ՀԱՅԱՍՏԱՆԻ ԳԻՏՈՒԹՅՈՒՆՆԵՐԻ ԱԶԳԱՅԻՆ ԱԿԱԴԵՄԻԱ
НАЦИОНАЛЬНАЯ АКАДЕМИЯ НАУК АРМЕНИИ
NATIONAL ACADEMY OF SCIENCES OF ARMENIA
ДОКЛАДЫ ԶԵԿՈՒՅՑՆԵՐ REPORTS

Հատոր Том Volume 124 2024 № 3–4

PHARMACOLOGY

УДК 556.3; 540.4
DOI: 10.54503/0321-1339-2024.124.3-4-95

A.S. Grigoryan, Zh.R. Alaverdyan, N.A. Pagutyan, O.A. Hunanyan, S.V. Baghdasaryan, K.G. Navoyan A.A. Agekyan, H.V. Gasparyan

Antimonoamine Oxidase and Antioxidant Properties of Phenolic Compounds Functionalized with Amine and Carboxyl Groups

(Submitted by corresponding member of V.O. Topuzyan 03/VI 2024)

Keywords: *phenolic compounds, carboxy and aminoderivatives, antimonoaminoxidaseactivity, antioxydative properties.*

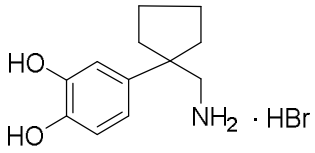
In recent decades., there has been increased interest in the molecular aspects of changes in the structure and expression of genes in the pathogenesis of the development of the tumor process. There is a point of view that in the development of the tumor process and a number of other diseases of the aging process, a significant role is played by the damaging effect of free radicals (FR) – highly reactive particles with unpaired electrons [8], in particular reactive oxygen species (ROS), such as hydrogen peroxide H_2O_2. Hydrogen peroxide is formed in the cell as a product of a number of enzymatic reactions, for example in the oxidation of β-D-glucose to glucono – 1,5-lactone, catalyzed by glucose oxidase.

Although ROS play an essential role in cell metabolism, in particular, as mediators of intracellular signaling, inducers of the immune system and repair processes, initiators of apoptosis, etc. [3], an increase in their intracellular level often leads to the so-called oxidative stress [6], that is destructive effects on the cell. The result of this destructive effect of ROS is, in particular, damage to nucleic acids, peroxidation of polyunsaturated fatty acids of lipids (LPO), oxidation of amino acids and cofactors of a number of enzymes, etc. [7]. Under physiological conditions, the elimination of ROS from the cell is carried out enzymatically, in which antioxidant enzymes (superoxide dismutase, catalase, peroxiredoxins) and through low molecular weight antioxidants (A, E, C, glutathione, uric acid, flavonoids, carotenoids, sulfur-containing compounds, etc.) are involved [4].

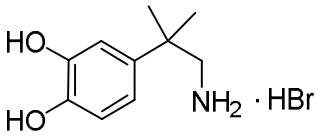
It is important to note that in the brain, the main source of free radicals is hydrogen peroxide, produced in deamination reactions catalyzed by monoamine oxidase (MAO). It is known that MAO is the main enzyme that controls the concentrations of biologically active amines – neurotransmitters through their deamination, and the reaction by-products are toxic aldehyde and ammonia. Since low concentrations of biogenic amines in the blood are considered as one of the possible mechanisms for the occurrence and development of neuropsychiatric diseases, increased MAO activity not only contributes to a decrease in the concentration of biogenic amines, but also to an increase in the concentration of hydrogen peroxide and an increase in destructive oxidative processes.

Taking into account the fact that modern antidepressant drugs and peroxidation inhibitors are not always quite effective and are not without side effects, it seems justified to search for new compounds that simultaneously exhibit both of these biological effects.

