COORDINATION COMPOUND AS INHIBITOR OF SUPEROXID RADICAL

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Rezumat

A fost obținut un nou compus coordinativ biologic activ, nitratul de cupru [2 - ({2 - [(ethylsulfanyl) (prop-2-en-1-yl) carbononoimidooyl] -hydrazinylidene} methyl) phenolate], care face parte din clasa isotiosemicarbazidelor metalelor de tranziție.S-a stabilit că acesta exercită proprietăți anti radicalice performante la acțiunea moleculei organice cu radicalul superoxidic. Datorită acestei poroprietăți compusul obținut poate avea o potențială aplicare in medicină in calitate de inhibitor al radicalilor superoxidici in corpul uman, care poate duce la prevenirea deteriorării tesutului cellular, aterosclerozii si carcinogenezei.Compusul coordinativ sintetizat extinde arsenalul inhibitorilor de radicali superoxidici cu activitate biologică importanta.

Cuvinte cheie: compuși coordinativi, derivați ai tiosemicarbazidei, inhibitori ai radicalilor superoxidici

Depus la redacţie 26 noiembrie 2019

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Introduction

Multifactorial diseases (MFDs), such as atherosclerosis, chronic inflammatory processes, the neurodegenerative diseases, cancer, etc., continue to remain in the

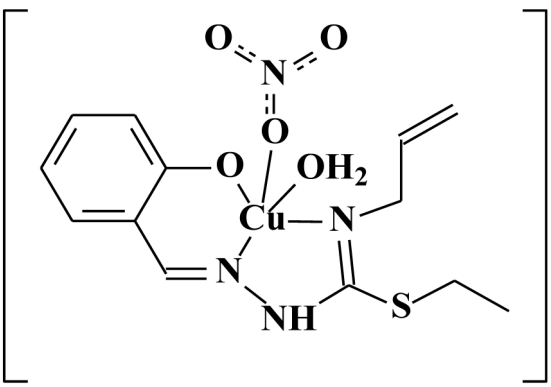
attention of physicians worldwide due to their increasing incidence and high mortality, caused by the imperfection of the therapeutic means proposed by modern medicine. Oxidative destruction mediated by free radicals at the molecular and cellular level plays a key role in the pathogenesis of these diseases is demonstrated [5]. Therefore, the problem of developing, highlighting and selecting new effective remedies, which could be used for MFDs prophylaxis and treatment, remains a problem of great importance and practical value.

Respectively, one of the priority directions of modern applicative chemistry is the synthesis of new compounds that capture and neutralize reactive oxygen species (ROS), in particular the superoxide radical, thus preventing the development of cellular and tissue damage, including inflammatory processes in the human body, atherosclerosis and carcinogenesis.

In this aspect, the coordination compounds of the transition metals, derivatives of thiosemicarbazones represent a special interest. Preliminary research has revealed the therapeutic efficacy and prospects of using these compounds [7, 9].

Materials and methods

A new, biologically active, copper-coordinating compound in the class of isothiosemicarbazides of transition metals - nitrato- [2 - ({2 - [(ethylsulfanyl) (prop-2-en-1-yl) -carbonoimidoyl] -hydrazinyliden} methyl) phenolato] aquacopper of formula (Scheme 1) was investigated.



Scheme 1. Nitrato-[2-({2-[(ethylsulfanyl)(prop-2-en-1-yl)-carbonoimidoyl]-hydrazinyliden} methyl)phenolato]aquacopper.

This compound at the State University of Moldova in the Laboratory "Advanced materials in biopharmaceutical and technical" was synthesized [3], but its influence on oxidative processes with ROS, such as superoxide radical, was not studied.

The superoxide radical scavenging activity was determined by the spectrophotometric method, described in [2, 8] with some modifications.

The method is based on the generation of superoxide radical by the phenazine methosulphate / nicotinamide adenine dinucleotide (PMS/NADH) system by oxidation of NADH, and the superoxide radical reduces the tetrazolium salt - Nitro Blue Tetrazolium (NBT) to blue-purple formazan.

