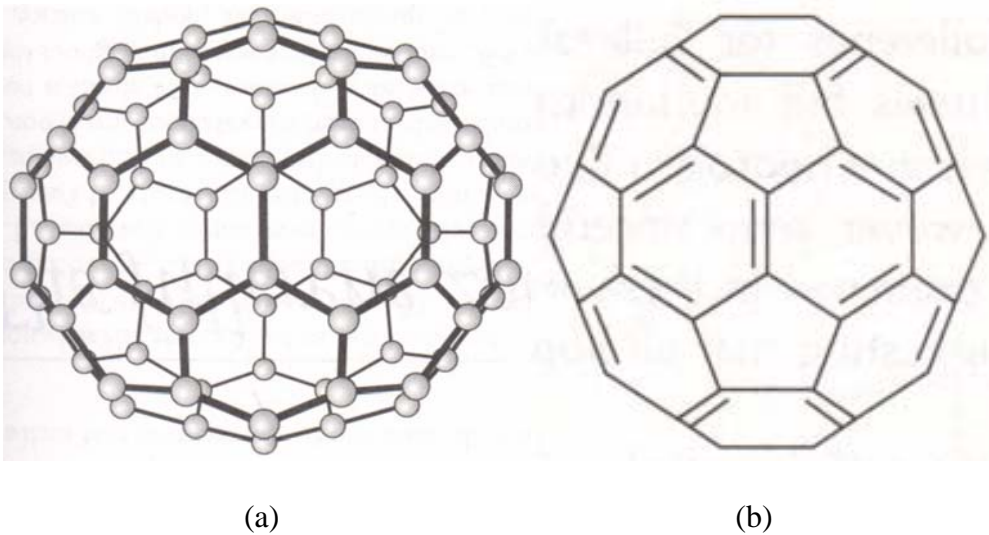
## Chapter 4

# Enantioselective, potentiometric membrane electrodes based on fullerenes

### 4.1 Fullerenes as chiral selector in the EPMEs design

A long search for molecular allotropic forms of carbon other than graphite and diamond culminated in 1985 in the discovery of a  $C_{60}$  molecule. Fullerenes (Figure 4.1) are “cage” molecules that are named after eccentric architect R. Buckminster Fuller, the inventor of the truncated icosahedron-shaped geodesic dome. The soccer-ball shaped  $C_{60}$  molecule and the rugby-ball shaped  $C_{70}$  were soon followed by higher-order fullerenes of 76, 84, 90, and 94 carbon “cages” as large as  $C_{240}$  and  $C_{540}$ .

The fullerenes appear as a whole system able to form a completely new type of chemistry with many surprising arrangements and applications, including analytical ones and it is also said that, when a new atom is put in a fullerene cage, or when an atom (or group) is attached to the outside, a new molecule is formed [1,2].



**Figure 4.1** Fullerenes (a) C<sub>70</sub> and (b) C<sub>60</sub>

Since the discovery of the fullerenes (C<sub>60</sub>, C<sub>70</sub> and higher fullerenes), electrochemistry has played a significant part in investigations of the properties of these fascinating new types of carbon. Initially, some important physiochemical properties, including the standard redox potentials, and confirming theoretical predictions of these molecules [3,4] were considered. Fullerenes (C<sub>60</sub> and C<sub>70</sub>) display anomalous behaviour in solution due to the formation of aggregates [5-13]. Since the specific surface energies of interactions of fullerene molecules are very close in magnitude [5], fullerenes in solution often tend to form aggregates.

## **4.2 Enantioanalysis of S-deprenyl using enantioselective, potentiometric membrane electrodes based on C<sub>60</sub> fullerenes**

Three enantioselective, potentiometric membrane electrodes based on C<sub>60</sub> fullerene derivatives impregnated on carbon-paste matrices as chiral selector are proposed for the enantioanalysis of S-deprenyl (dep) [14].

