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DIFFERENTIAL PULSE POLAROGRAPHIC DETERMINATION OF SOME 6-ACYL-2 (3H)-BENZOXAZOLONE DERIVATIVES

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SUMMARY

The synthesis and the differential pulse polarographic analysis of ten 6-acyl-2(3)-benzoxazolone derivatives are presented. pH 3.00 Britton-Robinson buffer was found to be the best supporting electrolyte. The electrochemical determination is based on the reduction of carbonyl group present in all molecules. Linear response was observed from 0.2-2 to 25-65 $mg.L^{-1}$ depending on the molecules.

Key Words: Benzoxazolone derivatives, synthesis, analysis, differential pulse polarography.

INTRODUCTION

Benzoxazolinones have been associated with various type of biological properties. Lespagnol and his co-workers (1,2) prepared and tested a number of derivatives of benzoxazolin-2-ones for their analgesic (3,4), antipyretic (3,4), anticonvulsive and hypnotic effects (1). Analgesic and antipyretic activity have been described in 6-acyl-benzoxazolin-2-ones (4,5) and in 3-aminoalkylbenzoxazolin-2-ones (6,7). Antibacterial (8,9) and fungicidal (10) activities have also been reported for benzoxazolin-2-ones. The pronounced biological activity of many 2-benzoxazolinone derivatives (4) and medicinal value of 5-chloro-2-benzoxazolinone (11) prompted the investigation of 3-substituted-2-benzoxazolinones.

In this work the preparation of ten different 6-acyl-2(3H)-benzoxazone derivatives have been given. Due the lack in the literature concerning the quantitative determination of these benzoxazolones, an accurate and rapid electrochemical method is proposed for the analysis of these molecules.

EXPERIMENTAL

Reagents

All chemicals used were of analytical grade purity and were used without any further purification. Britton-Robinson (BR) buffer was prepared by mixing 2.7 ml phosphoric acid, 2.3 ml glacial acetic acid and 2.47 g boric acid and then filled up to 1.0 liter with triple distilled water. The BR buffers with different pH values were prepared by adding appropriate volumes of 0.2 N NaOH to above acid mixture.

Apparatus

Polarographic analysis were performed with a Princeton Applied Research (PAR) polarographic analyzer, model 174A with PAR model 174/70 drop timer assembly. Polarograms were recorded on a Houston omnigraphic 2000 X-Y recorder. pH measurements were made with a pH meter Corning model 12. Melting points were determined on a Buchi SMP-20 apparatus. Infrared spectra were taken by a Beckman Acculab model 4 Infracord Spectrophotometer in KBr disc. NMR spectra were taken on a Varian model C-60 HL Jeol Spectrometer using TMS as the internal standard. Microanalyses were performed by "Le Centre de Microanalyse du C.N.R.S. de Villeurborne, France.

Procedures

5 mg of the molecules analyzed were dissolved in 5 ml methanol and a small volume (on the order of 10 µl) was added into a supporting

electrolyte by micropipet. Before each measuremenets, oxygen was removed from supporting electrolyte by sparging with nitrogen. During the measurement nitrogen was passed over the surface of the solution. The electrode assembly used in the course of the present work and other details of experimental conditions has been described priviously (12).

Synthesis

The methods of synthesis of 6-acylbenzoxazolin-2-ones and 3-piperazinoalkyl-6-acylbenzoxazolin-2-ones:

Method A: 6-Acylbenzoxazolin-2-ones

0.1 Mole benzoxazolin-2-one derivative and 200 g polyphosphoric acid were mixed in a two necked round-bottomed flask. On to this mixture, 0.1 mole of acid derivatives used for acylation were slowly added by constant strirring. Then they were put in a thermostatic oil bath at 150^{o}C and the mixing was continued untill the dark brown color was obtained. Later they were poured into 900 ml of ice-water mixture and stirred seven hours more. The precipitates obtained was washed with water, dried and recrystallized from appropriate solvent.

