

Antioxidant Activity of Phenyl Alanine Mandelates by Chemical and Electrochemical Methods

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Abstract

Food decomposition in human body due to the redox reactions results in the formation of reactive oxygen species (ROS). ROS obtained during metabolic activities in our body are responsible for cancerous diseases. ROS are scavenged by hydroxyl radicals present in the small organic molecules. Novel small organic molecules like mandelic acid - amino acid complexes, possess the weak hydrogen bonds and vanderwaals forces of attractions in the complex formation results in the antioxidant property. The title compounds, Rphenyl alanine-S-mandelate (RPASMA), Bis-L-phenyl alanine mandelate (BLPAMA) and L-phenyl alanine bis mandelate (BMALPA) are synthesised, carried out characterisation studies like FTIR, NMR, TG-DTA, mass, UV and melting point and grown single crystal by slow evaporation technique confirmed the structure by single crystal XRD. The electrochemical behaviour of the phenyl alanine mandelates show the existence of redox activity using cyclic voltammetry and is confirmed by comparing with the chemical behaviour using DPPH (1,1-diphenyl-2-picrylhydrazyl) radical scavenging method.

Index terms— ROS, phenyl alanine mandelates, EC50, IC50, DPPH, ARP.

1 I. Introduction

andelic acid (2-hydroxy-2-phenyl acetic acid) and Phenyl alanine (2-amino-3-phenyl propanoic acid) exist in racemic forms. The complexes of mandelic acid and phenyl alanine are having hydroxyl group, acid group and amino group which gives the salt formation due to the protonation of amino group through the formation of covalent bond and weak vanderwaals forces of attraction between acidic hydrogen, basic amino group and hydroxyl groups [1]. The zwitterionic structure of amino acid enhances the formation of salt complex with carboxylic acids. Donor and acceptor concept of hydrogen in the salt helps in the redox activity and the radical scavenging activity of the title compounds [2]. The present study indicates the usefulness of the title compounds having hydroxy substitution and conjugation act as antioxidant material to scavenge the free radicals formed during the metabolic activities in the human body. It is further supported by the electrochemical behaviour of the compounds due to the structure property activity of the title compounds [3][4]. The reactive oxygen species formed during metabolic activities are nullified by the exogenous antioxidant having high antiradical power. The increase in electron donating groups in the title compounds modulate antioxidant capacity and they can be used to fight against oxidative stress diseases like cancer, cardiovascular disorders, neurodegenerative pathologies [5].

2 II. Experiment

AlfaAaser mandelic acid and Nice chemicals Lphenyl alanine were mixed in water in 1:2 and 2:1 ratios respectively. Obtained almost clear solution after agitation at room temperature for 2-3 hours, filtered and kept for slow evaporation at room temperature. Observed the crystals formation after 8 days harvested crystals after 28 days showed homogenous on TLC and confirmed the melting point as 184°. The recrystallisation of 1:2 mandelic

acid [6] and L-phenyl alanine resulted in the diastereomeric isolation of R-phenyl alanine-S-mandelate [7] which showed homogenous on TLC and has melting point 174°C. Characterisation studies, mass analysis, single crystal XRD studies confirmed the structure of the title compounds and the possession of second order non linear susceptibilities due to non centrosymmetric structure.

The increase in the presence of hydroxyl groups and conjugation in the molecular structure favours the oxidation and reduction activity. It is confirmed by the electrochemical behaviour using cyclic voltammetry and the radical scavenging activity using DPPH for the title compounds.

3 Basis of DPPH scavenging method DPPH -free radical form DPPH -reduced form a) DPPH -free radical and reduced form

The molecule of 1,1-diphenyl-2-picryl-hydrazyl (DPPH) is characterised as a stable free radical by virtue of the delocalisation of the spare electron over the molecule as a whole, so that the molecules do not dimerise, as would be the case with most other free radicals. The delocalisation also gives rise to the deep violet colour, characterised by an absorption in methanol solution at 517 nm [8].

When a solution of DPPH is mixed with that of a substance that can donate a hydrogen atom, then this gives rise to the reduced form with the loss of this violet colour (although there would be expected to be a residual pale yellow colour from the picryl group still present). Representing the DPPH radical by Z

? and the donor molecule by AH.