### **4.2.1 Reagents and materials**

(1,2-methanofullerene C<sub>60</sub>)-61-carboxylic acid (I), diethyl (1,2-methanofullerene C<sub>60</sub>)-61-61-dicarboxylate (II) and tert-butyl (1,2-methanofullerene C<sub>60</sub>)-61-carboxylic acid (III) were supplied by Fluka (Buchs, Switzerland). Phosphate buffer of pH 5.82 was supplied by Merck (Darmstadt). Deionized water from a Modulab system (Continental Water Systems, San Antonio, TX, USA) was used for all solutions preparations. The S- and R-dep solutions necessarily in the characterization of the enantioselective potentiometric membrane electrodes were prepared from standard S- and R-dep solutions (10<sup>-2</sup> mol/L), respectively, by serial dilutions. All standard and diluted solutions were buffered with phosphate buffer (pH 5.82 mol/L) from Merck (Darmstadt, Germany) (1:1,v/v, buffer: deionised water). Lentogestic tablets (65 mg deprenyl per tablet) were obtained from Adcoc Ingram Limited (Johannesburg, South Africa).

## 4.2.2 Apparatus

A 663 VA Stand (Metrohm, Herisau, Switzerland) combined with a PGSTAT 20 and a software (Eco Chemie version 4.4) were used for all potentiometric measurements. A glassy carbon electrode and a Ag/AgCl (0.1 mol/L KCl) served as the counter and reference electrodes in the cell.

## 4.2.3 Electrodes design

Paraffin oil and graphite powder were mixed in a ratio of 1:4 (w/w) followed by the addition of chiral selector (fullerene (I), (II) or (III)) to carbon paste. A certain quantity of carbon paste free of chiral selector was prepared and it was placed into a plastic pipette peak leaving 3–4mm empty in the top to be filled with the carbon paste that contains the chiral selector. The diameter of the potentiometric, enantioselective membrane electrode was 3 mm. Electric contact was obtained by inserting a Ag/AgCl wire in the carbon paste. 0.1mol/L of KCl was used as internal solution. The surface of the electrodes was wetted with deionised water and polished with alumina paper (polishing strips 30144-001, Orion) before using them for each experiment. When it was not in use, the electrode was immersed in a  $10^{-3}$  mol/L S-dep solution.

## 4.2.4 Recommended procedure: Direct potentiometry

The potentiometric method was used for the potential determination of each standard solution ( $10^{-10}$ -  $10^{-3}$  mol/L, pH 5.82). The electrodes were placed in the stirred standard

solutions and graphs of E (mV) versus pS-dep were plotted. The unknown concentrations of S-dep were determined from the calibration plots.

#### 4.2.4.1 Content uniform assay of Lentogesic tablets

Each of the ten tablets were placed into 100 ml calibrated flask, dissolved and diluted to the mark using a phosphate buffer (pH 5.8) : deionized water 1:1. The unknown concentration of S-deprenyl was determined using the direct potentiometric method.

### 4.2.5 Results and discussion

#### 4.2.5.1 Response characteristics of the electrodes

The response characteristics exhibited by the proposed electrodes towards S-dep are summarized in Table 4.1. For all the calibration plots, the potentiometric membrane electrodes showed linear near Nernstian responses for S-dep, with correlation coefficients for the equations of calibration of 0.9998 (I), 0.9996 (II) and 0.9999 (III), respectively. R-dep on the other hand, showed non-Nernstian response.

**Table 4.1.** Response characteristics of the potentiometric, enantioselective membrane electrode based on C<sub>60</sub> and its derivative.

