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An efficient one-pot synthesis of polyphenolic amino acids and evaluation of their radical-scavenging activity

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ABSTRACT

A simple and efficient procedure for the synthesis of *N*-acyl 4-hydroxy, 4-hydroxy-3-methoxy and 3,4-dihydroxy phenylglycine amides by a strategy based on the multicomponent Ugi reaction is proposed. Hydroxybenzaldehyde derivatives were reacted with 4-methoxybenzylamine, cyclohexyl isocyanide and benzoic acid or 2-naphthylacetic acid to give Ugi adducts that were treated with trifluoroacetic acid yielding *N*-acyl hydroxyphenylglycine amides in good yields. The same procedure using as acid component protocatechuic acid or hydrocaffeic acid gave *N*-catechoyl 3,4-dihydroxyphenylglycine amides. The use of *N*-benzyloxycarbonylglycine as acid component allowed the preparation of a 3,4-dihydroxyphenylglycyl dipeptide derivative. Radical-scavenging activity studies of the polyphenolic amino acid derivatives showed a sharp increase in activity with the increase in number of hydroxyl or catechol groups present. Cyclic voltammetry experiments established a correlation between oxidation peak potentials and the radical-scavenging activity.

1. Introduction

Phenolic amino acids represent a wide class of compounds endowed with interesting biological activities. The natural phenolic amino acid, tyrosine displays antioxidant properties,[1] performing vital antioxidant functions inside lipid bilayers and protecting cells from oxidative destruction.[2] Non-proteinogenic phenolic amino acids are crucial components of certain peptidic natural products such as the vancomycin group of antibiotics[3] (4-hydroxyphenylglycine) and of cell-penetrating peptides (CPPs).[4] CPPs are short, nontoxic peptides containing cationic and non-coded hydrophobic amino acids that can carry small molecules. They are able to cross, not only the cellular membrane, but also to target inside specific cellular organelles such as nucleus and mitochondria. Of the CPPs tested as mitochondria targeted antioxidants[5,] the Szeto-Schiller (SS) peptides are the most promising.[6] There are a number of surprising features about these peptides. Despite being water soluble, with a net charge of 3+, they are readily taken up by all cells via passive diffusion.[7-9] The presence of a D-amino acid and non-coded amino acids at the tips of the peptide renders them resistant to aminopeptidase activity. The mechanism behind their cell permeability is unclear, but the aromatic rings may serve as electron cages to shield the cationic charges via cation-π interaction. These SS peptides may be viewed as "cloaked" or "stealth" as they can evade cellular membranes, even penetrating cell barriers with tight junctions including the blood-brain barrier.[7,10] The antioxidant action of SS peptides in scavenging radical oxygen species and inhibiting low density lipoprotein oxidation, is attributed to the 2,5-dimethyl-L-tyrosine residue.

The *ortho*-dihydroxyaryl function present in catechols gives compounds with a broad scope of properties that confer important biochemical functions.[11] Such properties result from; the ability to establish reversible equilibria at moderate redox potentials and pHs; irreversible cross-linking through complex oxidation mechanisms; excellent chelating properties; interaction of the vicinal hydroxyl groups with surfaces of different nature. Despite these properties, little attention has been given to amino acids with the *ortho*-dihydroxyaryl function, such as 3,4-dihydroxyphenylglycine. This non-natural amino acid has been studied as copper ligand[12] and as a substrate for tyrosinase, being converted to 3,4-dihydroxybenzaldehyde via spontaneous decarboxylation of the enzymatically generated *ortho*-quinone.[13]

Amino acids coupled with phenolic or catecholic groups are bioactive substances involved in suppression of deleterious effects of oxidative stress[14-17] and have a wide range of biological activities such as anti-atherogenic,[18] anticancer[19] and antimicrobial.[20-23] Studies have confirmed that conjugation between different types of compounds such as

amino acids and phenolic acids is useful, not only to investigate structure activity relationships, but can also constitute a strategy to improve antioxidant efficiency and bioactivity.[24,25] Recently, we established an innovative synthetic strategy that allowed the preparation of a library of *N*-phenolic and *N*-catecholic dehydroamino acid derivatives.[26]

The development of synthetic strategies that allow the synthesis of new amino acids with phenolic or catecholic side chains and conjugates of these amino acid with phenolic or cathecolic groups would give rise to a new repertoire of interesting synthetic building blocks. These may have a wide application in the preparation, not only of specific cellular organelle targeted antioxidant peptides,[5] but can also have other intrinsic biological activities or broader applications, such as: cross-linkers with proteins or carbohydrates;[27] transition metal ligands;[28] or used in the design of new peptide hydrogels that mimic mussel adhesive proteins.[29-32]

2. Results and Discussion

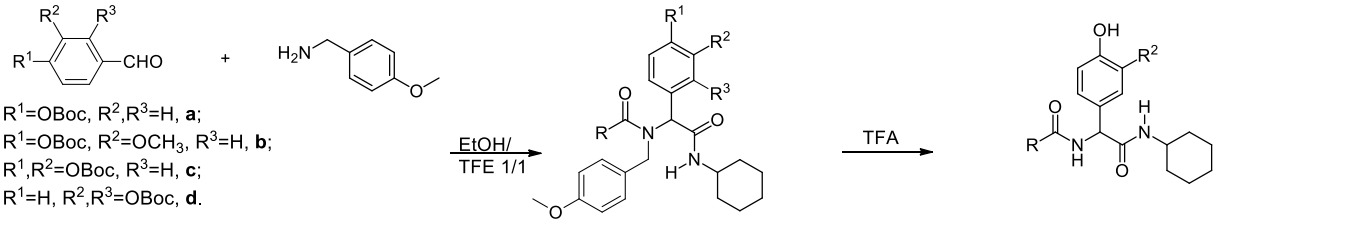
A simple approach to obtain new hydroxyphenylglycines and/or conjugates of hydroxyphenylglycines with phenolic or catecholic acids would be a strategy where simple natural phenols are joined together in a one-step economical way. One such multicomponent reaction is the Ugi reaction, first reported by Ivar Ugi.[33,34] It is classified as an isocyanide-based multicomponent reaction, consisting of the simultaneous joining of 4 different classes of compounds: an aldehyde; a carboxylic acid; an amine and an isocyanide.

Lambruschini et al.[35] using the Ugi reaction, were able to prepare a series of complex polyphenols containing two to four hydroxy-substituted aryl groups. This methodology, using hydroxyl substituted benzoic acids and benzaldehydes, gave hydroxylated bis-amides and their effect on quenching radicals and DNA oxidation was estimated.[36] However, derivatives with the *ortho*-catechol moiety as the amino acid side chain have not been report.

A strategy based on the modified Ugi reaction, using as amine component 4-methoxybenzylamine and as isocyanide the commercially available and relatively stable cyclohexyl isocyanide, followed by treatment of the adducts with trifluoroacetic acid (TFA) has previously been explored.[37,38] This procedure using as aldehyde component phenolic or catecholic benzyladehydes and as acid component catecholic acids, could be an efficient approach for the synthesis of novel *N*-(catechoyl)-hydroxy and 3,4-dihydroxy phenylglycines. Thus, a strategy based on the modified Ugi reaction using phenolic and/or catecholic benzaldehydes and carboxylic acid components was explored.

A severe limitation of the application of *ortho*-catechol moieties in synthesis is their high instability in basic environment. Thus, due to the basic nature of the amine component, protection of the hydroxyl function of the benzaldehyde would be required. *O-tert*-Butyloxycarbonylated benzaldehydes can be easily obtained by reaction of hydroxybenzaldehydes with *tert*-butyldicarbonate in the presence of dimethylaminopyridine as catalyst.[26] Furthermore, this temporary protection can be removed in the subsequent treatment with TFA, a reagent generally used in peptide synthesis to remove acid-labile protecting groups.[39,40]

A preliminary test was carried out using as reactants *tert*-butyl (4-formylphenyl) carbonate, 4-methoxybenzylamine, benzoic acid and cyclohexyl isocyanide in a 1/1 solution of ethanol and trifluorethanol (TFE) to give compound **1a'** in high yield (Scheme 1). The proton NMR spectrum of the compound gave the expected peaks with the diastereotopic CH_2 protons of the benzylamine group appearing as two signals at 4.41 ppm and 4.63 ppm. The Ugi adduct was treated with TFA at 80 °C for 10 min. Removal of TFA gave an oily residue that precipitated on addition of diethyl ether to give *N*-benzoyl, 4-hydroxyphenylglycine cyclohexylamide in good yield (compound **1a**, Scheme 1).