The free radical form reacts with the substance AHZ ? + AH = ZH + A ? [1]

where ZH is the reduced form and A

? is free radical produced in this first step. This latter radical will then undergo further reactions which control the overall stoichiometry, that is, the number of molecules of DPPH reduced (decolorised) by one molecule of the reductant.

The reaction [1] is therefore intended to provide the link with the reactions taking place in an oxidising system, such as the autooxidation of an unsaturated substance; the DPPH molecule Z ? is thus intended to represent the free radicals formed in the system whose activity is to be suppressed by the substance AH.

4 % of Inhibition = (A of control - A of Test)/A of control * b) Electrochemical study

Non-aqueous media cyclic voltammetry (CV) study using Pt electrodes show the possibility of electrooxidation, acceptor -donor interactions of the title compounds and the starting materials [9][10][11].

5 III. Results and Discussion

The DPPH annihilation activity of free radicals is calculated as % inhibition [12][13][14]. The control Ascorbic acid show the maximum % inhibition compared to the title compounds as shown in Table 1. The maximum antioxidant power is shown by more hydroxyl group containing BMALPA Figure ??5. The increase in hydroxyl group substitution in the title compounds, the presence of increase in conjugation increases the % inhibition is indicated in the Figure ??6. EC 50 or IC 50 is more for BMALPA compared to RPASMA and BLPAMA. The comparative respective efficient concentration and antiradical power shown by the title compounds is given in the Table 2. The presence of electron donating amino group, hydroxyl group and acidic hydrogen in the title compounds show low oxidation potential due to electrooxidation which corresponds to high antioxidant power [15][16][17].

The electrochemical behaviour of the title compounds and the starting materials are compared using the cyclic voltammetry measurements. Donor-acceptor interactions leads to hydrogen bond formation [18,19]. The electrochemical oxidation of the title compounds show higher area under anodic wave form which corresponds to higher antioxidant capacity. The presence of electron donating groups have lower half wave potential, higher antioxidant activity and higher reducing power. In the title compounds the presence of more hydroxyl groups, electron donating groups in BMALPA shows higher antioxidant activity. Radical scavenging activity, antiradical power, structure property activity leads to the high antioxidant activity of the title compounds and can be used as fighting agents to nullify the ROS generated during meabolic activities [20].

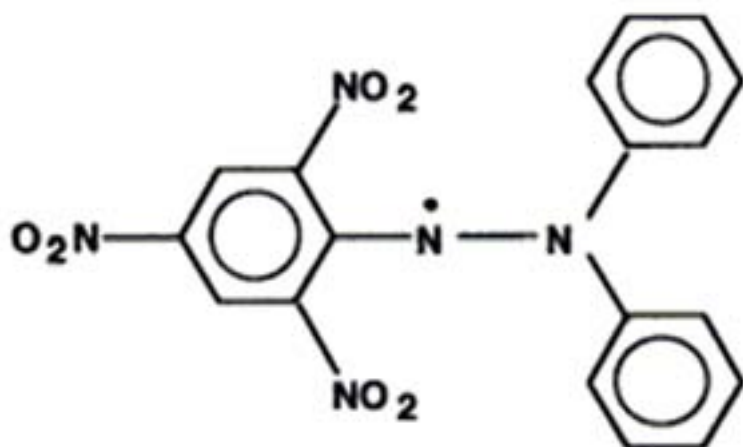
6 IV. Conclusion

Novel organic salt complexes can act as exogenic antioxidants is confirmed from the comparative study of title compounds using radical scavenging DPPH method and electrochemical cyclic voltammetric method. The comparable results from the both methods give optimistic thought to over come the prevailing health issues caused by the present life style of the modern world. The use of starting materials to synthesise the title compounds find



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Figure 1: Figure 1 :Figure 2 :Figure 3 :



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Figure 2: Figure 4 :Figure 5 :Figure 6 :

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Figure 3:

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Conc micro gram	% inhibition			
	ASCORBIC ACID	RPASMA	BLPAMA	BMALPA
25	58.88	40.55		15.69
50	64.42	53.83		26.74
100	83.09	63.28		39.49
250	96.44	74.77		54.72

Figure 4: Table 1 :

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SAMPLE	IC 50	ARP
BLPAMA	206.4	0.00485
BMALPA	140.47	0.00712
RPASMA	151.15	0.00662

Figure 5: Table 2 :

94 many medical applications , constituent to protect the central nervous system the harmless effects are expected
95 for the title compounds in the in-vivo studies. ^{1 2 3}

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