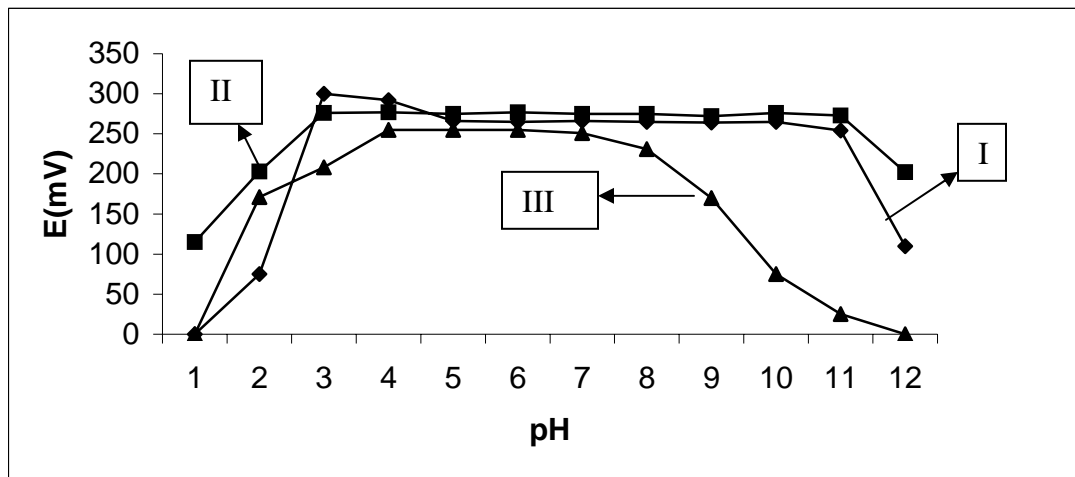
Chiral Selectors	Slope [mV/S-Dep]	Intercept, E° [mV]	Linear range [mol/L]	Detection limit [mol/L]
(I)	55.30	559.00	10 <sup>-10</sup> -10 <sup>-4</sup>	7.6 × 10 <sup>-11</sup>
(II)	56.30	560.40	10 <sup>-9</sup> -10 <sup>-3</sup>	1.12 × 10 <sup>-10</sup>
(III)	58.30	595.20	10 <sup>-10</sup> -10 <sup>-3</sup>	6.1 × 10 <sup>-11</sup>

All measurements were made at room temperature; all values are the average of ten determinations.

The response time was 1min for the fullerene (I) based electrode in concentration range  $10^{-10}$ - $10^{-9}$  mol/L and less than 30s between  $10^{-8}$  and  $10^{-5}$  mol/L. The response time for fullerene (II) based electrode was 2 min between  $10^{-9}$  and  $10^{-5}$  mol/L and 30s between  $10^{-4}$  and  $10^{-3}$  mol/L. The response times for fullerene (III) based electrode in the concentration range  $10^{-9}$ - $10^{-8}$  mol/L was higher 1.5 min and 20s in the concentration range  $10^{-7}$ - $10^{-5}$  mol/L. All electrodes displayed good stability and reproducibility over the test period (6 months) (RSD<0.1%) when used every day for measurements.

#### 4.2.5.2 Effect of pH on the response of the electrodes

The influence of pH on the response of the EPME was checked by recording the emf of the cell for solutions containing  $10^{-5}$  mol/L S-dep at different pH values (pH, 1-12).



**Figure. 4.2.** The influence of pH on the response of the enantioselective potentiometric membrane electrodes (S-dep =  $10^{-6}$  mol/L); for fullerenes I, II, and III based enantioselective, potentiometric membrane electrodes.

These solutions were prepared by adding very small volume of HCl/ NaOH solution (0.1 mol/L or 1 mol/L of each) to S-dep solution. The plots of E (mV) versus pH (Figure 4.2) indicate that the response of the electrodes does not depend upon the pH in the following range: 5.0-10.0 (I) 3.0-11.0 (II) and 4.0-7.0 (III).