Scheme 1 Synthesis of Ugi adducts and treatment with trifluoroacetic acid to give *N*-acyl hydroxyphenylglycine cyclohexylamides.

Treatment of Ugi adduct **1a'** with TFA allowed the simultaneous removal of the methoxybenzyl and the *tert*-butyloxycarbonyl groups, without affecting the cyclohexylamide. This is in agreement with previous results

by Maia et al. in the synthesis of *N*-acyl-α,α-dialkylglycines by cleavage of Ugi adducts with neat TFA.[38] Treatment in these conditions of *N*-benzoyl-*N,α,α*-trialkylglycine amides formed by reaction of 4-methoxybenzylamine, benzoic acid, cyclohexyl isocyanide and the more bulky dibenzylketone, resulted in removal of the methoxybenzyl group without amide cleavage. The authors reported that in neat TFA the *N*-alkyl group of the bulkier *N*-benzoyl, *N*-(4-methoxybenzyl) *α,α*-dibenzylglycine derivatives cleaves faster than their amide bond. They concluded that for acidolytic cleavage of the C-terminal amide to occur, the presence of an alkyl/aryl substituent at the amino acid nitrogen atom is required. Thus, the phenyl group directly linked to the α-carbon in adduct **1a'** seems to exert a steric effect comparable to that reported for the α,α-dibenzyl moiety in *N*-acyl-*N,α,α*-trialkylglycine amides.

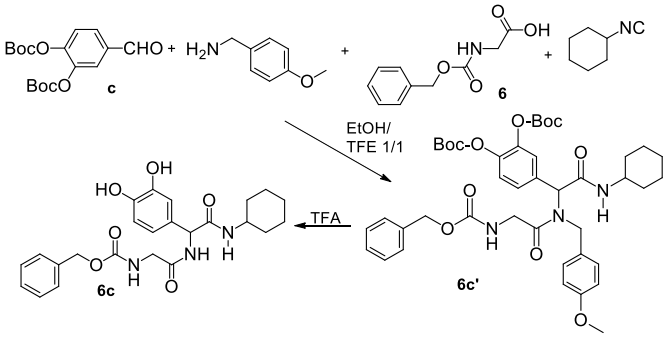
Compound **1a** could also be obtained in a one-pot procedure consisting of the condensation reaction, followed by evaporation of the solvent at reduced pressure and treatment of the residue with TFA (Scheme 1). *tert*-Butyl (4-formyl-2-methoxyphenyl) carbonate and di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate (Scheme 1, compounds **b** and **c**, respectively) were reacted in this one-pot procedure to give *N*-benzoyl, 4-hydroxy-3-methoxy and *N*-benzoyl, 3,4-dihydroxy phenylglycine cyclohexylamides in 76% and 66% yields, respectively (compounds **1b** and **1c**). An attempt to carry out the condensation reaction using the highly sterically hindered di-*tert*-butyl (3-formyl-1,2-phenylene) dicarbonate (compound **d**) and benzoic acid gave only trace amounts of the corresponding Ugi adduct (compound **1d'**).

The reaction was carried out substituting 2-naphthylacetic acid for benzoic acid and using di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate to give the *N*-(2-naphthyl)acetyl, 3,4-dihydroxyphenylglycine amide in 78% yield (compound **2c**, Scheme 1).

The results obtained led us to explore the possibility of using catecholic acids as acid component in the Ugi reaction, namely, protocatechuic and hydrocaffeic acid. These cathecolic acids were tested without hydroxyl protection since, when the acid component is added, all 4-methoxybenzylamine has been consumed. Thus, *tert*-butyl (4-formylphenyl) carbonate was reacted in the same conditions, substituting protocatechuic acid for benzoic acid to give the Ugi adduct **3a'** in quantitative yield (Scheme 1). Treatment of compound **3a'** with TFA gave the *N*-protocatechoyl 4-hydroxyphenylglycine derivative in 72% yield (compound **3a**). By reacting *tert*-butyl (4-formyl-2-methoxyphenyl) carbonate or di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate with protocatechuic acid, the corresponding *N*-protocatechoyl, hydroxyphenylglycine amides were obtained in good yields (compounds **3b** and **3c**, respectively). The reaction using hydrocaffeic acid and di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate gave compound **4c** in 74% yield. Thus, the use of di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate and protocatechuic or hydrocaffeic acid, allows the synthesis of amino acid derivatives baring two catecholic groups.

Synthesis of compound **4c** shows that the steric effect of the phenyl group linked to the α-carbon induces removal of the methoxybenzyl group without amide cleavage, even with the less sterically hindered hydrocaffeoyl substituent as *N*-acyl group. This was further confirmed by reacting *tert*-butyl (4-formylphenyl) carbonate with acetic acid as carboxylic component. Again, treatment of the Ugi adduct with neat TFA led to removal of the methoxybenzyl group without amide cleavage (compound **5a**).

The procedure was also carried out using as acid component a N-protected amino acid, namely, *N*-benzyloxycarbonylglycine. A Ugi adduct was obtained (compound **6c'**, Scheme 2) which, after treatment with TFA gave a 3,4-dihydroxyphenylglycyl dipeptide derivative in 74% yield (compound **6c**, Scheme 2). Thus, the methodology developed also allows the preparation of hydroxyphenylglycyl dipeptide derivatives.

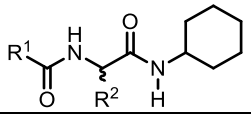


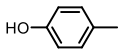
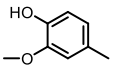
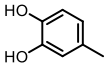
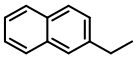
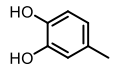
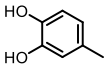
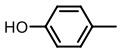
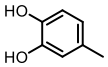
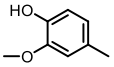
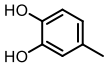
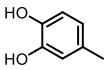
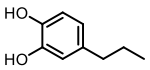
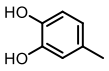
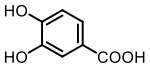
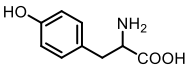
Scheme 2 Synthesis of a 3,4-dihydroxyphenylglycyl dipeptide derivative.

In order to establish the antioxidant capacity of the *N*-acyl hydroxyphenylglycines prepared, the radical-scavenging activity of selected compounds was determined using 1,1-diphenyl-2-picrylhydrazyl (DPPH•) as a stable radical.[41] The EC_{50} values (relative concentration of antioxidant required to lower the initial DPPH• concentration by 50%) were determined after several reaction times and compared to the values obtained for protocatechuic acid (Table 1). As expected, the radical-scavenging activity against the DPPH radical of the monophenol derivative **1a** was very weak. However, a significant increase in activity occurs when a methoxy group is added in *ortho* position to the hydroxyl group (compound **1b**) (a six-fold increase for EC_{50} determined after 5 minutes). The substitution of this methoxy group by another hydroxyl group (compound **1c**), increases more than ten times the radical-scavenging activity. A slightly higher activity is observed when (2-naphthyl)acetyl is substituted for benzoyl as *N*-acyl group (compound **2c**). The *N*-protocatechoyl 4-hydroxylphenylglycine derivatives (compound **3a**) showed radical-scavenging activity comparable to that of compound **1c**, which also has an *ortho*-dihydroxyaryl function as amino acid side chain. Introduction of a methoxy group in *ortho* position to the hydroxyl group (compound **3b**) causes a 25% increases in activity as determined after 5 minutes. Substitution of the methoxy group with a second hydroxyl group (compound **3c**) causes a further 30% increase in activity. The *N*-hydrocaffeoyl 3,4-dihydroxyphenylglycine derivative (compound **4c**) showed similar activity to the *N*-protocatechoyl derivative **3c**. Thus, the radical-scavenging activities of compounds **3c** and **4c** were almost double the activities of derivatives with a single catecholic group (compounds **1c** or **3a**).

