ELECTROCHEMICAL INVESTIGATION OF CERTAIN NOVEL HYDRAZIDES BEARING PYRAZOLINE-5-ONE AND INDOLE MOIETIES

DASTAGIRI REDDY MALIKI REDDY[1], RAGHAVENDRA GURU PRASAD ALURU[2], SPOORTHY YADATI NARASIMHA[3], RAVINDRANATH LAKSHMANA RAO KRISHNA RAO[4]

The electrochemical behaviour of [3-methyl-5-oxo-4-(4'-substituted aryl hydrazono)-4,5-dihydro-pyrazol-1-yl]-aceticacid-(2-oxo-1-piperidine-1-yl-methyl-1,2-dihydro–indol-3-yl-idene)-hydrazides (a-f) was studied in Britton-Robinson buffer solutions containing aqueous dimethylformamide 40% (v/v) using DC polarography and cyclic voltammetry. The kinetic parameters such as charge transfer coefficient, heterogeneous rate constant and activation free energy change were evaluated. Based on the experimental data, the electrochemical process was reported to be diffusion controlled and irreversible. The results from polarography and cyclic voltammetry (at hanging mercury drop electrode-HMDE and modified carbon pasted electrode-MCPE) were compared and a mechanism for the electrode reduction was proposed in acidic and basic media.

Keywords: polarography; cyclic voltammetry; HMDE; MCPE; mechanism of electrochemical reaction.

1. Introduction

Medicinal chemists exploit heterocyclic structures as the scaffold to design new drugs, as they demonstrate distinctive ability to mimic the structure of peptides and to bind reversibly to proteins [1–2]. Numerous pharmacologically active compounds such as Vincristine (Anticancer), Delavirdine (Anti-HIV), Arbidol (Antiviral), Indomethacin (Anti-inflammatory) etc have an indole nucleus. The diverse pharmacological activities of indole system are evident from the literature [3-8]. On the other hand, pyrazolon-5-ones also has versatile medicinal applications and exhibit anti microbial [9,10], anti cancer [11], analgesic [12], anti covasculant [13], anti viral [14] and anti inflammatory [15] activities. Many such frameworks, along with their medicinal activity are reported by the authors [9,10].

[1] Sri Krishnadevaraya University, Anantapur, A.P., India
[2] The ICFAI Foundation for Higher Education, Hyderabad, A.P., India. e mail: guruar@rediffmail.com
[3] Sri Krishnadevaraya University, Anantapur, A.P., India
[4] Sri Krishnadevaraya University, Anantapur, A.P., India

Electrochemical techniques are most suitable to investigate the redox properties i.e metabolic information of a new drug. It is a practice to bring about the analogy between the electrochemical reactions taking place at the electrode and biochemical reactions [16]. Many successful efforts have been conducted to understand the pharmacological activity of a drug in terms of *in vivo* redox processes through electrochemical investigations [17, 18].
The versatile medicinal applications of indoles and pyrazolon-5-ones have inspired the authors to undertake the electrochemical studies of certain frameworks containing both these moieties. The authors have reported the electrochemical reduction mechanism of these novel compounds, which reveals the information about the biochemical reactions and their possible metabolites if are used as drugs.

2. Materials and Methods

2.1. Chemicals and instruments

The chemicals employed in the studies were of analytical reagent grade obtained from Merck India Limited. The pH of Britton-Robinson buffer solutions [19] were measured using pH meter, LI–10, Elico Private Limited, Hyderabad, India. Analar mercury was further purified according to the procedure described by Vogel [20] and was vacuum distilled.
A CL–25 Pen Recording Polarograph manufactured by ELICO Private Limited, Hyderabad, India was used to record current voltage curves. The capillary having the characteristics 1.80 $mg^{2/3}$ $s^{-1/2}$ at h = 80 cm was employed in the studies.
The cyclic voltammeter used consists of an X–Y recorder (Model RE 0074), a PAR 175 Potentiostat and a PAR 175 Universal Programmer. A single compartment cell model 303 SMDE supplied by PAR with silver wire as reference electrode and platinum wire as counter electrode was used in the studies. A stationary mercury drop electrode (SMDE 303) with a drop area 0.0096 cm^2 was used as the working electrode. A circulating type thermostat supplied by Thoshniwal, Bombay, India, was employed to maintain a constant temperature with the range of $\pm$ 0.01°C.

2.2. Procedure for polarographic studies

8.0 mL of the buffer solution of desired pH (1.1 – 10.1), 2 mL of the stock solution of the substrate (1.0×10^{-2} M) in dimethylformamide (DMF), 6 mL of dimethylformamide (DMF) and 4.0 mL of distilled water were mixed thoroughly in the polarographic/ cyclic voltammetric cell and the polarograms/ cyclic voltammograms were recorded after removing the dissolved oxygen by passing pure and dry nitrogen gas through the solution for about fifteen minutes.

2.3. Preparation of chemically modified electrode

Chemically modified carbon paste electrodes were prepared by grinding the crown – ether crystals with a mortar and pestle. The mixture was ground with graphite powder, then nujol oil was added and the composition was thoroughly mixed. The final composition of the mixture was 54:36:10% w/w graphite/oil/modifier. The paste was packed in one end of a glass tube (3 mm bore; 1mm wall) to make contact with a copper wire inserted in the tube.

3. Results and Discussion

3.1. Polarographic behaviour of [3-methyl-5-oxo-4-(4'-substituted aryl hydrazono)-4,5-dihydro-pyrazol-1-yl]-acetic acid-(2-oxo-1-piperidine-1-yl-methyl-1,2-dihydro–indol-3-ylidene)-hydrazides (a-f)

Compound	a	b	c	d	e	f
-R	H	4'-CH_3	4'-OCH_3	4'-OC_2H_5	4'-Cl	4'- Br

3.1.1. General polarographic behaviour

The compounds under study 'a-f' were synthesised according to the procedure mentioned in the literature [21].
The compounds gave two well defined cathodic waves in the pH range 1.1-7.1 and three cathodic waves in the pH range 8.1-10.1. The model polarograms are shown in the Fig. 1.

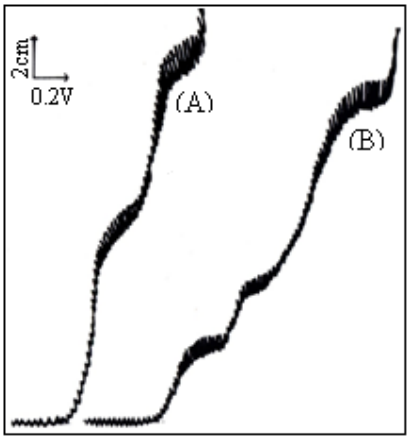


Fig. 1. Polarograms of 'a'; Medium = Dimethyl formamide (40% v/v); (A) pH 4.1, Starting potential 0.3 V; (B) pH 8.1, Starting potential 0.7 V.

An inspection of the structure of above compounds reveals that the sites susceptible for reduction at the dropping mercury electrode are π systems namely exocylic >C=N, exocyclic >C=O, cyclic >C=N and weak N-N sigma bonds. Among these bonds (groups) exocyclic azomethine group is most susceptible for reduction than other groups. The reduction of the exoyclic >C=O and cyclic >C=N generally occurs at higher negative potentials at the dropping mercury electrode. The polarographic behaviour of ([3-methyl-4,5-dioxo-4,5-dihydro-pyrazol-1-yl]-acetic acid hydrazide) under similar experimental conditions reveals that the compound does not exhibit any reduction wave under experimental conditions (pH 1.1-10.1). These observations, therefore, suggest the waves observed were due to reduction of two exocyclic azomethine groups [22].
Half wave potentials of first and second waves were shifted to more negative potentials with pH in acidic pH range 1.1-6.1 (Table 1). The values of $\Delta E_{½}/\Delta pH$ for both the waves lie in the range of 0.089-0.094. The half wave potential of the wave in alkaline solution was not altered with the change in pH of the solutions. The typical $E_{1/2}$–pH graphs shown in Fig. 2 consist of two linear portions intersecting at pH 8.1.

The sigmoid shaped $E_{1/2}$-pH graphs were observed in the pH range 1.1-10.1. These graphs suggest that both the protonated species (acidic) and the unprotonated species (basic) of the depolariser were electroactive. As the half wave potentials were also very close to each other, the polarographic waves of both forms, therefore, merge with each other resulting in a single wave. The limiting currents of compounds a-f recorded at different heights of the mercury column vary linearly with the square root of mercury column height ($h^{½}$) to confirm the diffusion controlled nature [23] of the first and second waves (Fig. 3). This was further confirmed by linear relationship between the limiting current and concentration of hydrazides under study (Fig. 4).