Method B: 3-piperazinoalkyl-6-acylbenzoxazolin-2-ones

a) 3-piperazinomethyl-6-acylbenzoxazolin-2-ones

0.1 mole benzoxazolin-2-one derivative, 30 ml of methanolic solution of 0.01 mole piperazine derivative and 1 ml (0.013 mole) 37% formaldehyde solution were mixed. The mixture was heated half an hour on a steam bath and the precipitate obtained was recrystalized form suitable solvent.

b) 3-piperazininoethyl-and 3-piperazinopropyl-6-acybenzoxazolin-2-ones;

To suspension of sodium 6-acylbenzoxazolin-2-one (0.01 mole) in 50 ml of diemethylformamide, appropriate 1-(chloroalkyl)-4-arylpiperazine (0.01 mole) was added and refluxed for an hour. The reaction mixture was cooled and DMF was remowed under vacuum. The product thus separeted was recrystallized from suitable solvent. All products were obtained in 60-80 % yield (Table 1 and 2).

Table 1. 6-acyl-and 3-piperazinoalkyl-acylbenzoxazolin-2-ones

Group	comp.	R_1	R_2	R_3	m.p. (°C)	IR (cm^{-1})
I	1	Cl(o)	Cl	H	199	1675
	2	Cl(m)	Cl	H	214	1675
II	3	H	Cl	H	185	1675
	4	F(p)	H	H	228	1675
	5	H	Cl	$CH_2N\square NC_6H_4F(p)$	149	1650
III	6	F(p)	Cl	H	235	1685
IV	7	$OCH_3(p)$	H	$CH_2N\square NC_6H_4OCH_3(o)$	179	1645
	8	F(p)	H	$(CH_2)_2N\square NC_6H_4OCH_3(o)$	136	1655
	9	$OCH_3(p)$	H	$(CH_2)_2N\square NC_6H_4CF_3(m)$	114	1650
	10	$OCH_3(p)$	H	$(CH_2)_3N\square NC_6H_4OCH_3(m)$	118	1650

RESULTS AND DISCUSSION

Polarography is becoming increasingly important in the analysis of compounds of pharmaceutical interest. This is due to the fact that pharmaceutical preparations are in most cases the mixture of well-defined molecules of approximately known composition. Differential pulse polarography has been found to be the best polarographic method (13). No previous data is available regarding the quantitative analysis of the molecules studied.

Table 2. The results of elemental analysis of benzoxazolones studied

Comp.	Formula		%C	%H	%Cl	%F	%N
1	$C_{14}H_7Cl_2NO_3$	Calc.	54.57	2.29	23.01	-	4.55
		Foun.	54.85	2.34	23.18	-	4.49
2	$C_{14}H_7Cl_2NO_3$	Cal.	54.57	2.29	23.01	-	4.55
		Fo.	54.96	2.42	22.82	-	4.39
3	$C_{14}H_8ClNO_3$	C.	61.44	2.95	12.95	-	5.12
		F.	61.33	3.02	12.96	-	5.12
4	$C_{14}H_8FNO_3$	C.	65.37	3.14	-	7.39	5.45
		F.	65.29	3.26	-	7.48	5.45
5	$C_{25}H_{21}ClFN_3O_3$	C.	64.05	4.69	7.48	4.08	9.15
		F.	64.45	4.54	7.61	4.08	9.02
6	$C_{14}H_7ClFNO_3$	C.	57.65	2.42	12.15	6.51	4.80
		F.	57.30	2.33	12.10	6.30	4.73
7	$C_{27}H_{27}N_3O_5$	C.	68.48	5.70	-	-	8.87
		F.	68.09	5.65	-	-	8.58
8	$C_{27}H_{26}FN_3O_4$	C.	63.34	5.32	6.92	3.71	8.21
		F.	63.34	5.02	6.45	3.57	8.30
9	$C_{28}H_{26}F_3N_3O_4$	C.	64.00	4.98	-	10.85	7.99
		F.	63.84	4.99	-	11.02	7.74
10	$C_{29}H_{31}N_3O_5$	C.	69.44	6.23	-	-	8.38
		F.	69.50	6.21	-	-	8.23

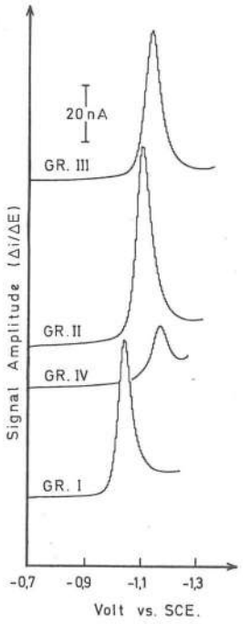

Figure 1. Differential pulse polarograms obtained at pH 3.00 of 10 mg/L of various benzoxazolones