These results show a direct correlation between radical-scavenging capacity against the DPPH radical and the number of hydroxyl or catechol groups present in the phenylglycine derivatives. A high increase in activity with the increase in number of hydroxyl or catechol groups is observed with the monocatecholic derivatives (compounds **1c** and **3a**) having twice the radical-scavenging activity of protocatechuic acid, while the *N*-catechoyl derivatives **3c** and **4c** have almost four times this activity. Thus, the introduction of the catecholic moiety in an amino acid structure induces a significant increase in its intrinsic radical scavenging activity.

Table 1 DPPH• Radical-scavenging capacity, EC50[a], oxidation peak potentials and Log *P*[b] of *N*-acyl 4-hydroxy, 4-hydroxy-3-methoxy and 3,4-dihydroxy phenylglycine cyclohexylamides and of protocatechuic acid and tyrosine.



Compound	R1	R2	EC_{50} 5 min	EC_{50} 60 min	$E_{p,a}$ (V *vs* Ag/AgCl)	Log *P*
1a			24.5 ± 13.8	11.0 ± 11.9	0.703 ± 0.016	3.25
1b			3.76 ± 0.20	2.08 ± 0.60	0.500 ± 0.006	3.06
1c			0.32 ± 0.01	0.22 ± 0.01	0.272 ± 0.006	2.76
2c			0.26 ± 0.01	0.22 ± 0.01	0.266 ± 0.005	4.03
3a			0.37 ± 0.01	0.23 ± 0.01	0.274 ± 0.003	2.28
3b			0.28 ± 0.01	0.19 ± 0.01	0.280 ± 0.006	2.10
3c			0.20 ± 0.01	0.13 ± 0.01	0.272 ± 0.015	1.79
4c			0.19 ± 0.01	0.16 ± 0.01	0.145 ± 0.008 0.233 ± 0.010	2.40
Protocatechuic acid			0.69 ± 0.03	0.31 ± 0.01	0.324 ± 0.005	0.88
Tyrosine			26.5 ± 11.70	nd	0,715± 0.018	-2.20

[a]The antiradical activity was defined as the relative concentration of antioxidant required to lower the initial DPPH• concentration by 50% [EC50 (M phenolic compound per unit DPPH concentration)] obtained at different reaction times.
[b]Partition coefficient between water and *n*-octanol. Estimated using Molinspiration Cheminformatics software (Molinspiration, Slovensky Grob, Slovak Republic, 2017, http://www.molinspiration.com).

Correlation between antioxidant profile and redox properties of low molecular weight antioxidants is well established.[42] Cyclic voltammetry has been used as an important tool for the evaluation of antioxidant capacity of this type of antioxidants. Compounds with lower oxidation potentials are more readily oxidized and usually show better antioxidant activities. Thus, oxidation peak potentials for the hydroxyphenylglycine derivatives were determined by cyclic voltammetry using a glassy carbon working electrode and measured *vs* a Ag/AgCl electrode (Table 1). Experiments were run using as supporting electrolyte a phosphate buffer at the physiological pH of 7.30.

Compound **1a**, which showed very weak radical-scavenging activity, presented the highest oxidation peak potential (0.703 V). A decrease in over 0.2 V in potential occurs when a methoxy group is added in *ortho* position to the hydroxyl group (compound **1b**) in agreement with a significant increase in radical-scavenging activity against the DPPH radical. The substitution of the methoxy group by a second hydroxyl group (compound **1c**) causes a further significant decrease in the oxidation potential (0.228 V), which also agrees with the ten-fold increase observed in radical-scavenging activity. Compounds with a single catecholic group (compounds **2c**, **3a** and **3b**) gave anodic peaks with similar potentials *ca.* 0.270 V, that corresponded to similar radical-scavenging activities. These potentials were lower than the peak potential for the catecholic reference protocatechuic acid (0.324 V). Compound **3c** baring two similar catecholic groups gave only one anodic peak at a similar potential to the other catecholic compounds (0.272 V). Thus, the increase in radical-scavenging activity of compound **3c**, when compared to the activities of the monocatecholic

compounds, must result from an additive effect of the two catechol groups.

On the other hand, two anodic peaks were observed for compound **4c**, one at 0.145 V and another at 0.233 V. The first peak probably results from oxidation of the *N*-hydrocaffeoyl group, since the potential of this peak was similar to that determined by us for octyl hydrocaffeate (0.145 V, data not shown). The potential of the second peak, although slightly lower, is in line with the peak potentials presented by the other glycine derivatives with an *ortho*-dihydroxyaryl side chain (compounds **1c**, **2c** and **3c**). Despite the lower oxidation potential of the first peak, compound **4c** did not present higher radical-scavenging activity against the DPPH radical than compound **3c**, which also has catecholic groups both as *N*-acyl and as side chain moiety.

These cyclic voltammetry experiments indicate a significant decrease in oxidation potential with the increase in number of hydroxyl groups. Thus, a correlation between oxidation potentials and radical-scavenging activity can be established, with lower oxidation potentials corresponding to higher radical-scavenging activities.

Despite the increase in the number of hydroxyl groups in these non-coded amino acid, their log *P* is significantly higher (Table 1) than that for tyrosine. This probably confers these new non-coded amino acids with better cell penetrating capacities.

3. Conclusions

A simple and efficient one-pot procedure for the synthesis of *N*-acyl hydroxyphenylglycine amides has been established. These novel hydroxyphenylglycine derivatives, in particular the polyphenolic amino acids *N*-protocatechoyl and *N*-hydrocaffeoyl 3,4-dihydroxyphenylglycine amides exhibited high radical-scavenging activity.

The synthesis of the *N*-benzyloxycarbonyl-3,4-dihydroxyphenylglycyl dipeptide derivative shows that the methodology developed gives rise to a substrate that, after N-acyl deprotection, can be coupled with other amino acid residues to give, among others, cell-penetrating type peptides that can be tested as mitochondria targeted antioxidants.

In addition, these non-coded amino acid derivatives can have other intrinsic biological activities or be applied as: cross-linkers; transition metal ligands; or used in the design of new peptide hydrogels.

4. Experimental Section

4.1. General methods

Melting points (°C) were determined in a Gallenkamp apparatus and are uncorrected. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker Avance II^{+} instrument. Spectra were taken at room temperature in $CDCl_3$, CD_3COCD_3 and CD_3OD at 400 MHz (^{1}H) and 100.6 MHz (^{13}C) by using TMS as an internal standard (^{1}H NMR in $CDCl_3$: 0.000 ppm). ^{1}H-^{1}H spin-spin decoupling, DEPT θ 45°, HMQC and HMBC were used to attribute some signals. Chemical shifts are given in ppm and coupling constants in Hz. HRMS data were recorded by the Laboratory for Structural Elucidation of the Materials Centre of the University of Porto on an LTQ Orbitrap^{TM} XL hybrid mass spectrometer (Thermo Fischer Scientific, Bremen, Germany) controlled by *LTQ Tune Plus 2.5.5* and *Xcalibur 2.1.0*. Elemental analysis was performed on a LECO CHNS 932 elemental analyzer. The reactions were monitored by thin layer chromatography (TLC). Column chromatography was performed on Macherey-Nagel silica gel 230-400 mesh. Petroleum ether refers to the boiling range 40-60 °C. Solvents were used without purification except for acetonitrile which was dried using standard procedures.