E_{dme} versus log (i/i_d-i) graphs at typical pH value 4.1 are shown in the Fig. 5. The plots were linear and the slopes were in the range 0.066V-0.10V. The αn_a values (α is the transfer coefficient and n_a is the number of electrons involved) were calculated and presented in Table 1. The irreversible nature of the two waves at low pH values was attributed to the bulky group present at the end of >C=N-NH- linkage [24]. The kinetic parameters, i.e. k^0_{fh} (heterogeneous rate constant) and ΔG* (activation free energy change) of the electrode reaction at typical pH values evaluated for first and second waves are presented in the Table1. The decrease in the value of k^0_{fh} and the increase in the value of ΔG* with increase in pH further confirms the irreversible nature of the waves [25].

Table 1

Polarographic characteristics and kinetic parameters of [3-methyl-5-oxo-4-(4'-substituted aryl hydrazono)-4,5-dihydro-pyrazol-1-yl]-acetic acid (2-oxo-1-piperidine-1-yl-methyl-1,2-dihydro–indol-3-ylidene)- hydrazide (1×10^{-3}M); Medium : Aqueous dimethyl formamide (40% v/v)

pH	$\Delta E_{1/2}/\Delta pH$ (Δ/v)		α_{na}			No. of protons p	$D \times 10^{-6}$ cms^{-1}	$I^* \times 10^3$		K^0_h cm s^{-1}			ΔG^* k cal mole^{-1}			$\Delta E_{1/2}/\Delta pH$ (Δ/v)		
	I Wave	II Wave	I Wave	II Wave	III Wave	I Wave	II Wave	I Wave	II Wave	I Wave	II Wave	III Wave	I Wave	II Wave		I Wave	II Wave	I Wave
-H																		
2.1	0.089	0.094	0.42	0.38		0.63	0.60	2.1	0.089	0.094	0.42	0.38		0.63	0.60	2.1	0.089	0.094	0.42
4.1	0.089	0.094	0.42	0.38		0.63	0.60	4.1	0.089	0.094	0.42	0.38		0.63	0.60	4.1	0.089	0.094	0.42
6.1	0.089	0.094	0.40	0.34		0.6	0.52	6.1	0.089	0.094	0.40	0.34		0.6	0.52	6.1	0.089	0.094	0.40
8.1	0.089	0.094	0.36	0.31	0.37	0.54	0.47	8.1	0.089	0.094	0.36	0.31	0.37	0.54	0.47	8.1	0.089	0.094	0.36
10.1	0.089	0.094	0.36	0.31	0.37	0.54	0.47	10.1	0.089	0.094	0.36	0.31	0.37	0.54	0.47	10.1	0.089	0.094	0.36
methyl																			
2.1	0.11	0.090	0.47	0.43		0.87	0.66	2.1	0.11	0.090	0.47	0.43		0.87	0.66	2.1	0.11	0.090	0.47
4.1	0.11	0.090	0.47	0.43		0.87	0.66	4.1	0.11	0.090	0.47	0.43		0.87	0.66	4.1	0.11	0.090	0.47
6.1	0.11	0.090	0.45	0.39		0.84	0.60	6.1	0.11	0.090	0.45	0.39		0.84	0.60	6.1	0.11	0.090	0.45
8.1	0.11	0.090	0.41	0.36	0.40	0.76	0.55	8.1	0.11	0.090	0.41	0.36	0.40	0.76	0.55	8.1	0.11	0.090	0.41
10.1	0.11	0.090	0.41	0.36	0.40	0.76	0.55	10.1	0.11	0.090	0.41	0.36	0.40	0.76	0.55	10.1	0.11	0.090	0.41
methoxy																			
2.1	0.10	0.066	0.53	0.47		0.90	0.52	2.1	0.10	0.066	0.53	0.47		0.90	0.52	2.1	0.10	0.066	0.53
4.1	0.10	0.066	0.53	0.47		0.90	0.52	4.1	0.10	0.066	0.53	0.47		0.90	0.52	4.1	0.10	0.066	0.53
6.1	0.10	0.066	0.51	0.43		0.86	0.48	6.1	0.10	0.066	0.51	0.43		0.86	0.48	6.1	0.10	0.066	0.51
8.1	0.10	0.066	0.47	0.40	0.42	0.80	0.45	8.1	0.10	0.066	0.47	0.40	0.42	0.80	0.45	8.1	0.10	0.066	0.47
10.1	0.10	0.066	0.47	0.40	0.42	0.80	0.45	10.1	0.10	0.066	0.47	0.40	0.42	0.80	0.45	10.1	0.10	0.066	0.47
ethoxy																			
2.1	0.123	0.108	0.50	0.44		1.04	0.80	2.1	0.123	0.108	0.50	0.44		1.04	0.80	2.1	0.123	0.108	0.50
4.1	0.123	0.108	0.50	0.44		1.04	0.80	4.1	0.123	0.108	0.50	0.44		1.04	0.80	4.1	0.123	0.108	0.50
6.1	0.123	0.108	0.48	0.40		1.0	0.73	6.1	0.123	0.108	0.48	0.40		1.0	0.73	6.1	0.123	0.108	0.48
8.1	0.123	0.108	0.46	0.37	0.44	0.96	0.68	8.1	0.123	0.108	0.46	0.37	0.44	0.96	0.68	8.1	0.123	0.108	0.46
10.1	0.123	0.108	0.46	0.37	0.44	0.96	0.68	10.1	0.123	0.108	0.46	0.37	0.44	0.96	0.68	10.1	0.123	0.108	0.46
chloro																			
2.1	0.08	0.09	0.39	0.36		0.53	0.55	2.1	0.08	0.09	0.39	0.36		0.53	0.55	2.1	0.08	0.09	0.39
4.1	0.08	0.09	0.39	0.36		0.53	0.55	4.1	0.08	0.09	0.39	0.36		0.53	0.55	4.1	0.08	0.09	0.39
6.1	0.08	0.09	0.37	0.32		0.5	0.49	6.1	0.08	0.09	0.37	0.32		0.5	0.49	6.1	0.08	0.09	0.37
8.1	0.08	0.09	0.33	0.29	0.34	0.45	0.44	8.1	0.08	0.09	0.33	0.29	0.34	0.45	0.44	8.1	0.08	0.09	0.33
10.1	0.08	0.09	0.33	0.29	0.34	0.45	0.44	10.1	0.08	0.09	0.33	0.29	0.34	0.45	0.44	10.1	0.08	0.09	0.33
bromo																			
2.1	0.36	0.33	0.53	0.55		1.82	1.47	2.1	0.36	0.33	0.53	0.55		1.82	1.47	2.1	0.36	0.33	0.53
4.1	0.36	0.33	0.53	0.55		1.47	1.15	4.1	0.36	0.33	0.53	0.55		1.47	1.15	4.1	0.36	0.33	0.53
6.1	0.32	0.29	0.50	0.49		0.64	0.38	6.1	0.32	0.29	0.50	0.49		0.64	0.38	6.1	0.32	0.29	0.50
8.1	0.30	0.26	0.45	0.44		0.35	0.17	8.1	0.30	0.26	0.45	0.44		0.35	0.17	8.1	0.30	0.26	0.45
10.1	0.30	0.26	0.45	0.44		0.35	0.17	10.1	0.30	0.26	0.45	0.44		0.35	0.17	10.1	0.30	0.26	0.45