### 4.2.5.3 Selectivity of the electrodes

The selectivity of the potentiometric membrane electrode was checked using the mixed solutions method. The concentrations of interfering ions and S-dep were  $10^{-4}$  and  $10^{-5}$  mol/L, respectively. The values obtained for the potentiometric selectivity coefficients for R-dep, PVP, creatine and creatinine demonstrated the enantioselectivity and selectivity properties of the proposed EPMEs for the assay of S-dep (Table 4.2).

**Table 4.2 Potentiometric selectivity coefficients for the enantioselective, potentiometric membrane electrode for S-dep.**

EPMEs	Interfering species					
	R-dep	PVP	Creatine	Creatinine	Paracetamol	L-glutamine
(I)	$5.1 \times 10^{-3}$	$4.0 \times 10^{-3}$	$\ll \times 10^{-4}$	$1.8 \times 10^{-3}$	$4.3 \times 10^{-4}$	$6.5 \times 10^{-3}$
(II)	$\ll \times 10^{-4}$	$2.3 \times 10^{-3}$	$3.3 \times 10^{-3}$	$6.3 \times 10^{-3}$	$3.9 \times 10^{-3}$	$1.4 \times 10^{-3}$
(III)	$8.2 \times 10^{-4}$	$1.3 \times 10^{-3}$	$2.0 \times 10^{-3}$	$1.2 \times 10^{-3}$	$2.5 \times 10^{-3}$	$7.9 \times 10^{-3}$

All measurements were made at room temperature; all values are the average of ten determinations.

### 4.2.5.4 Analytical applications

To assess the feasibility of the proposed direct potentiometry method for enantioanalysis of S-deprenyl, recovery tests were performed for S-deprenyl in the presence of its

antipode. The assay of S-dep in the presence of R-dep was conducted by using different ratios between S-dep and R-dep. The results obtained (Table 4.3) demonstrated the suitability for the proposed enantioselective, potentiometric membrane electrodes for testing the enantiopurity of deprenyl tablets due to the good recovery values obtained for the assay of one of the enantiomers in the presence of its antipode. No significant differences in the recovery values were recorded for the ratios between S:R enantiomers varying from 1:9 to 1:99.9.

**Table 4.3** Determination of S-deprenyl in the presence of R-deprenyl.

Sample	S-Dep, Recovery (% of nominal value)		
	[I]	[II]	[III]
2:1	99.60 ± 0.02	100.00 ± 0.01	99.99 ± 0.02
1:1	99.59 ± 0.02	99.05 ± 0.02	99.98 ± 0.02
1:2	99.62 ± 0.01	99.32 ± 0.02	99.98 ± 0.01
1:4	99.98 ± 0.01	100.00 ± 0.02	100.00 ± 0.01
1:9	99.45 ± 0.02	99.99 ± 0.01	99.99 ± 0.02

All measurements were made at room temperature; all values are average of ten measurements.

The proposed enantioselective, potentiometric membrane electrodes proved to be useful for the uniformity content test of Lentogesic tablets. The results obtained for content uniformity test of Lentogesic tablets using the proposed electrode showed, that the tested pharmaceutical formulations contain 98.12±0.14, 98.13±0.15, and 98.20±0.17% S-deprenyl, when electrodes based on fullerenes I, II, and III, respectively were used.



### 4.3 Conclusions

This chapter describes new enantioselective, potentiometric membrane electrodes designed using (I), (II) and (III) as chiral selectors for the enantioanalysis of deprenyl. The electrodes can be successfully used for the assay of S-deprenyl in the presence of R-deprenyl. The enantioselectivity is good for all the three proposed electrodes; the best being recorded when tert-butyl (1,2-methanofullerene C<sub>60</sub>)-61-carboxylic acid is used as chiral selector. The proposed EPME based on fullerene derivatives have good feature in enantioselective analysis. The simple, fast and reproducible construction of the electrode assures the reliable response characteristics. The good enantioselectivity of designed electrodes is allowed to perform the enantiopurity assay of S-deprenyl raw material and in its pharmaceutical formulation Lentogesic.

## 4.4 References

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