4.1.1. DPPH• Radical Essay

DPPH• radical-scavenging activity was assessed as described previously.[41,43] Methanolic DPPH• stock solution (1.93 mM) was diluted to give a 0.10 mM working solution. The reaction between DPPH• and each antioxidant was monitored at 515 nm by using a Powerwave XS Microplate Reader (Bio-Tek Instruments, Inc) thermostated at T = 25.0 ± 0.1 °C. The wells of a 96-well microplate contained a methanolic solution of the antioxidant (3-200 μM) and 80 μM DPPH•. The absorbance of each well was recorded at 1 min. intervals for a 60 min. period. The absorbance of each solution was subtracted from the blank (80 μM DPPH• without antioxidant). The antiradical activity was defined as the relative concentration of antioxidant required to lower the initial DPPH• concentration by 50% [EC_{50} (mol/L phenolic compound per unit DPPH concentration)].

4.1.2. Cyclic voltammetry

The cyclic voltammetry experiments were carried out using a Hi-Tek potentiostat, type DT 2101, and a Hi-Tek wave generator type PPRl, connected to a Philips recorder, type PM 8043, and to a three electrode, home-built glass cell. The working electrode was a vitreous carbon disc (diameter: 3 mm), the counter electrode a platinum spiral and the reference electrode a mercury pool. The supporting electrolyte was a phosphate buffer (0.10 M) with pH adjusted to 7.30. Stock solutions of each compound (0.01 M) were prepared by dissolving an appropriate amount in ethanol. The voltammetric working solutions were prepared in the electrochemical cell by diluting 0.10 mL of the stock solution in 10 mL of supporting electrolyte in order to obtain a final concentration of 0.0001 M. At the end of each experiment the potential of the mercury pool was measured *vs* a Ag/AgCl electrode. Between experiments the working electrode was repolished with alumina powder (~0.05 μm).

4.1.3. Statistical analysis

All the DPPH• radical scavenging assays and cyclic voltammetry experiments were run at least in quadruplicate. SPSS 21.0 software was used for statistical analysis by one-way analysis of variance (ANOVA, with Tukey's HSD multiple comparison) with the level of

significance set at $P < 0.05$. Data are presented as means ± standard deviation.

4.2. Synthesis

Procedure A: Synthesis of *O*-*tert*-butyloxycarbonylated benzaldehydes. To a solution of the hydroxybenzaldehyde (5.00 mmol) in dry acetonitrile (0.20 M), 0.10 equiv of dimethylaminopyridine was added. This was followed by addition under rapid stirring at room temperature of 1.10 equiv of *tert*-butyldicarbonate for the monohydroxylated benzaldehydes and 2.20 equiv for the dihydroxylated benzaldehyde. When all the reactant had been fully *tert*-butyloxycarbonylated the solvent was evaporated at reduced pressure and the residue was dissolved in ethyl acetate (100 mL) and washed with $KHSO_4$ (1 M) and brine (3 times 25 mL each). The organic layer was dried with $MgSO_4$ and the solvent evaporated at reduced pressure. The product obtained was then recrystallized or subject to column chromatography.

***tert*-Butyl (4-formylphenyl) carbonate** (**a**).[44] Procedure A using 4-hydroxybenzaldehyde (0.611 g, 5.000 mmol) was followed to give **a**.
Yield: 1.066 g (96%); white solid (from diethyl ether/*n*-hexane). The spectroscopic properties were in agreement with those previously reported.[44]

***tert*-Butyl (4-formyl-2-methoxyphenyl) carbonate** (**b**).[45] Procedure A using 4-hydroxy-3-methoxybenzaldehyde (0.761 g, 5.000 mmol) was followed to give **b**.
Yield: 1.197 g (95%); white solid (from diethyl ether/*n*-hexane). The spectroscopic properties were in agreement with those previously reported.[45]

di-*tert*-Butyl (4-formyl-1,2-phenylene) dicarbonate (**c**).[46] Procedure A using 3,4-dihydroxybenzaldehyde (0.691 g, 5.000 mmol) was followed to give **c**.
Yield: 1.575 g (93%); light brown oil. The spectroscopic properties were in agreement with those previously reported.[46]

di-*tert*-Butyl (3-formyl-1,2-phenylene) dicarbonate (**d**). Procedure A using 2,3-dihydroxybenzaldehyde (0.691 g, 5.000 mmol) was followed to give **d**.
Yield: 1.622 g (96%); light brown solid.
R_f = 0.43 (petroleum ether/diethyl ether 1:1).
M.p. 58-59 °C.
^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.56 [s, 18H, $2C(CH_3)_3$], 7.36-7.40 (m, 1H, ArH), 7.53 (dd, *J* = 8.0 Hz, *J* = 1.6 Hz, 1H, ArH), 7.75 (dd, *J* = 8.0 Hz, *J* = 1.6 Hz, 1H, ArH), 10.19 (s, 1H, CHO) ppm.
^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 27.11 [$C(CH_3)_3$], 27.14 [$C(CH_3)_3$], 83.94 [$C(CH_3)_3$], 84.37 [$C(CH_3)_3$], 126.07 (CH), 126.39 (CH), 128.53 (CH), 129.35 (C), 143.08 (C), 143.74 (C) 149.67 (C=O), 149.99 (C=O), 187.27 (CH=O) ppm.
HRMS (ESI): m/z M^+ calcd for $C_{17}H_{22}O_7$: 338.1366; found: 338.1597.

Procedure B: Two step synthesis of *N*-acyl, hydroxyphenylglycine cyclohexylamides. To a solution of the *O*-*tert*-butyloxycarbonylated benzaldehyde (1.00 mmol) in ethanol/2,2,2-trifluoroethanol (1/1) (0.17 M) under a stream of nitrogen, 1.10 equiv of 4-methoxybenzylamine was added. After 4 hours, 1.10 equiv of the carboxylic acid and 1.10 equiv of cyclohexyl isocyanide were added and left to react for 2 days. The reaction mixture was then evaporated at reduced pressure and the residue was dissolved in ethyl acetate (100 mL) and washed with $KHSO_4$ (1 M), $NaHCO_3$ (1 M) and brine (3 times 25 mL each). The organic layer was dried with $MgSO_4$ and the solvent evaporated at reduced pressure to give the corresponding Ugi adduct. Trifluoroacetic acid was added to the adduct (0.25 M) and the solution refluxed at 80 °C for 10 min. TFA was then evaporated at reduced pressure. The product obtained was then recrystallized or subject to column chromatography.