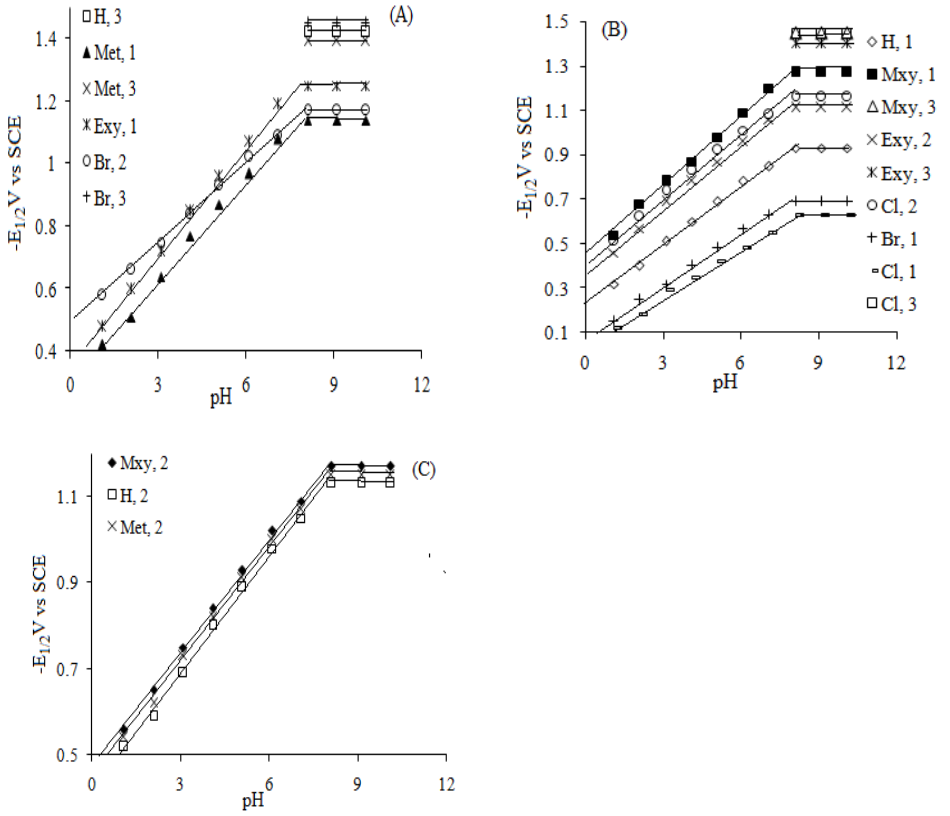


Fig. 2. Effect of pH on half-wave potential; [Hydrazide] = 1×10^{-3}M; Medium = Dimethylformamide (40% by volume). 1 and 2 indicate first and second wave respectively of corresponding compound.

3.1.2. Controlled potential electrolysis

The electrochemical reduction of a has been studied by the method of controlled potential electrolysis at pH 4.1. The controlled potential electrolysis was carried out in a Lingane H-type cell. The cathode compartment contains 10 mL of a (0.01M), 30 mL of DMF, 20 mL of 1.0 M KCl and 40 mL of the buffer solution (pH 4.1). A potential of -1.2 V was applied and was kept constant. The procedure outlined by Lingane [26] was applied to find the number of electrons per molecule and was found to be 8.

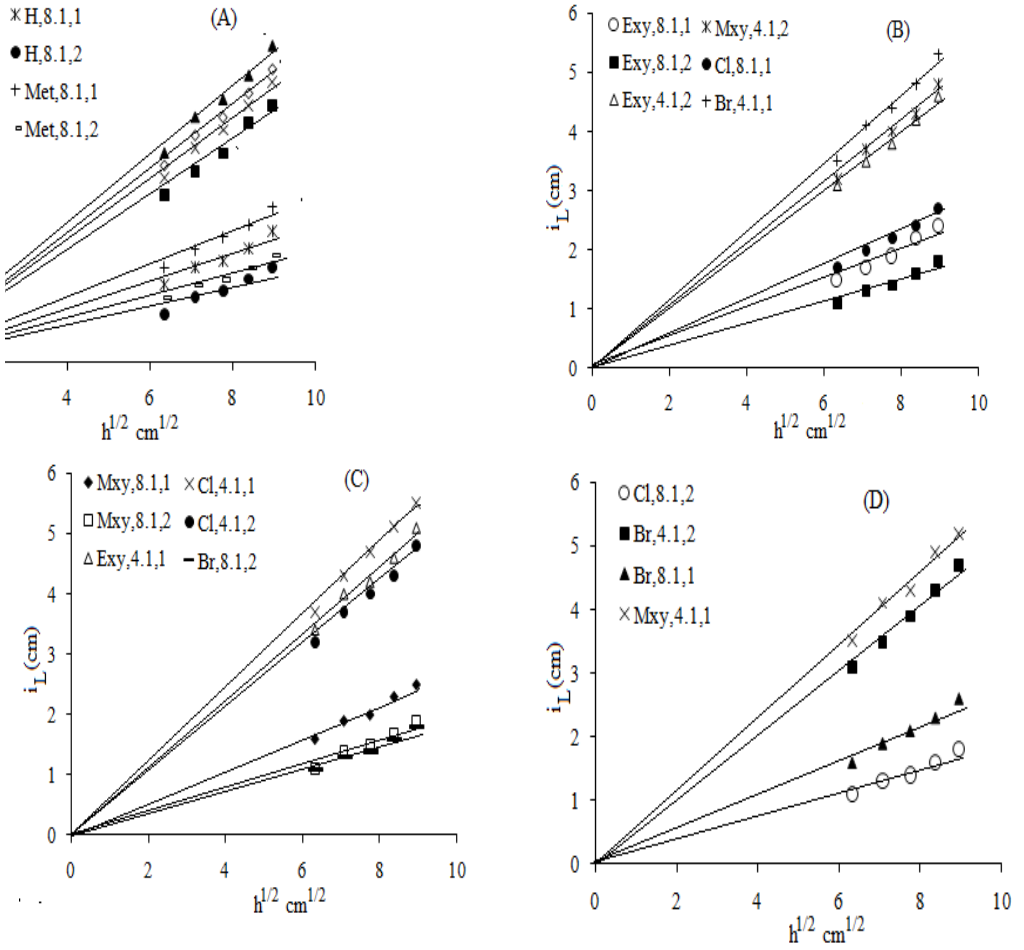


Fig. 3. Effect of mercury column height on limiting current; [Hydrazide] = 1×10^{-3}M; Medium = Dimethylformamide (40% by volume). 1 and 2 indicate the first and second wave, respectively, of the corresponding compound at indicated pH, as shown in figures (A), (B), (C) and (D).

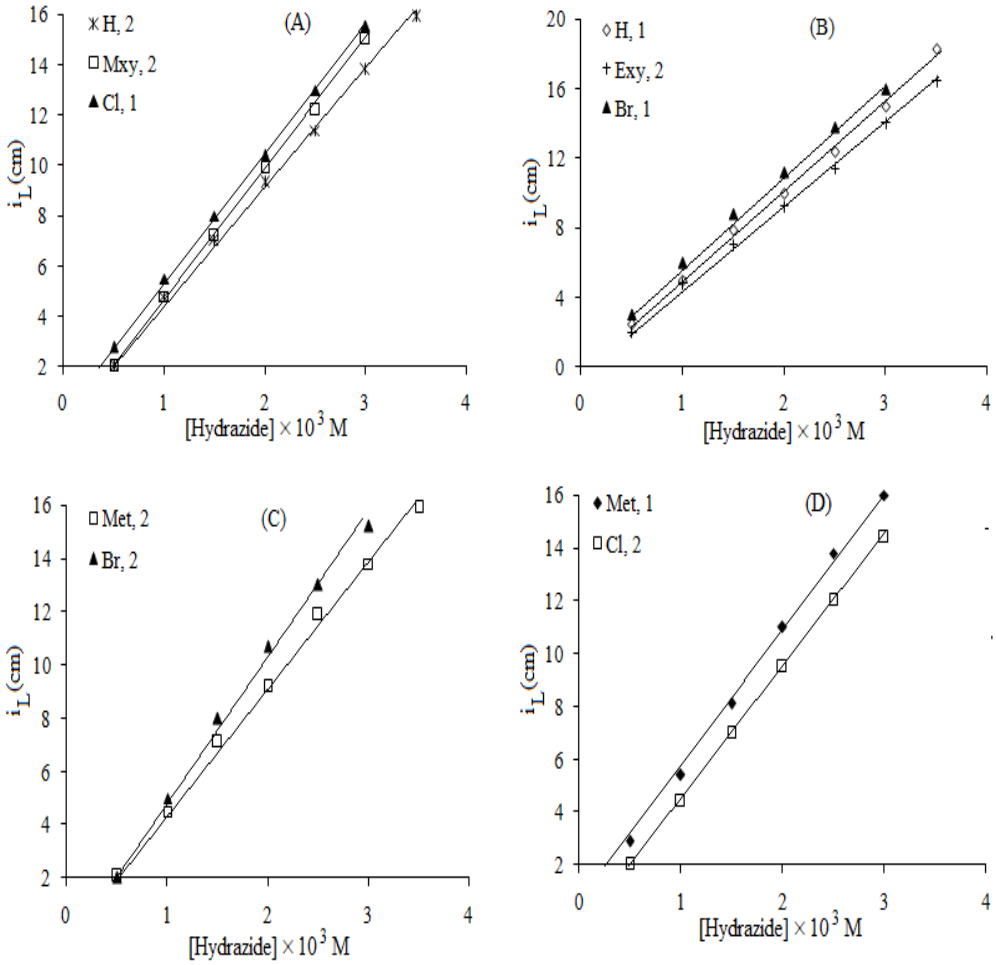
3.1.3. Reduction mechanism in acidic medium

Based on the experimental results, it was proposed that two azomethine groups were reduced separately each involving four electrons. The reduction steps manifest as two waves in acidic solutions of pH 1.1-6.1 through a mechanism which involves the azomethine, imine intermediate and amine via usual sequence.