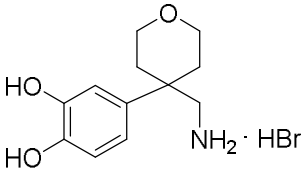
PHENOLIC COMPOUNDS FUNCTIONALIZED WITH AMINE (1,2) AND CARBOXYL (3–5) FUNCTIONS [1]



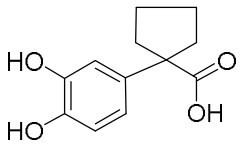
4-(1-(aminomethyl)cyclopentyl)benzene-1,2-diol hydrobromide (**1**)



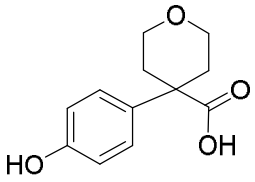
4-(1-amino-2-methylpropan-2-yl)benzene-1,2-diol (**2**)



4-(4-(aminomethyl)tetrahydro-2*H*-pyran-4-yl)benzene-1,2-diol hydrobromide (**3**)



1-(3,4-dihydroxyphenyl)cyclopentanecarboxylic acid (**4**)



4-(4-hydroxyphenyl)tetrahydro-2 *H*-pyran-4-carboxylic acid (**5**)

Materials and methods. The research was carried out on outbred white male rats weighing 180–220g, kept on a normal diet. The test compounds were dissolved in 1 ml of DMSO and administered intraperitoneally to experimental animals at a dose of 0,2 mg/kg per rat, and control animals were administered 1 ml of DMSO.

Euthanasia of animals was carried out under Nembutal anesthesia administered intraperitoneally at a dose of 40 mg/kg. At the final stage, the sacrificed animals were cremated. The work is carried out in accordance with the European Council Directive (2010163IEU) on the care and use of experimental animals.

After autopsy, the brain and liver were isolated and washed with saline. The solution was cleared of blood vessels and homogenized in Tris–HCl buffer (pH–7.,4). The level of lipid peroxides was determined in a non-enzymatic (ascorbate-dependent) peroxidation system based on the yield of the final product – malondialdehyde (MDA), which forms a complex compound with thiobarbituric acid in the form of a pink chromogen, the color intensity of which was recorded spectrophotometrically (at a wavelength of 535 nm) and corresponded to the amount of peroxide formed [2].

The antioxidant activity (AOA) of the tested compounds was judged by the percentage changes in the amount of MDA in the experimental samples compared with the control samples per 1 g of a predetermined amount of protein.

The effect of anti-MAO on the activity of phenolic compounds was also studied.

The source of MAO was 50% bovine brain homogenate, which was obtained by homogenizing the brain in a glass homogenizer with an equal (by weight) volume of 2,5% Arcopal solution [5]. The activity of MAO in the resulting homogenate was determined. The test samples contained 0,2 ml of homogenate, 0,18 ml of a solution of the test compound and 0,18 ml of a substrate solution. The sample volume was adjusted to 1,8 ml with 0,1 M Na-K phosphate buffer to pH-7,4. Serotonin (5-OT) creatinine sulfate monohydrate was used as a substrate,

which was added to the samples after a 30-minute preincubation of the enzyme with the test substance at room temperature 18–25°C. Oxygen saturation was carried out for 5 min. at 37°C. The reaction was stopped by adding 0,2 ml of 50% trichloroacetic acid. The protein precipitate was separated by centrifugation at 3000 rpm. In the protein-free sedimentary liquid, the ammonia content was determined by isometric distillation for 24 hours, followed by non-sterilization and photometry using an FEK 56–2 photometer-nephelometer (at a wavelength of 450 nm). MAO activity is expressed as a % ratio to the control (indopan). Each compound was tested in 3 experiments. The results obtained were processed statically using the Grafpad-Instat method.

Results. 5 compounds were studied for antioxidant and anti-MAO activity.