Analytical application of differential pulse polarography to ten different benzoxazone derivatives gives rise to a single, well-defined, diffusion controlled and reversible peak in aqueous solutions as shown in Figure 1. Over the range 20-50 ^{o}C, the graphs of log (Δ i) versus temperature suggest a linear relationship with a temperature coefficient of 0.87 % $^{o}C^{-1}$. Also, a linear relationship was determined for $(\Delta i)_{max}$ versus the square root of the height of the mercury column. All these data indicate the electroreduction of benzoxazolones being diffusion controlled. The reversibility of the electrode process was investigated by performing cathodic and anodic scan. Later anodic andcathodic currents and potential values were compared. These results together with logarithmic analysis show us the reversible nature of the process.

The effect of pH on peak potential which was shown in Table 3 confirms the presence of four groups of compound. We have divided all ten molecules to four groups according to the substituents (Table 1).

The linear responce was found to be the greatest in BR buffer at pH 3.00 which was taken as supporting electrolyte. It was clear that there was a correlation between the structure of the benzoxazolones and the peak potential values in a selected supporting electrolyte. The separation of the molecules into four groups was based on the fact that the molecules having at least 30 mV different peak potential velues.

The molecules show polarographic activity due to carbonyl groups present. The carbonyl group adjacent to etheric linkage cannot be reduced easily. The reduction of the other carbonyl group was investigated by controlled potential electrolysis and involves the addition of two electron and two proton to form corresponding hydroxide.

Table 3. The effect of pH on the peak potential (V.vs.SCE) of various benzoxazones shown in Table 1 (c=10 mg/L)

Comp.		0.1N HCl	0.1N NaOH	pH 3 BR	pH 12 BR	1M KNO_3
1	I	-0.93	-1.62	-1.05	-1.60	-1.04
2		-0.93	-1.53	-1.05	-1.54	-1.03
3		-1.95	-1.63	-1.11	-1.60	-1.08
4	II	-0.96	-1.75	-1.10	-1.70	-1.10
5		-0.97	-1.64	-1.12	-1.65	-1.10
6	III	-0.98	-1.65	-1.15	-1.64	-1.10
7		-1.00	-1.75	-1.18	-1.75	-1.15
8	IV	-1.00	-1.64	-1.20	-1.65	-1.12
9		-1.00	-1.75	-1.19	-1.75	-1.12
10		-1.02	-1.70	-1.18	-1.72	-1.16

in acidic medium. This mechanizm can be easily seen by IR spectra. In Table 1 the last column shows us carbonyl streching bands which cannot be seen after electroreduction. Instead of carbonyl streching bands, hydroxyl bands appear in IR spectra of electroreduced benzoxazolones. 2-electron reduction of the carbonyl group of 2-methylamino-

5-chlorobenzophene in acidic medium shown by Oelschlager et al (14).

Linear calibration curves for each groups of benzoxazolones were obtained in pH 3.00 BR buffer. The dependence of the cathodic current on benzoxazolones concentration is shown in Figure 2. Table 4 shows the statistical analysis of the benzoxazolones studied.

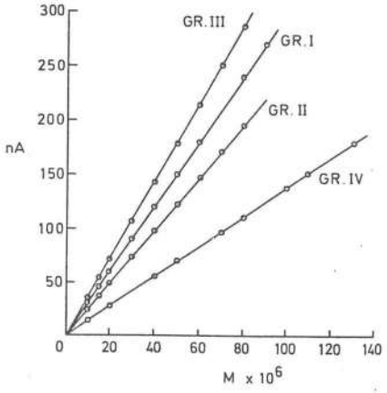

Figure 2. The linear calibration curves obtained by differential pulse polarography for benzoxazolones

Table 4. Characteristics of linear regression of calibration graphs for benzoxazolones

GC	LL	UL	N	CC	SL	CI
I	0.99	99.00	10	0.986	2.98	1.72
II	0.99	90.90	10	0.994	2.43	0.82
III	0.99	43.06	10	0.996	3.57	0.65
IV	1.99	130.43	10	0.999	1.37	-0.30

(GC) Group of Compounds, (LL) Lower Limit of detection ($Mx10^6$), (UL) Upper Limit of Detection ($Mx10^6$), (N) Number of data points, (CC) Correlation Coefficient, (SL) Slope, (CI) Concentration Intercept.

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