Hydrazone ⟶ Imine ⟶ Amine

The azomethine group of 'a' was protonated to yield the protonated form II. Weak >C=N-NH single bond undergoes cleavage [27, 28] with the uptake of

four electrons and two protons to form unstable imine intermediates IV & V which further undergo two electron reduction to form VI & VII. It was reported that the



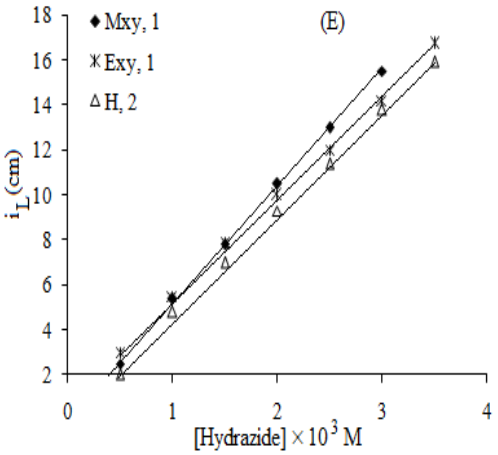
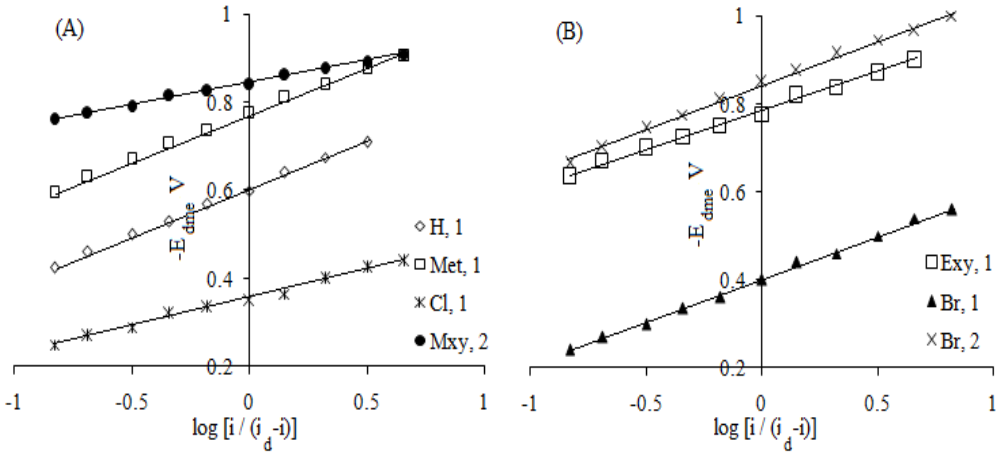


Fig. 4. Effect of concentration on limiting current; pH = 4.1; Medium = Dimethylformamide (40% by volume). 1 and 2 indicate first and second wave respectively of corresponding compound as shown in the figures (A), (B), (C), (D) and (E).



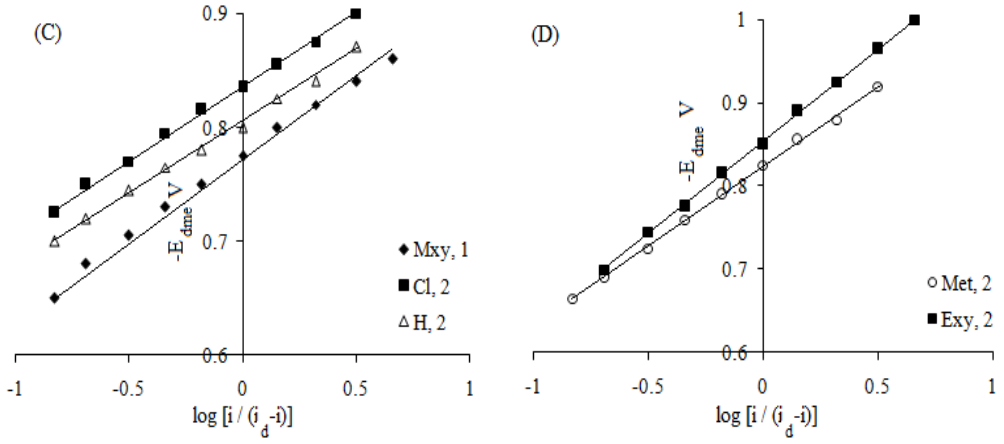


Fig. 5. Semi log polts of hydrazides; [Hydrazide] = 1×10^{-3}M; pH = 4.1; Medium=Dimethylformamide (40% by volume). 1 and 2 indicate first and second wave respectively of corresponding compound as shown in the figures (A), (B), (C) and (D).

above mentioned two steps of reduction occur at the same potential and this fact substantiates the appearance of two single four electron waves as first and second waves.

3.1.4. Reduction mechanism in basic medium

In alkaline medium (pH>pKa) 'a' exist in azomethine anionic form (II) and the later was susceptible to chemical cleavage partially in alkaline solutions to corresponding carbonyl compounds IV and V as shown in the scheme 2. The first and second waves noticed in alkaline solutions were attributed to two 4 electron reductions of azomethine anionic form to amine stage. The third wave was attributed to two 2 electron reductions of heterocyclic carbonyl compounds (IV and V) obtained during the chemical cleavage of dianion II. The heterocyclic carbonyl compounds were reduced to carbinols by two electron reduction process. Decrease in the height of the first and second waves with increase in alkali concentration was attributed to the partial chemical cleavage of dianion.

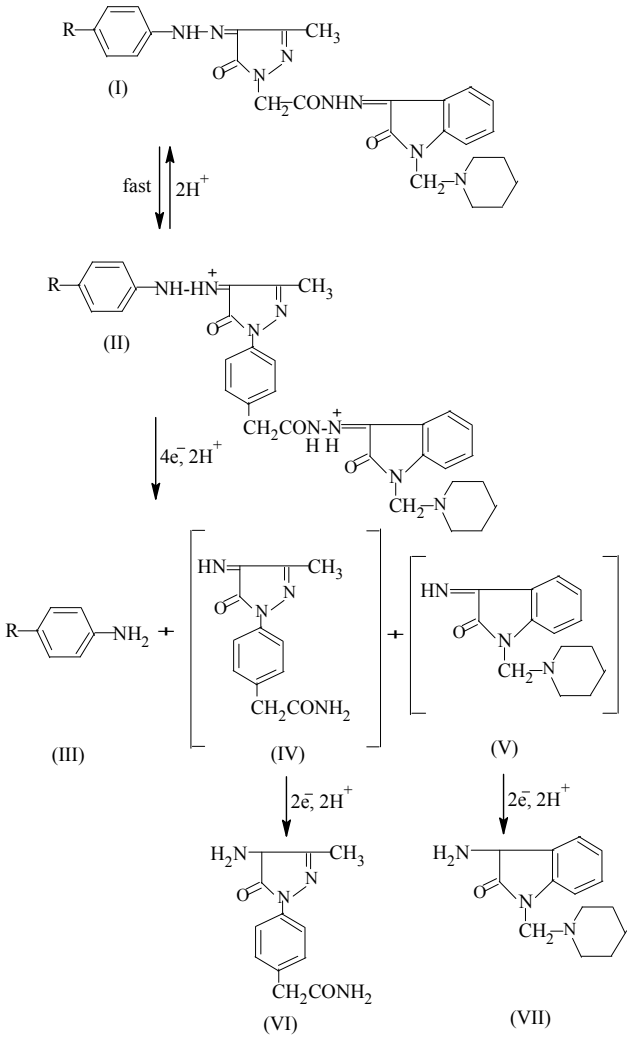
3.2. Cyclic voltammetric studies of a-f at HMDE

The cyclic voltammetric data of a-f in solutions of pH 2.1, 4.1, 6.1, 8.1 and 10.1 are shown in Table 2. The model cyclic voltammograms of 'a' in typical pH media are given in the Fig. 6.