***N*-Benzoyl, (4-hydroxyphenyl)glycine cyclohexylamide** (**1a**). Procedure B using *tert*-butyl (4-formylphenyl) carbonate (0.222 g, 1.000 mmol) and benzoic acid was followed to give **1a'**.
Yield: 0.537 g (94%); white solid (from diethyl ether).
R_f = 0.68 (petroleum ether/diethyl ether 1:5).
M.p. 65-66 °C.
^{1}H NMR (400 MHz, $CDCl_3$): δ = 0.86-1.93 (m, 10H, CH_2 cyclohexyl), 1.57 [s, 9H, $C(CH_3)_3$], 3.76 (s, 3H, OCH_3), 3.79-3.82 (m, 1H, CH cyclohexyl), 4.41 (br. s, 1H, $C_6H_4CH_2$), 4.63 (br. s, 1H, $C_6H_4CH_2$), 5.40 (br. s, 1H, αCH), 5.74 (br. s, 1H, NH), 6.74 (d, *J* = 8.4 Hz, 2H, ArH), 7.00 (br. d, *J* = 4.8 Hz, 2H, ArH), 7.13 (d, *J* = 8.8 Hz, 2H, ArH), 7.35-7.37 (m, 4H, ArH), 7.46-7.49 (m, 3H, ArH) ppm.
^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 24.70 (CH_2), 24.78 (CH_2), 25.44 (CH_2), 27.66 [$C(CH_3)_3$], 32.69 ($2CH_2$), 48.64 (CH cyclohexyl), 55.20 (OCH_3), 64.08 (αCH), 65.82 ($C_6H_4CH_2$), 83.80 [$C(CH_3)_3$], 113.87 (3CH), 121.61 (3CH), 126.68 (CH), 128.38 (CH), 128.49 (CH), 129.84 (CH), 130.07 (CH), 130.70 (CH), 132.65 (2C), 133.35 (CH), 136.08 (C), 151.08 (C), 151.54 (C), 158.76 (C=O), 168.04 (C=O), 173.07 (C=O) ppm.
Anal. calcd. for $C_{34}H_{40}N_2O_6$ (572.69): C 71.31, H 7.04, N 4.89; found C 71.39, H 7.32, H 4.51.
Trifluoroacetic acid was added to **1a'** (0.307 g, 0.537 mmol) (0.25 M) and the solution refluxed at 80 °C for 10 min. TFA was then evaporated at reduced pressure to give **1a**.
Yield: 0.118 g (63%); white solid (from diethyl ether).
R_f = 0.48 (petroleum ether/diethyl ether 1:5).
M.p. 141-142 °C.
^{1}H NMR (400 MHz, CD_3COCD_3): δ = 1.14-1.36 (m, 5H, CH_2 cyclohexyl), 1.57-2.12 (m, 5H, CH_2 cyclohexyl), 3.68-3.76 (m, 1H, CH cyclohexyl), 5.64 (s, 1H, αCH), 6.82 (d, *J* = 8.4 Hz, 2H, ArH), 7.37 (d, *J* = 8.4 Hz, 2H, ArH), 7.46-7.50 (m, 2H, ArH), 7.53-7.57 (m, 1H, ArH), 7.93-7.95 (m, 2H, ArH) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 25.48 (CH_2), 25.57 (CH_2), 26.22 (CH_2), 33.21 (CH_2), 33.30 (CH_2), 49.10 (CH cyclohexyl), 57.27 (αCH), 115.91 (2CH), 128.08 (2CH), 129.17 (2CH), 129.58 (2H), 131.04 (C), 132.13 (CH), 135.40 (C), 157.83 (C), 166.36 (C=O), 170.12 (C=O) ppm.

HRMS (ESI): m/z [M + H]$^+$ calcd for $C_{21}H_{25}N_2O_3$: 353.1865; found: 353.1856.

Tentative synthesis of *N*-benzoyl, (2,3-dihydroxyphenyl)glycine cyclohexylamide (**1d**). Procedure B using di-*tert*-butyl (3-formyl-1,2-phenylene) dicarbonate (0.338 g, 1.000 mmol) and benzoic acid was followed. The residue obtained was chromatographed through silica using as eluent ethyl acetate/petroleum ether 1:3 to give the Ugi adduct **1d'** in low yield.
Yield: 0.116 g (17%); light yellow oil.
R_f = 0.64 (petroleum ether/diethyl ether 1:5).
^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.15-1.96 (m, 10H, CH_2 cyclohexyl), 1.46, 1.57, 1.76 [s, 18H, $C(CH_3)_3$], 3.79 (br. s, 4H, OCH_3 + CH cyclohexyl), 4.25 (d, *J* = 4.8 Hz, 2H, $C_6H_4CH_2$), 4.80 (br. s, 1H, αCH), 5.92 (br. d, *J* = 7.6 Hz, 1H, NH), 6.86 (d, *J* = 8.8 Hz, 2H, ArH), 6.94 (t, *J* = 8.0 Hz, 1H, ArH), 7.14 (dd, *J* = 8.0 Hz, *J* = 1.6 Hz, 1H, ArH), 7.18-7.22 (m, 3H, ArH), 7.47 (t, *J* = 8.0 Hz, 2H, ArH), 7.59 (t, *J* = 7.2 Hz, 1H, ArH), 8.01 (dd, *J* = 8.0 Hz, *J* = 1.2 Hz, 2H, ArH) ppm.
^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 24.59 (CH_2), 24.69 (CH_2), 25.47 (CH_2), 27.57 [$C(CH_3)_3$], 28.38 [$C(CH_3)_3$], 32.91 ($2CH_2$), 48.06 (CH cyclohexyl), 55.26 (OCH_3), 58.43 ($C_6H_4CH_2$), 82.02 [$2C(CH_3)_3$], 113.96 (2CH), 119.46 (CH), 120.20 (CH), 120.56 (C), 121.56 (CH), 124.29 (CH), 128.49 (2CH), 128.79 (CH), 129.49 (2CH), 129.62 (C), 130.51 (C), 130.87 (CH), 130.94 (C), 130.97 (C), 133.18 (CH), 144.88 (C), 158.89 (2C=O), 168.82 (C=O), 172.00 (C=O) ppm.
HRMS (ESI): m/z M$^+$ calcd for $C_{39}H_{48}N_2O_9$: 688.3360; found: 688.3968.

***N*-protocatechoyl, (4-hydroxyphenyl)glycine cyclohexylamide** (**3a**). Procedure B using *tert*-butyl (4-formylphenyl) carbonate (0.444 g, 2.000 mmol) and protocatechuic acid was followed to give **3a'**.
Yield: 1.208 g (quantitative); colourless oil.
R_f = 0.31 (petroleum ether/diethyl ether 1:5).
M.p. 119-120 °C.
^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.03-1.86 (m, 10H, CH_2 cyclohexyl), 1.57 [s, 9H, $C(CH_3)_3$], 3.72-3.75 (m, 4H, OCH_3 + CH cyclohexyl), 4.39 (br. s, 1H, $C_6H_4CH_2$), 4.76 (br. s, 1H, $C_6H_4CH_2$), 5.16 (br. s, 1H, αCH), 5.81 (br. s, 1H, NH), 6.72-6.79 (m, 3H, ArH), 6.89 (br. d, *J* = 8.4 Hz, 1H, ArH), 7.00-7.02 (m, 2H, ArH), 7.10-7.12 (m, 3H, ArH), 7.32 (br. d, *J* = 8.4 Hz, 2H, ArH) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 24.60 (CH_2), 24.66 (CH_2), 25.32 (CH_2), 27.67 [$C(CH_3)_3$], 32.46 ($2CH_2$), 48.94 (CH cyclohexyl), 55.21 (OCH_3), 60.40 (αCH), 83.89 [$C(CH_3)_3$], 113.96 (3CH), 114.86 (CH), 115.05 (CH), 119.67 (CH), 121.70 (3CH), 126.75 (C), 128.68 (CH), 130.03 (C), 130.60 (CH), 132.22 (C), 144.34 (C), 147.07 (C), 151.18 (C), 151.55 (C), 158.87 (C=O), 168.84 (C=O), 171.18 (C=O) ppm.
HRMS (ESI): m/z [M + H]$^+$ calcd for $C_{34}H_{41}N_2O_8$: 605.2863; found: 605.2856.
Trifluoroacetic acid was added to **3a'** (0.453 g, 0.750 mmol) (0.25 M) and the solution refluxed at 80 °C for 10 min. TFA was then evaporated at reduced pressure. The residue was dissolved in ethyl acetate (100 mL) and washed with $KHSO_4$ (1 M), $NaHCO_3$ (1 M) and brine (3 times 25 mL each). The organic layer was dried with $MgSO_4$ and the solvent evaporated at reduced pressure to give **3a**.
Yield: 0.207 g (72%); white solid (from ethyl acetate/dichloromethane).
R_f = 0.90 (petroleum ether/ethyl acetate 1:4).
M.p. 158-159 °C.
^{1}H NMR (400 MHz, CD_3COCD_3): δ = 1.11-1.39 (m, 5H, CH_2 cyclohexyl), 1.57-1.78 (m, 4H, CH_2 cyclohexyl), 1.87-1.90 (m, 1H, CH_2 cyclohexyl), 3.69-3.75 (m, 1H, CH cyclohexyl), 5.60 (d, *J* = 7.6 Hz, 1H, αCH), 6.80-6.82 (m, 2H, ArH), 6.89 (d, *J* = 8.0 Hz, 1H, ArH), 7.34-7.36 (m, 3H, ArH), 7.46 (d, *J* = 2.0 Hz, 1H, ArH), 7.75 (d, *J* = 7.6 Hz, 1H, NH), 8.42 (br. s, 3H, OH) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 25.48 (CH_2), 25.56 (CH_2), 26.21 (CH_2), 33.22 (CH_2), 33.29 (CH_2), 49.21 (CH cyclohexyl), 57.29 (αCH), 115.50 (CH), 115.61 (CH), 115.92 (2CH), 120.38 (CH), 127.16 (C), 129.53 (2CH), 131.30 (C), 145.60 (C), 149.20 (C), 157.79 (C), 166.23 (C=O), 170.41 (C=O) ppm.
HRMS (ESI): m/z [M + H]$^+$ calcd for $C_{21}H_{25}N_2O_5$: 385.1764; found: 385.1698.