Table 1

The influence of new compounds on the MDA content (mg/kg protein) in the brain of white rats in in vitro experiments

№	Control mg/kg (n=10)	Experience mg/kg (n=10)	% Experience from control	% Difference from control
1.	12.5±0,9	1.60±0,2	13,28	86,72
2.	12.5±0,9	2,12±0,4	17,59	82,41
3.	12.5±0,9	1,99±0,3	16,51	83,49
4.	12.5±0,9	1,79±0,3	14,85	85,15
5.	12.5±0,9	1,41±0,2	11,70	88,3

Table 2

Effect of the studied compounds on the MDA content (mg/kg) in the liver of white rats in in vitro experiments

№	Control mg/kg (n=10)	Experience mg/kg (n=10)	% Experience from control	% difference from control
1.	12.5±0,9	2,18±0,4	18,09	81,09
2.	12.5±0,9	3,21±0,4	26,64	73,38
3.	12.5±0,9	2,56±0,3	21,24	78,76
4.	12.5±0,9	2,24±0,3	18,59	81,41
5.	12.5±0,9	2,5±0,3	17,01	82,99

The results of the study showed that the studied compounds 1–5 exhibit high antioxidant activity, which reduces the intensity of oxidative processes in the body (Table 1.2).

According to the studies conducted, it can be concluded that the compounds have antioxidant activity on the process of free radical oxidation of lipids in the brain and liver of white rats. According to the data obtained, it can be assumed that this group of compounds is promising in terms of searching for new antioxidant compounds.

Table 3

The effect of studied compounds on the deamination of serotonin (5-OT) by bovine brain MAO in vitro.

Compounds	Inhibition MAO activity 0.,5 µmol/ml	Inhibition MAO activity 1.0 µmol/ml	P
1	32±4,0	68±,3,8	<0,05
2	50±3.8	84±5,2	<0,05
3	36±3,2	78±4,6	<0,05
4	25*	48±3,2	<0,05
5	22*	48±3,2	<0,05
indopan	54±5,8	86±6,0	

** reliability not calculated due to low activity of compounds*

The intensity of serotonin deamination in control samples was accepted as 100%

Indopan [9] (a reversible inhibitor of monoamine oxidase) was used as a control, exhibiting a pronounced anti-MAO effect at concentrations of 0,5 and 1,0 µmol/ml. It significantly inhibited the deamination of serotonin creatinine sulfate monohydrate (5-OT), making it suitable for comparison with our compounds. Based on the fact that the compounds at 1,0 µmol/ml showed the highest activity (84%), further studies were conducted at 0,5 µmol/ml, where the activity was 50%. The data obtained demonstrated that the activity of the tested compound at 0,5 µmol/ml was nearly identical to the control. The remaining compounds exhibited varying degrees of anti-MAO activity at concentrations of 0,5 and 1,0 µmol/ml.

As shown in the table, the effect of five compounds on the activity of monoamine oxidase (MAO) in bovine brain was studied in vitro. The results demonstrated that the tested compounds exhibited weak (derivative 4), moderate (derivatives 1, 3, and 5), and, in the case of cyclopentane derivative 2, pronounced anti-MAO properties, inhibiting serotonin deamination by 84%.

Thus, the data suggest that 2-(1-ethylcyclopentyl)-cyclohexa-1,3-hydrobromide with ethane has a dual pharmacological effect and can be considered both as a potential MAO inhibitor for the treatment of depression and as a potential inhibitor of peroxide oxidation.

Scientific and Technological Center of Organic and Pharmaceutical Chemistry of the National Academy of Sciences of the Republic of Armenia.
Institute of Fine Organic Chemistry after A.L. Mnjoyan, Yerevan, Armenia.
e-mail: anna.js@mail.ru

A.S. Grigoryan, Zh.R. Alaverdyan, N.A. Pagutyan, O.A. Hunanyan, S.V. Baghdasaryan, K.G. Navoyan, A.A. Agekyan, H.V. Gasparyan

Antimonoamine Oxidase and Antioxidant Properties of Phenolic Compounds Functionalized with Amine and Carboxyl Groups

The effect of carboxy- and amino-substituted phenolic compounds on the activity of monoamine oxidase (MAO) in bovine brain and on oxidative processes in the brain and liver of rats was studied*in vitro*. It has been established that some of the studied compounds exhibit certain anti-MAO and antioxidant properties. An attempt was made to compare the biological properties of the compounds with the activity of known drugs. In the series of aminosubstituted derivatives, 4-(1-amino-2-methylpropan-2-yl) benzene-1,2-diol displaed high antimonoamine oxidase and antioxidant activity.