Table 2

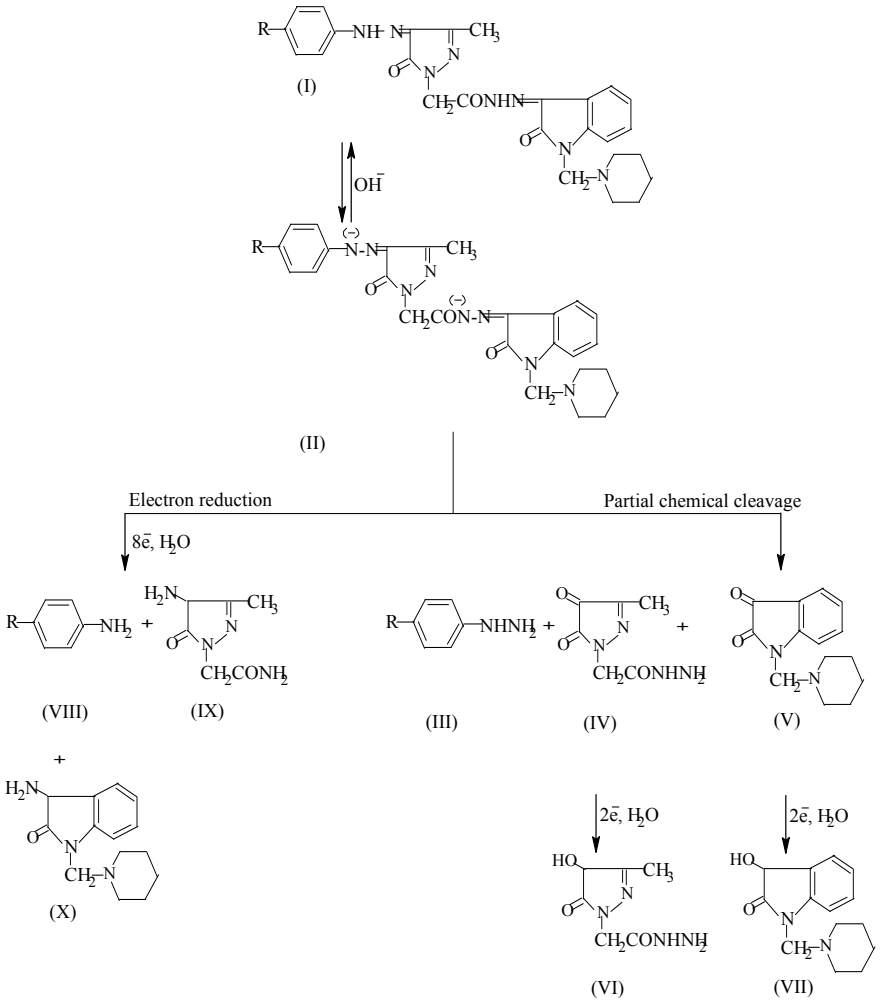
Cyclic voltammetric results of [3-methyl-5-oxo-4-(4'-substituted aryl hydrazono)-4,5-dihydro-pyrazol-1-yl]-acetic acid (2-oxo-1-piperidine-1-yl-methyl-1,2-dihydro–indol-3-yl-idene)- hydrazide at HMDE; Medium : Aqueous di-methyl formamide (40% v/v).

pH	Scan Rate $V s^{-1}$	-Epc I (V)	-Epc II (V)	-Epc III (V)	-Epc inv (V)	Ipc I (uA)	IpcII (uA)	IpcIII (uA)	Ipc inv (uA)	-Epc I (V)	-Epc II (V)	-Epc III (V)	-Epc inv (V)	Ipc I (uA)	IpcII (uA)	IpcIII (uA)	Ipc inv (uA)	-Epc I (V)	-Epc II (V)	-Epc III (V)	-Epc inv (V)	Ipc I (uA)	IpcII (uA)	IpcIII (uA)	Ipc inv (uA)
		H								**Methyl**								**Methoxy**							
4.1	0.010	0.7	0.80		0.75	1.9	1.3		0.7	0.87	0.97		0.72	2.3	1.7		0.8	0.97	1.07		0.78	2.2	2.8		0.8
	0.050	0.78	0.88		0.87	4.2	2.9		1.5	0.95	1.05		0.84	5.1	3.8		1.8	1.05	1.15		0.90	4.9	6.2		1.8
	0.100	0.82	0.92		0.96	6.0	6.0		2.2	0.99	1.09		0.93	7.2	5.3		2.5	1.09	1.19		0.99	6.9	8.8		2.5
	0.200	0.86	0.96		1.05	8.5	5.8		3.1	1.03	1.13		1.02	10.2	7.6		3.6	1.13	1.23		1.08	9.8	12.5		3.6
	0.500	0.98	1.08		1.23	13.4	9.1		4.9	1.15	1.25		1.20	16.2	12.0		5.6	1.25	1.35		1.26	15.5	19.7		5.6
6.1	0.010	0.9	1.0		0.84	1.1	1		0.9	1.09	1.19		0.87	1.5	1.2		1.1	1.21	1.31		0.93	1.4	1.3		1.1
	0.050	0.98	1.08		0.96	2.4	2.2		2.0	1.17	1.27		0.99	3.3	2.7		2.4	1.29	1.39		1.05	3.1	2.9		2.4
	0.100	1.02	1.12		1.05	3.4	3.1		2.8	1.21	1.31		1.08	4.7	3.8		3.5	1.33	1.44		1.14	4.4	4.1		3.5
	0.200	1.06	1.16		1.14	4.9	4.4		4.0	1.25	1.35		1.17	6.7	5.3		4.9	1.37	1.47		1.23	6.2	5.8		4.9
	0.500	1.18	1.28		1.32	7.7	7.0		6.3	1.37	1.47		1.35	10.6	8.4		7.7	1.49	1.59		1.41	9.9	9.1		7.7
8.1	0.010	1.07	1.17	1.56		1.0	0.7	1.8		1.28	1.38	1.53		1.4	0.9	2.2		1.41	1.51	1.59		1.3	1.0	2.1	
	0.050	1.15	1.25	1.64		2.2	1.5	4.0		1.36	1.46	1.61		3.1	2.0	4.9		1.49	1.59	1.67		2.9	2.2	4.7	
	0.100	1.19	1.29	1.68		3.1	2.2	5.6		1.40	1.50	1.65		4.4	2.8	6.9		1.53	1.63	1.71		4.1	3.1	1.6	
	0.200	1.23	1.33	1.72		4.4	3.1	8.0		1.44	1.54	1.69		6.2	4.0	9.8		1.57	1.67	1.75		5.8	4.4	9.4	
	0.500	1.35	1.45	1.84		7.0	4.9	12.7		1.56	1.66	1.81		10.6	6.3	15.5		1.69	1.79	1.87		9.1	7.0	14.8	
		Ethoxy								**Chloro**								**Bromo**							
4.1	0.010	0.95	1.05		0.84	2.1	1.6		0.9	0.45	0.55		0.42	2.5	1.8		0.8	0.50	0.60		0.45	2.3	1.7		0.8
	0.050	1.03	1.13		0.96	4.7	0.6		2.0	0.53	0.63		0.54	5.6	4.0		1.8	0.58	0.68		0.57	5.1	3.8		1.58
	0.100	1.07	1.17		1.05	6.6	5.0		2.8	0.57	0.67		0.63	7.9	5.6		2.5	0.62	0.72		0.66	7.2	5.3		2.5
	0.200	1.11	1.21		1.14	9.4	7.1		4.0	0.61	0.71		0.72	11.2	8.0		3.6	0.66	0.76		0.75	10.25	7.6		3.6
	0.500	1.23	1.33		1.32	14.8	11.3		6.3	0.73	0.83		0.90	17.7	12.7		5.6	0.78	0.88		0.93	16.2	12.0		5.6
6.1	0.010	1.19	1.29		0.99	1.3	1.2		1.3	0.64	0.74		0.60	1.6	1.2		1.2	0.69	0.79		0.66	1.5	1.2		1.3
	0.050	1.27	1.37		1.11	2.9	2.7		2.9	0.72	0.82		0.72	0.36	2.7		2.7	0.77	0.87		0.78	3.3	2.7		2.9
	0.100	1.31	1.41		1.20	4.1	3.8		4.1	0.76	0.86		0.81	5.0	3.8		3.8	0.81	0.91		0.87	4.7	3.8		4.1
	0.200	1.35	1.45		1.29	5.8	5.3		5.8	0.80	0.90		0.90	7.1	5.3		5.3	0.85	0.95		0.96	6.7	5.3		5.8
	0.500	1.47	1.57		1.47	9.1	8.4		9.1	0.92	1.02		1.08	11.3	8.4		8.4	0.97	1.07		1.14	10.6	8.4		9.1
8.1	0.010	1.39	1.49	1.54		1.2	0.9	2.2		0.77	0.87	1.58		1.5	0.9	2.1		0.83	0.93	1.59		1.4	0.9	2.2	
	0.050	1.47	1.57	1.62		1.7	2.0	4.9		0.85	0.95	1.66		3.3	2.0	4.7		0.91	1.01	1.67		3.1	2.0	4.9	
	0.100	1.51	1.61	1.66		3.8	2.8	6.9		0.89	0.99	1.70		4.7	2.8	6.6		0.95	1.05	1.71		4.4	2.8	6.9	
	0.200	1.55	1.65	1.70		5.3	4.0	9.8		0.93	1.03	1.74		6.7	4.0	9.4		0.99	1.09	1.75		6.2	4.0	9.8	
	0.500	1.67	1.77	1.82		8.4	6.3	15.5		1.05	1.15	1.86		10.6	6.3	14.8		1.11	1.21	1.87		9.9	6.3	15.5	