***N*-Acetyl, (4-hydroxyphenyl)glycine cyclohexylamide** (**5a**). Procedure B using *tert*-butyl (4-formylphenyl) carbonate (0.222 g, 1.000 mmol) and acetic acid was followed to give **5a'**.
Yield: 0.510 g (quantitative); light yellow oil.
R_f = 0.31 (petroleum ether/diethyl ether 1:5).
^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.01-1.15 (m, 3H, CH_2 cyclohexyl), 1.20-1.48 (m, 3H, CH_2 cyclohexyl), 1.54 [s, 9H, $C(CH_3)_3$], 1.56-1.69 (m, 2H, CH_2 cyclohexyl), 1.81-1.93 (m, 2H, CH_2 cyclohexyl), 2.08 (s, 3H, CH_3CO), 3.68-3.78 (m, 4H, NHCH + OCH_3), 4.46 (d, *J* = 17.2 Hz, 1H, NCH_2), 4.65 (d, *J* = 17.2 Hz, 1H, NCH_2), 5.83 (br. s, 2H, αCH + NH), 6.74 (d, *J* = 8.4 Hz, 2H, ArH), 6.91 (d, *J* = 8.4 Hz, 2H, ArH), 7.08 (d, *J* = 8.4 Hz, 2H, ArH), 7.36 (d, *J* = 8.4 Hz, 2H, ArH) ppm.
^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 22.36 (CH_3), 24.65 (CH_2), 24.71 (CH_2), 25.36 (CH_2), 27.58 [$C(CH_3)_3$], 32.61 ($2CH_2$), 48.60 (CH cyclohexyl), 50.35 ($C_6H_4CH_2$), 55.13 (OCH_3), 62.04 (αCH), 83.73 [$C(CH_3)_3$], 113.83 (2CH), 121.44 (2CH), 127.32 (2CH), 129.08 (C), 130.70 (2CH), 132.68 (C), 150.98 (C), 151.47 (C), 158.56 (C=O), 168.43 (C=O), 172.63 (C=O) ppm.
HRMS (ESI): m/z [M + H]$^+$ calcd for $C_{29}H_{39}N_2O_6$: 511.2808; found: 511.2796.
Trifluoroacetic acid was added to **5a'** (0.255 g, 0.500 mmol) (0.25 M) and the solution refluxed at 80 °C for 10 min. TFA was then evaporated at reduced pressure to give **5a.**
White solid (from acetone).
R_f = 0.78 (petroleum ether/ethyl acetate 1:4).
M.p. 241-242 °C.
^{1}H NMR (400 MHz, DMSO): δ = 1.16-1.24 (m, 5H, CH_2 cyclohexyl), 1.49-1.75 (m, 5H, CH_2 cyclohexyl), 1.85 (s, 3H, CH_3CO), 3.40-3.45 (m, 1H, NHCH), 5.31 (d, *J* = 8.4 Hz, 1H, αCH), 6.66 (d, *J* = 8.4 Hz, 2H, ArH), 7.15 (d, *J* =

8.4 Hz, 2H, ArH), 7.99 (d, *J* = 8.0 Hz, 1H, NH), 8.29 (d, *J* = 8.4 Hz, 1H, NH), 9.34 (s, 1H, OH) ppm.
^{13}C NMR (100.6 MHz, DMSO): δ = 22.46 (CH_3), 24.37 (CH_2), 24.49 (CH_2), 25.17 (CH_2), 32.18 (CH_2), 32.30 (CH_2), 47.52 (CH cyclohexyl), 55.38 (αCH), 114.88 (2CH), 128.11 (2CH), 129.62 (C), 156.59 (C), 168.69 (C=O), 169.29 (C=O) ppm.
HRMS (ESI): m/z [M + H]$^+$ calcd for $C_{16}H_{23}N_2O_3$: 291.1709; found: 291.1705.

***N*-Benzyloxycarbonylglycyl, (3,4-dihydroxyphenyl)glycine cyclohexylamide (6c)**. Procedure B using di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate (0.338 g, 1.000 mmol) and *N*-benzyloxycarbonylglycine was followed to give **6c'**.
Yield: 0.731 g (94%); light pink solid (from ethyl acetate/petroleum ether).
R_f = 0.60 (petroleum ether/diethyl ether 1:5).
M.p. 113-114 °C.
^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.10-1.12 (m, 3H, CH_2 cyclohexyl), 1.31-1.34 (m, 3H, CH_2 cyclohexyl), 1.56 [s, 18H, 2C(CH_3)$_3$], 1.65-1.70 (m, 2H, CH_2 cyclohexyl), 1.84-1.88 (m, 2H, CH_2 cyclohexyl), 3.74-3.80 (m, 4H, OCH_3 + CH cyclohexyl), 3.93 (br. d, *J* = 16.8 Hz, 1H, αCH_2), 4.10 (br. d, *J* = 16.8 Hz, 1H, αCH_2), 4.43 (br. d, *J* = 17.2 Hz, 1H, $C_6H_4CH_2$), 4.60 (br. d, *J* = 17.2 Hz, 1H, $C_6H_4CH_2$), 5.10 (s, 2H, CH_2 Z), 5.66 (d, *J* = 7.6 Hz, 1H, αCH), 5.71-5.73 (m, 2H, 2NH), 6.75 (d, *J* = 8.4 Hz, 2H, ArH), 6.94 (d, *J* = 8.4 Hz, 2H, ArH), 7.15 (s, 2H, ArH), 7.30-7.35 (m, 6H, ArH) ppm.
^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 24.73 (CH_2), 24.81 (CH_2), 25.40 (CH_2), 27.58 [2C(CH_3)$_3$], 29.63 (CH_2), 31.90 (CH_2), 32.65 (CH_2), 43.46 (αCH_2), 48.91 (CH cyclohexyl), 49.27 ($C_6H_4CH_2$), 55.17 (OCH_3), 62.75 (αCH), 66.85 (CH_2 Z), 84.03 [2*C*(CH_3)$_3$], 114.17 (CH), 123.37 (CH), 125.00 (CH), 127.69 (CH), 127.95 (2CH), 128.03 (2CH), 128.46 (3CH), 133.04 (C), 136.39 (C), 142.59 (C), 142.75 (C), 150.39 (C), 150.43 (C), 156.14 (C=O), 158.91 (2C=O), 167.37 (C=O), 170.12 (C=O) ppm.
HRMS (ESI): m/z [M + H]$^+$ calcd for $C_{42}H_{54}N_3O_{11}$: 776.3758; found: 776.3768.
Trifluoroacetic acid was added to **6c'** (0.583 g, 0.750 mmol) (0.25 M) and the solution refluxed at 80 °C for 10 min. TFA was then evaporated at reduced pressure to give **6c.**
Yield: 0.252 g (74%); white solid (from ethyl acetate/diethyl ether).
R_f = 0.82 (petroleum ether/ethyl acetate 1:4).
M.p. 162-163 °C.
^{1}H NMR (400 MHz, CD_3COCD_3): δ = 1.11-1.71 (m, 10H, CH_2 cyclohexyl), 3.63-3.67 (m, 1H, CH cyclohexyl), 3.87 (d, *J* = 6.0 Hz, 2H, αCH_2), 5.10 (s, 2H, CH_2 Z), 5.32 (d, *J* = 7.6 Hz, 1H, αCH), 6.70 (br. d, *J* = 7.6 Hz, 1H, NH), 6.73 (s, 2H, ArH), 6.91 (s, 1H, ArH), 7.26-7.39 (m, 6H, ArH + NH), 7.66 (d, *J* = 7.2 Hz, 1H, NH) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 25.62 (CH_2), 26.27 (CH_2), 29.26 (CH_2), 33.26 (CH_2), 33.38 (CH_2), 45.01 (αCH_2), 49.20 (CH cyclohexyl), 57.16 (αCH), 66.93 (CH_2 Z), 115.45 (CH), 115.90 (CH), 119.80 (CH), 128.63 (2CH), 129.24 (3CH), 131.60 (C), 138.16 (C), 145.66 (C), 145.78 (C), 157.63 (C=O), 169.13 (C=O), 170.00 (C=O) ppm.
HRMS (ESI): m/z [M + H]$^+$ calcd for $C_{24}H_{30}N_3O_6$: 456.2135; found: 456.2141.