Ա.Ս. Գրիգորյան, Ժ.Ռ. Ալավերդյան, Ն.Ա. Պագուտյան, Օ.Ա. Հունանյան, Ս.Վ. Բաղդասարյան, Ք.Հ. Նավոյան, Ա.Ա. Աղեկյան, Հ.Վ. Գասպարյան

Հակամոնոամինօքսիդազային և հակաօքսիդանտային հատկությունների ուսումնասիրումը ամինային և կարբոքսիլ խմբերի ֆենոլային միացություններում

Հետազոտվել է նոր սինթեզված հեքսաբրոմիդների ակտիվությունը մոնոամինօքսիդազա (ՄԱՕ) ֆերմենտի և հակաօքսիդանտների նկատմամբ առնետի ուղեղի և լյարդի վրա in vitro փորձերում: Հաստատվել է, որ որոշ հետազոտվող միացություններ՝ հիդրոբրոմիդ, հիդրոքսիֆենիլ, 3,4-դիհիդրոքսիցիկլոպենտիլի ածանցյալները ցուցաբերում են որոշակի հակաՄԱՕ և հակաօքսիդանտ ակտիվություն: Փորձ է արվել համեմատելու միացությունների կենսաբանական ակտիվությունը հայտնի միացությունների հակաՄԱՕ և հակաօքսիդանտ ակտիվությունների հետ:

А.С. Григорян, Ж.Р. Алавердян, Н.А. Пагутян, О.А. Унанян, С.В. Багдасарян, К.Г. Навоян, А.А. Агекян, Г.В. Гаспарян

Антимоноаминоксидазные и антиоксидантные свойства фенольных соединений, функционализированных аминными и карбоксильными группами

Изучено влияние карбокси и аминозамещенных фенольных соединений на активность моноаминоксидазы (МАО) в мозгу крупного рогатого скота и на окислительные процессы в мозгу и печени крыс *in vitro*. Установлено, что некоторые из изученных соединений проявляют определенные антиМАО и антиоксидантные свойства. Была предпринята попытка сравнить биологические свойства соединений с активностью известных препаратов. В ряду аминозамещенных производных 4-(1-амино-2-метилпропан-2-ил)бензол-1,2-диол обладает высокой антимоноаминоксидазной и антиоксидантной активностью.

References

1. *Агекян А.А., Маркарян Э.А.* Исследования в области синтеза N 3, 4-диметоксифенилциклопентилметил-N-замещенных диамидов щавелевой кислоты. Хим.ж. Армении. 2013, т. 66, № 4, с. 628–635.
2. *Владимиров Ю.А., Арчаков А.И.* Перекисное окисление липидов в биологических мембранах. М., 1972, с. 252.
3. *Владимиров Ю.А.* Свободные радикалы в биологических системах. Соросовский образовательный журнал. 2000, № 12, с. 13–19.
4. *Владимиров Ю.А., Азизова О.А., Деев А.И.* и др. Свободные радикалы в живых системах. Итоги науки и техники. Биофизика, 1992, т. 29, с. 3–250.
5. *Горькин В.З.* Методы, основанные на измерении освобождаемого аммиака. М., 1981, с. 34.
6. *Зенков Н.К.* Окислительный стресс. Биохимические и патофизиологические аспекты. М., Наука, 2001, 343 с.
7. *Иванова И.П., Зуймач Е.А., Спиров Г.М.* Опухолевые процессы и высокоэнергетические импульсные факторы. Нижегородский медицинский журнал. 2003, № 1, с. 81–85.
8. *Кондакова И.В., Какурина Г.В., Смирнова Л.П., Борунов Е.В.* Регуляция пролиферации и апоптоза опухолевых клеток свободными радикалами. Сибирский онкологический журнал. 2005, №1(13), с. 58–62.
9. *Машковский М.Д.* “Лекарственные средства”. М., “Новая волна”, 2010, с. 851.