Scheme 1. Reduction mechanism in acidic medium

It is seen from the data that two cathodic peaks in acidic solutions and three cathodic peaks in basic solutions at all sweep rates were noticed in the pH range 2.1-10.1. The peak potentials were shifted to more negative values and there was an increase in peak height with increase in sweep rates. The number of peaks was the same as that of waves observed in DC polarographic studies. The reduction mechanism is therefore assumed to be similar that described in Scheme 1 and 2.



Scheme 2. Reduction mechanism in basic mediu**m**

The peak potentials of the cathodic peaks were shifted to more negative values and the cathodic peak currents increase with the increase in the scan rate [29]. The anodic peak was absent in the reverse scan. The diffusion controlled nature of the reduction process was confirmed by linear i_{pc} *vs* $\gamma^{1/2}$ plots passing through the origin. The increase in peak currents with increase of the concentration of the depolarizer further supports this conclusion. The plot of $i_{pc/\gamma}{}^{1/2}$ *vs* γ confirms irreversible nature of reduction process and was similar to the case II of Nicholson Shain criteria [30]. Cathodic peak potentials (Table 2) were shifted to more negative values with the rise in pH of the solution. The first and second cathodic peak currents decrease with increase in pH of solutions from 2.1 to 10.1, while

that of third cathodic peak noticed in alkaline solutions remain constant. The results were similar to those obtained in polarographic studies. Voltammograms recorded at typical pH value 4.1 under repeated cycles reveal that the peak height diminishes with repetition of cycles. However, no significant change was noticed in the shape of the voltammogram. This may be ascribed to the adsorption of depolarizer on the mercury solution interface.

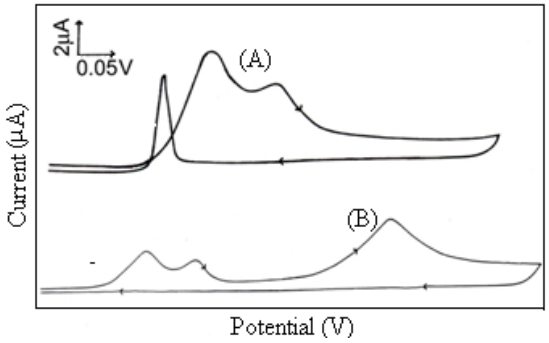


Fig. 6. Cyclicvoltammograms of 'a' at HMDE; Medium = Dimethyl formamide (40% v/v); Sweep rate: 100 mv/s; (A) pH 4.1, Starting potential 0.5 V; (B) pH 8.1, Starting potential 1.0 V, Sweep rate: 100 mv/s.

3.3. Cyclic voltammetric studies of [3-methyl-5-oxo-4-(4'-substituted aryl hydrazono)-4,5-dihydro-pyrazol-1-yl]-acetic acid-(2-oxo-1-piperidine-1-ylmethyl-1,2-dihydro–indol-3-yl-idene)-hydrazide (a-f) with crown – ether modified carbon paste electrode.

Cyclic voltammetric behavior of a-f were carried out at modified carbon paste electrode in buffer solutions of pH 2.1, 4.1, 6.1, 8.1 and 10.1 at different scan rates i.e. 10 mVs^{-1}, 20 mVs^{-1}, and 50 mVs^{-1}, 100 mVs^{-1}, 200 mVs^{-1}, 300 mVs^{-1}. The model cyclic voltammograms of 'a' in typical pH media are given in the Fig. 7.

The compounds a-f exhibit four well defined cathodic peaks at high scan rates (100-500 mVs^{-1}) in solutions of pH 2.1-6.1 and show six cathodic peaks in alkaline solutions of pH 8.1-10.1. Peak potentials and peak currents change with the variation in scan rates. The cathodic peak potentials were shifted to more negative values with increase in pH. The peak current deceases with increase in pH. (Table 3 and 4).

The irreversible nature of electrode process was characterized by the dependence of peak potential on sweep rate, the absence of anodic peak in the reverse scan, and a plot of $i_{pc}/ \upsilon^{1/2}$ vs sweep rate which was a straight line parallel to sweep rate axis. The above mentioned facts clearly rules out the possibility of a fast electron

transfer which is characteristic of a reversible behaviour. The linear variation of i_{pc} with $\upsilon^{1/2}$ suggests the diffusion controlled nature of the electrode process [30]. The plots of Epc vs pH were similar to $E_{½}$ vs pH plots and this lends support to the findings in DC polarography.

3.4. Comparison between polarographic and cyclic voltammetric studies

Polarographic studies reveal that the compounds a–f exhibit two well defined waves in solutions of pH of 2.1-6.1 and three waves in solutions of pH 8.1 – 10.1. Cyclic voltammetric studies reveal that the compounds under study give two cathodic peaks in acidic solutions of pH 2.1 – 6.1 and three cathodic peaks in alkaline solutions of pH 8.1-10.1. The number of polarographic waves was same as that of number of cyclic voltammograms observed at HMDE. However, a quite different behaviour was noticed in cyclic voltammetric studies at MCPE. The data presented in the tables indicates that four cathodic peaks were observed at higher sweep rates (100 mVs^{-1}, 200 mVs^{-1}, 300 $mVs^{-1}$and 500 mVs^{-1}) in acidic solutions of pH 2.1 – 6.1.

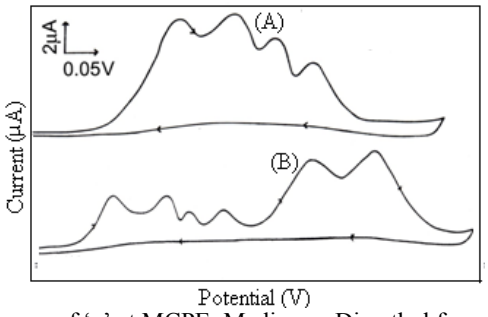


Fig. 7. Cyclicvoltammograms of 'a' at MCPE; Medium = Dimethyl formamide (40% v/v); Sweep rate: 100 mv/s; (A) pH 4.1, Starting potential 0.8 V; (B) pH 8.1, Starting potential 1.1 V.

The first polarographic wave manifest itself as two cathodic peaks ($-E_{pcI}$ and $-E_{pcII}$) in cyclic voltammetric studies (at MCPE) under similar experimental conditions, whereas the second polarographic wave was split into two cathodic peaks ($-E_{pcIII}$ and $-E_{pcIV}$). The first and second reduction steps were attributed to the four electron reduction of azomethine groups (>C=N-NH-) in different chemical environments. Compounds a-f contain two such groups. The reduction of each azomethine group was taking place in two steps through imine intermediate involving four electron transfer.

An inspection of the peak potentials data (Table 3 and 4) suggest that potentials of the first peak were close to that of second peak. Therefore these two peaks were attributed to the two reduction steps of hydrazone group to the amine stage via imine intermediate.