Procedure C: One-pot synthesis of *N*-acyl hydroxyphenylglycine cyclohexylamides. To a solution of the *O-tert*-butyloxycarbonylated benzaldehyde (1.00 mmol) in ethanol/2,2,2-trifluoroethanol (1/1) (0.17 M), 1.10 equiv of 4-methoxybenzylamine was added under a stream of nitrogen. After 4 hours, 1.10 equiv of the carboxylic acid and 1.10 equiv of cyclohexyl isocyanide were added and left to react for 2 days. The reaction mixture was then evaporated at reduced pressure and trifluoroacetic acid added to the residue (0.25 M). The solution was refluxed at 80 °C for 10 min. after which TFA was evaporated at reduced pressure. The product obtained was then recrystallized or subject to column chromatography

***N*-Benzoyl, (4-hydroxyphenyl)glycine cyclohexylamide (1a)**. Procedure C using *tert*-butyl (4-formylphenyl) carbonate (0.222 g, 1.000 mmol) and benzoic acid was followed to give **1a.**
Yield: 0.221 g (63%).

***N*-Benzoyl, (4-hydroxy-3-methoxyphenyl)glycine cyclohexylamide (1b)**. Procedure C using *tert*-butyl (4-formyl-2-methoxyphenyl) carbonate (0.252 g, 1.000 mmol) and benzoic acid was followed to give **1b**.
Yield: 0.291 g (76%); light yellow solid (from diethyl ether).
R_f = 0.31 (petroleum ether/diethyl ether 1:5).
M.p. 169-170 °C.
^{1}H NMR (400 MHz, CD_3COCD_3): δ = 1.15-1.36 (m, 5H, CH_2 cyclohexyl), 1.57-1.91 (m, 5H, CH_2 cyclohexyl), 3.69-3.75 (m, 1H, CH cyclohexyl), 3.83 (s, 3H, OCH_3), 5.67 (s, 1H, αCH), 6.81 (d, *J* = 8.0 Hz, 1H, ArH), 7.01 (dd, *J* = 8.0 Hz, *J* = 2.0 Hz, 1H, ArH), 7.19 (d, *J* = 2.0 Hz, 1H, ArH), 7.46-7.57 (m, 3H, ArH), 7.94-7.96 (m, 2H, ArH), 8.04 (s, 1H, NH) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 25.48 (CH_2), 25.56 (CH_2), 26.22 (CH_2), 33.21 (CH_2), 33.29 (CH_2), 49.09 (CH cyclohexyl), 56.23 (CH_3), 57.60 (αCH), 111.94 (CH), 115.53 (CH), 121.15 (CH), 128.12 (2CH), 129.16 (2CH), 131.49 (C), 132.13 (CH), 135.43 (C), 147.05 (C), 148.17 (C), 166.46 (C=O), 170.07 (C=O) ppm.
HRMS (ESI): m/z [M + H]$^+$ calcd for $C_{22}H_{27}N_2O_4$: 383.1971; found: 383.1973.

***N*-Benzoyl, (3,4-dihydroxyphenyl)glycine cyclohexylamide (1c)**. Procedure C using di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate (0.338 g, 1.000 mmol) and benzoic acid was followed. The residue obtained after removal of TFA was chromatographed through silica using as eluents from ethyl acetate/petroleum ether 1:3 to neat ethyl acetate to give **1c**.
Yield: 0.241 g (66%); white solid.
R_f = 0.88 (petroleum ether/ethyl acetate 1:4).
M.p. 127-128 °C.

^{1}H NMR (400 MHz, CD_3COCD_3): δ = 1.28-1.36 (m, 5H, CH_2 cyclohexyl), 1.57-1.92 (m, 5H, CH_2 cyclohexyl), 3.69-3.77 (m, 1H, CH cyclohexyl), 5.59 (d, *J* = 7.6 Hz, 1H, αCH), 6.80 (d, *J* = 8.4 Hz, 1H, ArH), 6.88 (dd, *J* = 8.4 Hz, *J* = 2.0 Hz, 1H, ArH), 7.04 (d, *J* = 2.0 Hz, 1H, ArH), 7.33 (d, *J* = 7.6 Hz, 1H, $C_6H_5CON\mathit{H}$), 7.46-7.55 (m, 3H, ArH), 7.90-7.97 (m, 5H, 2ArH + 2OH + $N\mathit{H}C_6H_{11}$) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 25.49 (CH_2), 25.57 (CH_2), 26.22 (CH_2), 33.24 (CH_2), 33.32 (CH_2), 49.23 (CH cyclohexyl), 57.53 (αCH), 115.58 (CH), 115.91 (CH), 119.97 (CH), 128.07 (2CH), 129.17 (2CH), 131.73 (C), 132.12 (CH), 135.42 (C), 145.66 (C), 145.77 (C), 166.39 (C=O), 170.17 (C=O) ppm.
HRMS (ESI): m/z $[M + H]^+$ calcd for $C_{21}H_{25}N_2O_4$: 369.1814; found: 369.1811.

***N*-(2-Naphthyl)acetyl, (3,4-dihydroxyphenyl)glycine cyclohexylamide** (**2c**). Procedure C using di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate (0.338 g, 1.000 mmol) and 2-naphthylacetic acid was followed. The residue obtained after removal of TFA was dissolved in ethyl acetate (100 mL) and washed with $KHSO_4$ (1 M), $NaHCO_3$ (1 M) and brine (3 times 25 mL each). The organic layer was dried with $MgSO_4$ and the solvent evaporated at reduced pressure to give **2c**.
Yield: 0.338 g (78%); light yellow solid (from ethyl acetate/dichloromethane).
R_f = 0.75 (petroleum ether/ethyl acetate 1:4).
M.p. 220-221 °C.
^{1}H NMR (400 MHz, CD_3COCD_3): δ = 1.06-1.33 (m, 5H, CH_2 cyclohexyl), 1.63-1.83 (m, 5H, CH_2 cyclohexyl), 3.59-3.66 (m, 1H, CH cyclohexyl), 3.81 [d, *J* = 2.8 Hz, CH_2 (2-naphthyl)acetyl], 5.38 (d, *J* = 8.0 Hz, 1H, αCH), 6.75 (s, 2H, ArH), 6.95 (s, 1H, ArH), 7.17 (d, *J* = 8.0 Hz, 1H, $C_6H_3CON\mathit{H}$), 7.46-7.53 (m, 3H, ArH), 7.70 (d, *J* = 7.6 Hz, 1H, $N\mathit{H}C_6H_{11}$), 7.84-7.89 (m, 6H, ArH + 2OH) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 25.42 (CH_2), 25.52 (CH_2), 26.18 (CH_2), 33.19 (CH_2), 33.28 (CH_2), 43.57 [CH_2 (2-naphthyl)acetyl], 49.01 (CH cyclohexyl), 57.28 (αCH), 115.46 (CH), 115.84 (CH), 119.79 (CH), 126.32 (CH), 126.80 (CH), 128.40 (2CH), 128.54 (CH), 128.59 (CH), 128.65 (CH), 131.80 (C), 133.29 (C), 134.49 (C), 134.85 (C), 145.57 (C), 145.74 (C), 170.04 (C=O), 170.25 (C=O) ppm.
HRMS (ESI): m/z $[M + H]^+$ calcd for $C_{26}H_{29}N_2O_4$: 433.2127; found: 433.2131.

***N*-Protocatechoyl, (4-hydroxyphenyl)glycine cyclohexylamide** (**3a**). Procedure C using *tert*-butyl (4-formylphenyl) carbonate (0.222 g, 1.000 mmol) and benzoic acid was followed. The residue obtained after removal of TFA was dissolved in ethyl acetate (100 mL) and washed with $KHSO_4$ (1 M), $NaHCO_3$ (1 M) and brine (3 times 25 mL each). The organic layer was dried with $MgSO_4$ and the solvent evaporated at reduced pressure to give **3a**.
Yield: 0.279 g (72%).

***N*-Protocatechoyl, (4-hydroxy-3-methoxyphenyl) glycine cyclohexylamide** (**3b**). Procedure C using *tert*-butyl (4-formyl-2-methoxyphenyl) carbonate (0.252 g, 1.000 mmol) and protocatechuic acid was followed. The residue obtained after removal of TFA was dissolved in ethyl acetate (100 mL) and washed with $KHSO_4$ (1 M), $NaHCO_3$ (1 M) and brine (3 times 25 mL each). The organic layer was dried with $MgSO_4$ and the solvent evaporated at reduced pressure to give **3b**.
Yield: 0.283 g (68%); white solid (from ethyl acetate/dichloromethane).
R_f = 0.47 (petroleum ether/ethyl acetate 1:4).
M.p. 129-130 °C.
^{1}H NMR (400 MHz, CD_3COCD_3): δ = 1.17-1.35 (m, 5H, CH_2 cyclohexyl), 1.58-1.89 (m, 5H, CH_2 cyclohexyl), 3.68-3.73 (m, 1H, CH cyclohexyl), 3.81 (s, 3H, OCH_3), 5.63 (d, *J* = 7.2 Hz, 1H, αCH), 6.80 (d, *J* = 8.0 Hz, 1H, ArH), 6.89 (d, *J* = 8.4 Hz, 1H, ArH), 6.99 (dd, *J* = 8.4 Hz, *J* = 2.0 Hz, 1H, ArH), 7.17 (d, *J* = 2.0 Hz, 1H, ArH), 7.36 (dd, *J* = 8.4 Hz, *J* = 2.0 Hz, 1H, ArH), 7.40 (br. s., 1H, NH), 7.48 (d, *J* = 2.0 Hz, 1H, ArH), 7.81 (d, *J* = 7.2 Hz, 1H, NH) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 25.48 (CH_2), 25.56 (CH_2), 26.21 (CH_2), 33.24 (CH_2), 33.30 (CH_2), 49.20 (CH cyclohexyl), 56.22 (CH_3), 57.66 (αCH), 111.91 (CH), 115.58 (2CH), 115.62 (CH), 120.44 (CH), 121.07 (CH), 127.15 (C), 131.70 (C), 145.63 (C), 147.05 (C), 148.16 (C), 149.25 (C), 166.41 (C=O), 170.39 (C=O) ppm.
HRMS (ESI): m/z $[M + H]^+$ calcd for $C_{22}H_{27}N_2O_6$: 415.1869; found: 415.1864.

***N*-Protocatechoyl, (3,4-dihydroxyphenyl)glycine cyclohexylamide** (**3c**). Procedure C using di-*tert*-butyl (4-formyl-1,2-phenylene) dicarbonate (0.338 g, 1.000 mmol) and protocatechuic acid was followed. The residue obtained after removal of TFA was chromatographed through silica using as eluents from ethyl acetate/petroleum ether 1:3 to neat ethyl acetate to give **3c**.
Yield: 0.292 g (69%); white solid (from ethyl acetate/dichloromethane).
R_f = 0.41 (petroleum ether/ethyl acetate 1:4).
M.p. 177-178 °C.
^{1}H NMR (400 MHz, CD_3COCD_3): δ = 1.21-1.33 (m, 6H, CH_2 cyclohexyl), 1.57-1.91 (m, 4H, CH_2 cyclohexyl), 3.71-3.74 (m, 1H, CH cyclohexyl), 5.55 (d, *J* = 7.6 Hz, 1H, αCH), 6.78 (d, *J* = 8.0 Hz, 1H, ArH), 6.85 (dd, *J* = 8.4 Hz, *J* = 2.0 Hz, 1H, ArH), 6.90 (d, *J* = 8.0 Hz, 1H, ArH), 7.03 (d, *J* = 2.0 Hz, 1H, ArH), 7.35 (dd, *J* = 8.4 Hz, *J* = 2.0 Hz, 1H, ArH), 7.48 (d, *J* = 2.0 Hz, 1H, ArH), 7.72 (d, *J* = 7.2 Hz, 1H, $C_6H_3CON\mathit{H}$), 7.95 (br. s, 1H, OH), 8.08 (br. s, 1H, OH), 8.45 (br. s, 1H, OH), 8.49 (br. s, 1H, OH) ppm.
^{13}C NMR (100.6 MHz, CD_3COCD_3): δ = 25.49 (CH_2), 25.57 (CH_2), 26.23 (CH_2), 33.21 (CH_2), 33.27 (CH_2), 49.23 (CH cyclohexyl), 57.35 (αCH), 115.54 (CH), 115.86 (CH), 119.84 (2CH), 120.36 (2CH), 127.07 (C), 127.10 (C), 131.91 (C), 131.94 (C), 145.65 (C), 149.26 (C), 166.17 (C=O), 170.34 (C=O) ppm.
HRMS (ESI): m/z $[M + Na]^+$ calcd for $C_{21}H_{24}N_2NaO_6$: 423.1532; found: 423.1528.

***N*-Hydrocaffeoyl, (3,4-dihydroxyphenyl)glycine cyclohexylamide** (**4c**). Procedure C using di-*tert*-butyl (4-

formyl-1,2-phenylene) dicarbonate (0.338 g, 1.000 mmol) and hydrocaffeic acid was followed. The residue obtained after removal of TFA was chromatographed through silica using as eluents from ethyl acetate/petroleum ether 1:3 to neat ethyl acetate to give **4c**.
Yield: 0.317 g (74%); white solid.
R_f = 0.45 (petroleum ether/ethyl acetate 1:4).
M.p. 112-113 °C.
^{1}H NMR (400 MHz, CD_3OD): δ = 1.18-1.36 (m, 6H, CH_2 cyclohexyl), 1.61-1.90 (m, 4H, CH_2 cyclohexyl), 2.32-2.54 (m, 2H, CH_2), 2.72-2.77 (m, 2H, CH_2), 3.72-3.88 (m, 1H, CH cyclohexyl), 5.23 (s, 1H, αCH), 6.54 (s, 1H, ArH), 6.64-6.68 (m, 2H, ArH), 6.74-6.68 (m, 5H, ArH), 7.02 (d, *J* = 8.4 Hz, 2H, ArH) ppm.
^{13}C NMR (100.6 MHz, CD_3OD): δ = 26.06 (CH_2), 26.12 (CH_2), 26.59 (CH_2), 29.27 (CH_2), 33.47 (CH_2), 33.5 (CH_2), 38.28 (CH_2), 49.97 (CH cyclohexyl), 58.28 (αCH), 114.77 (CH), 115.70 (CH), 116.37 (CH), 117.49 (CH), 118.69 (CH), 120.23 (CH), 130.31 (C), 130.59 (CH), 131.47 (C), 131.68 (C), 134.96 (C), 144.61 (C), 146.46 (C), 159.31 (C), 171.97 (C=O), 174.80 (C=O) ppm.
HRMS (ESI): m/z $[M + H]^+$ calcd for $C_{23}H_{29}N_2O_6$: 429.2026; found: 429.2030.

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Conflict of Interest: the authors declare that they have no conflict of interest.

This article does not contain any studies with human participants or animals performed by any of the authors.

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