Table 3

Cyclic voltammetric results of [3-methyl-5-oxo-4-(4'-substituted aryl hydrazono)-4,5-dihydro-pyrazol-1-yl]-acetic acid (2-oxo-1-piperidine-1-ylmethyl-1,2-dihydro–indol-3-yl-idene)- hydrazide at MCPE; Medium : Aqueous di-methyl formamide (40% v/v)

pH	Scan rate Vs^{-1}	$-E_{pci}$ (V)	$-E_{pcII}$ (V)	$-E_{pcIII}$ (V)	$-E_{pcIV}$ (V)	$-E_{pcV}$ (V)	$-E_{pcVI}$ (V)	I_{pci} (µA)	I_{pcII} (µA)	I_{pcIII} (µA)	I_{pcIV} (µA)	I_{pcV} (µA)	I_{pcVI} (µA)
-H													
4.1	0.100	0.98	1.10	1.10	1.14	-	-	8.8	9.1	5.7	5.6	-	-
	0.200	1.02	1.14	1.14	1.18	-	-	12.5	12.9	8.0	8.5	-	-
	0.300	1.08	1.20	1.20	1.24	-	-	15.3	15.8	9.8	10.4	-	-
	0.500	1.14	1.26	1.26	1.30	-	-	19.7	20.5	12.7	13.4	-	-
8.1	0.100	1.29	1.45	1.49	1.59	1.78	1.93	4.7	5.0	3.1	3.5	8.5	8.8
	0.200	1.33	1.49	1.53	1.63	1.82	1.97	6.7	7.1	4.4	4.9	12.0	12.5
	0.300	1.39	1.55	1.59	1.69	1.88	2.03	8.2	8.7	5.4	6.0	14.7	15.3
	0.500	1.45	1.61	1.65	1.75	1.94	2.09	10.6	11.3	7.07	7.7	19.0	19.7
4'-methyl													
4.1	0.100	1.15	1.27	1.27	1.41	-	-	10.7	11.0	7.9	8.2	-	-
	0.200	1.19	1.31	1.31	1.45	-	-	15.2	15.6	11.2	11.6	-	-
	0.300	1.25	1.37	1.37	1.51	-	-	18.6	19.1	13.7	14.2	-	-
	0.500	1.31	1.43	1.43	1.57	-	-	24.0	24.7	17.7	18.4	-	-
8.1	0.100	1.50	1.66	1.70	1.80	1.75	1.90	6.2	6.6	4.1	4.4	10.1	10.4
	0.200	1.54	1.70	1.74	1.84	1.79	1.94	8.9	9.4	5.8	6.2	14.3	14.7
	0.300	1.60	1.76	1.80	1.90	1.85	2.0	10.9	11.5	7.1	7.3	17.5	18.0
	0.500	1.66	1.82	1.86	1.96	1.91	2.06	14.1	14.8	9.1	9.9	22.6	23.3
4'-methoxy													
4.1	0.100	1.25	1.37	1.37	1.51	-	-	10.1	10.4	12.9	13.2	-	-
	0.200	1.28	1.41	1.41	1.55	-	-	14.3	14.7	18.3	18.7	-	-
	0.300	1.35	1.47	1.47	1.61	-	-	17.5	18.0	22.4	23.0	-	-
	0.500	1.41	1.53	1.53	1.67	-	-	22.6	23.3	29.0	29.7	-	-
8.1	0.100	1.63	1.79	1.83	1.93	1.81	1.96	506	6.0	4.7	5.0	9.8	10.1
	0.200	1.67	1.83	1.87	1.97	1.85	2.0	8.0	8.5	6.7	7.1	13.8	14.3
	0.300	1.73	1.89	1.93	2.03	1.91	2.06	9.8	10.4	8.2	8.7	17.0	17.5
	0.500	1.79	1.95	1.99	2.09	1.97	2.12	12.7	13.4	10.6	11.3	21.9	22.6

Table 4

Cyclic voltammetric results of [3-methyl-5-oxo-4-(4'-substituted aryl hydrazono)-4,5-dihydro-pyrazol-1-yl]-acetic acid (2-oxo-1-piperidine-1-ylmethyl-1,2-dihydro–indol-3-yl-idene)- hydrazide at MCPE; Medium : Aqueous DMF (40% v/v)

pH	Scan rate Vs^{-1}	$-E_{pc,I}$ (V)	$-E_{pc,II}$ (V)	$-E_{pc,III}$ (V)	$-E_{pc,IV}$ (V)	$-E_{pc,V}$ (V)	$-E_{pc,VI}$ (V)	$I_{pc,I}$ (µA)	$I_{pc,II}$ (µA)	$I_{pc,III}$ (µA)	$I_{pc,IV}$ (µA)	$I_{pc,V}$ (µA)	$I_{pc,VI}$ (µA)
4'-ethoxy													
4.1	0.100	1.23	1.35	1.35	1.31	-	-	9.8	10.1	7.6	7.9	-	-
	0.200	1.27	1.39	1.39	1.35	-	-	13.8	14.3	10.7	11.2	-	-
	0.300	1.33	1.45	1.45	1.41	-	-	17.0	17.5	13.1	13.7	-	-
	0.500	1.39	1.51	1.51	1.47	-	-	21.9	22.6	17.0	17.7	-	-
8.1	0.100	1.81	1.77	1.81	1.91	1.76	41.91	5.6	6.0	4.1	4.4	10.4	10.7
	0.200	1.65	1.81	1.85	1.95	1.80	1.95	8.0	8.5	5.8	6.2	14.7	15.2
	0.300	1.71	1.87	1.91	2.01	1.86	2.01	9.8	10.4	7.1	7.6	18.0	18.6
	0.500	1.77	1.93	1.97	2.07	1.92	2.07	12.7	13.4	9.1	9.9	23.3	24.0
4'-chloro													
4.1	0.100	0.73	0.85	0.85	0.99	-	-	13.9	15.1	8.5	8.8	-	-
	0.200	0.77	0.89	0.89	1.03	-	-	21.0	21.4	12.0	12.5	-	-
	0.300	0.83	0.95	0.95	1.09	-	-	25.7	26.2	14.7	15.3	-	-
	0.500	0.89	1.01	1.01	1.15	-	-	31.1	33.9	19.0	17.7	-	-
8.1	0.100	0.99	1.15	1.19	1.29	1.80	1.95	6.9	47.2	4.1	4.4	9.8	10.1
	0.200	1.03	1.19	1.23	1.33	1.84	1.99	9.8	10.2	5.8	6.2	13.8	14.3
	0.300	1.09	1.25	1.29	1.39	1.90	2.05	12.0	12.5	7.1	7.6	17.0	17.5
	0.500	1.15	1.31	1.35	1.45	1.96	2.11	15.5	16.2	9.1	9.9	21.9	22.6
4'-bromo													
4.1	0.100	0.78	0.90	0.90	1.04	-	-	10.7	11.0	7.9	8.2	-	-
	0.200	0.82	0.94	0.94	1.08	-	-	15.2	15.6	11.2	11.6	-	-
	0.300	0.88	1.00	1.00	1.14	-	-	18.6	19.1	13.7	14.2	-	-
	0.500	0.94	1.06	1.06	1.20	-	-	24.0	24.7	17.7	18.4	-	-
8.1	0.100	1.05	1.21	1.25	1.35	1.81	1.96	6.6	6.9	4.1	4.4	10.4	10.7
	0.200	1.09	1.25	1.29	1.39	1.85	2.0	9.4	9.8	5.8	6.2	14.7	15.2
	0.300	1.15	1.31	1.35	1.45	1.91	2.06	11.5	10.0	7.1	7.6	18.0	18.6
	0.500	1.21	1.37	1.41	1.51	1.97	2.12	14.8	15.5	9.1	9.9	23.3	24.0

The reason for the appearance of a single wave in polarographic studies may be due to the fact that the half wave potentials of the two waves on the potential axis were so close to each other that the two waves merge and they are observed as a single polarographic wave.

In alkaline medium (pH>pKa) 'a' exists in the azomethine anionic form (II) and the later was susceptible to chemical cleavage partially in alkaline solutions to the corresponding carbonyl compounds (V and VI). Each of the first and second waves noticed in the polarographic studies have appeared as two cathodic peaks ($-E_{pcI}$, $-E_{pcII}$ and $-E_{pcIII}$, $-E_{pcIV}$) in cyclic voltammetric studies at MCPE. Similarly, the third wave noticed in polarographic studies was a composite wave ascribed to the two-electron reductions of non equivalent heterocyclic ketones to carbinol stage. This wave has been separated in two cathodic peaks ($-E_{pc}$v, $-E_{pcVI}$) in cyclic voltammetric studies at modified electrode.

4. Conclusion

An electrochemical study of certain novel hydrazides bearing pyrazoline-5-one and indole moieties were performed by polarography at HMDE, cyclic voltammetry at HMDE and cyclic voltammetry MCPE. The electrochemical reduction of these compounds was diffusion controlled, irreversible and pH dependent. The results from these studies were correlated to propose plausible electrode reduction mechanism in acidic and basic media.

REFERENCES

[1]. *R.E. Dolle,* Comprehensive survey of combinatorial library synthesis, J. Comb. Chem., 3, 2000, pp. 477–517

[2]. *S. Hanessian, G. McNaughton-Smith, H.G. Lombart and W.D. Lubell,* Design and synthesis of conformationally constrained amino acids as versatile scaffolds and peptide mimetics, Tetrahedron, 53, 1997, pp. 12789–12854

[3]. *H. Abdel-Gawad, H. A. Mohamed, K. M. Dawood and F. A. Badria,* Synthesis and antiviral activity of new indole-based heterocycles, Chem Pharm Bull (Tokyo), 58, 2010, pp. 1529-1531

[4]. *M. C. Lucas, R. J. Weikert, D. S. Carter, H. Y. Cai, R. Greenhouse, P. S. Iyer,* Design, synthesis and biological evaluation of new monoamine reuptake inhibitors with potential therapeutic utility in depression and pain, Bioorg. Med. Chem Lett., 20, 2010, p. 5559–5566

[5]. *N. I. Ziedan, F. Stefanelli, S. Fogli, A. D. Westwell,* Design, synthesis and pro-apoptotic antitumour properties of indole-based 3,5-disubstituted oxadiazoles, Eur J Med Chem., 45, 2010, pp. 4523-4530

[6]. *R. Ben-Daniel, W. Deuther-Conrad, M. Scheunemann, J. Steinbach, P. Brust and E. Mishani,* Carbon-11 labeled indolylpropylamine analog as a new potential PET agent for imaging of the serotonin transporter, Bioorg Med Chem. 16, 2008, pp. 6364–6370

[7]. *P. Wang, J. Liu, H. Xing,, Y. Liu, W. Xie and G. Zhao,* Synthesis and anticancer activity of novel 5-(indole-2-yl)-3-substituted 1,2,4-oxadiazoles, Drug Discov Ther., 6, 2012, pp. 133-139

[8]. *J. S. Biradar, B. S. Sasidhar, R. Parveen,* Synthesis, antioxidant and DNA cleavage activities of novel indole derivatives, Eur J Med Chem., 45, 2010, pp. 4074-4079

[9]. *M. Dastagiri Reddy, A. Raghavendra Guru Prasad, Y. N. Spoorthy and L. K. Ravindranath,* Synthesis, Characterization and Antimicrobial Activity of Certain novel Aryl Hydrazone Pyrazoline-5-Ones Containing Thiazole Moiety, Advanced Pharmaceutical Bulletin, 3, 2013, pp. 153-159

[10]. *Krishna Naik, A. Raghavendra Guru Prasad, Y. N. Spoorthy and L. K. Ravindranath,* Design, synthesis, characterization and antimicrobial evaluation of new pyrazoline-5-ones, Journal of Applied Pharmacy, 4, 2013, pp. 720-730

[11]. *L. Jing, L. Wang, Y. Zhao, R. Tan, X. Xing, T. Liu, W. Huang, Y. Luo and Z. Li,* Synthesis, crystal structure and evaluation of cancer inhibitory activity of 4-[indol-3-yl-methylene]-1*H*-pyrazol-5(4*H*)-one derivatives, Journal of Chemical Research, 36, 2012, pp. 691-696

[12]. *M. Amir and S. Kumar,* Synthesis and anti-inflammatory, analgesic, ulcerogenic and lipid peroxidation activities of 3,5-dimethyl pyrazoles, 3-methyl pyrazol-5-ones and 3,5-disubstituted pyrazolines, Indian Journal of Chemistry, B 44, 2005, pp. 2532–2537

[13] *E. Badawey and I. M. El-Ashmawey,* Non-steroidal anti- inflammatory agents- Part 1: Anti-inflammatory, analgesic and antipyretic activity of some new 1(pyrimidin-2-yl)-3-pyrazoline-5-ones and 2-(pyrimidin-2yl)-1,2,4,5,6,7-hexahydro-3H-indazol 3-ones, Eur J Med Chem., 33, 1998, pp. 349-361

[14]. *O. I. El-Sabbhagh, M. M. Baraka, S. M. Ibrahim, C. Pannecouque, G. Andrei, R. Snoeck, J. Balzarini and A. A. Rashad,* Synthesis and antiviral activity of new pyrazole and thiazole derivatives, Eur J Med Chem., 44, 2009, pp. 3746-3753

[15]. *T. Chandra, N. Garg, S. Lata, K. K. Saxena and A. Kumar,* Synthesis of substituted acridinyl pyrazoline derivatives and their evaluation for anti-inflammatory activity, Eur J Med Chem., 45, 2010, pp. 1772–1776

[16]. *J. C. Vire and J. M. Kauffmann,* Trends in electrochemistry in drug analysis, Current Topics in Electrochemistry, 3, 1994, pp. 493–515

[17]. *S. Suzen and S. A. Ozkan,* Combination of electrochemical, spectrometric and other analytical techniques for high throughput screening of pharmaceutically active compounds, Combinatorial Chemistry and High Throughput Screening, 13, 2010, pp. 658–664

[18]. *H. Shirinzadeh, A. D. Yilmaz, M. Gumustas, S. Suzen, S. Ozden and S. A. Ozkan,* Electrochemical behavior of indole-3-carboxaldehyde izonicotinoyl hydrazones: discussion on possible biological behavior, Combinatorial Chemistry and High Throughput Screening, 13, 2010, pp. 619–627

[19] *H. T. K.* B*ritton and R. A. Robinson,* Universal buffer solutions and the dissociation constant of veronal, J. Chem. Soc., 1931, pp. 1456–1462.

[20]. *A. I. Vogel,* A Text Book of Quantitative Inorganic Analysis. Longman, Third Edition, 1961

[21]. *L. K.Ravindranath, K. Srikanth, M. Dastagiri Ready, S. D. Ishrath Begum,* Synthesis of novel mannich bases containing pyrazolones and indole systems, Heterocyclic Communications, 15, 2009, pp. 443-449

[22]. *Benoit Soucaze-Guillous and Henning Lund,* Electrochemical Reduction of Benzaldehyde Benzoylhydrazone in Aprotic Media Acta Chem. Scand., 51, 1997, pp. 354-356

[23]. *A. Raghavendra Guru Prasad, V. Seshagiri and L.K. Ravindranath,* Polarographic Investigations of Certain Propiophenone benzoic acid hydrazones, *Jordan Journal of Chemistry, 6,* 2011, pp. *51-64*

[24]. *W. V. Malik, R. N. Goyal and R. Jain,* Polarographic reduction of some arylazopyrazoles in N, N'-dimethylformamide, J. Electroanal. Chem., 87, 1978, pp. 129-135
[25]. *K. Ramana Kumar, A. Raghavendra Guru Prasad, V. Srilalitha, G. Narayana Swami and L. K. Ravindranath,* Synthesis and electrochemical investigations on certain pyrazolin-5-ones, Scientica Iranica (Transactions in Chemistry and Chemical Engineering), 19, 2012, pp. 605-618
[26]. *J.J. Lingane,* Coulometric Analysis, J. Am. Chem. Soc. 67, 1945, pp. 1916-1922
[27]. *B. Nygard,* Azobenzene-hydrazobenzene polarographic reversibility problem, Arkiv. Kemi., 20, 1962, pp. 163-183
[28]. *R. N. Adams,* Electrochemistry at Solid Electrodes, Marce Dekkar Inc., New York, 1969
[29]. *J. W. Ross and R. D. Demars*, I. Shain, Analytical Applications of Hanging Mercury Drop Electrode, Anal. Chem. 28, 1956, pp. 1768-1772
[30]. *R. S. Nicholson and Shain,* Theory of Stationary Electrode Polarography - Single Scan and Cyclic Methods Applied to Reversible, Irreversible, and Kinetic Systems, Anal. Chem., 36, 1964, pp. 706-723