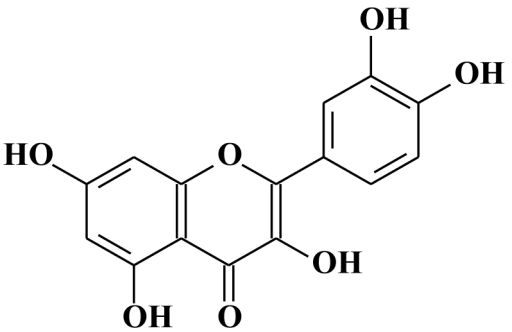
The method was carried out as follows: The working dilutions of the tested compound dissolved in DMSO were prepared: 1.0; 10.0; 100 μM. Then, 20 μl of each working dilution of the tested compound was pipetted into 96-well microplate wells. Each dilution was poured in duplicate. Then 180 μl of reaction medium (mixture)

containing 20 mM phosphate buffer solution (pH 7.4), NADH (0.1 mM) and NBT (0.09 mM) was added. The control sample was mounted the same as the test sample, but instead of dilutions of the tested compound, an equivalent amount of 20 mM phosphate buffer solution (pH 7.4) was added. It was prepared in duplicate. After mixing the absorbance was measured at 560 nm [Ao]. Then, in all the wells, 20 µl of 8.0 µM phenazine methosulphate (PMS) solution was added, for 10-15 s was stired and at room temperature for exactly 5 min was incubated. Absorbance was measured again at 560 nm [A1]. As a reference substance, quercetin in concentrations 1.0; 10.0; 100 µM was used.

The percent of superoxide radical scavenging activity (SRSA) was calculated according to the formula:

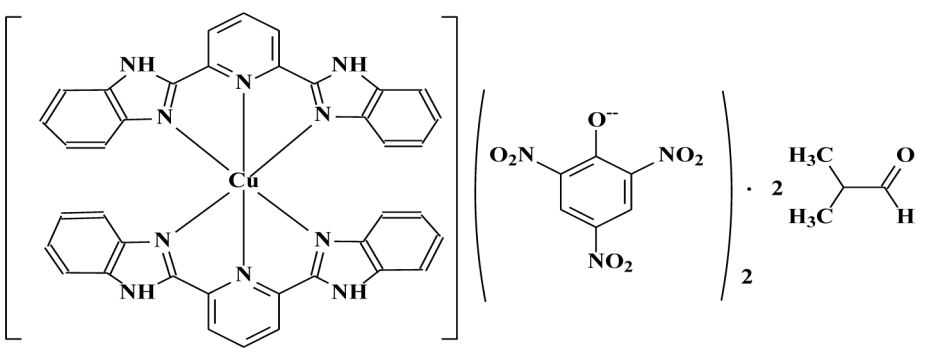
SRSA (%) = [100-(A1 / Ao)] x 100

Quercetin (3,3', 4,5,6-pentahydroxyphlavone) as a reference substance to determine the superoxide radical scavenging activity was used (Scheme 2):



Scheme 2. Quercetin (3,3',4,5,6-pentahydroxyphlavone).

which represents a natural flavonol from the polyphenolic flavonoid group [1] and bis(2,4,6-trinitrophenolate) bis (2,2'-pyridin-2,6-diyl-kN) -bis-1H-benzimidazole] -copper (II) bis(N,N-dimethylformamide) solvate [4] of the formula (Scheme 3).



Scheme 3. Bis(2,4,6-trinitrophenolate) bis[2,2'-pyridin-2,6-diyl-kN)-bis-1H-benzimidazol]-copper (II) bis(N,N-dimethylformamide) solvate.

Results and discussion

The nitrato- [2- ({2- [(ethylsulphanyl) (prop-2-en-1-yl) - carbononoidido] -hydrazinylidene} methyl) phenolate] aquacompound has anti-radical activity has been established with IC50 equal to 0.86±0.15 µM, which was 71.1 times higher than quercetin

activity, used as a standard for determining the inhibitory activity of superoxide radical and was 1.15 times more effective than bis (2,4,6-trinitrophenolate) bis (2,2'-pyridin-2,6-diyl-kN) -bis-1H-benzimidazole]-copper (II) bis (N, N-dimethylformamide) solvate (Table 1).

Table 1. Anti-radical activity of the investigated compound in comparison to quercetin.

Compound	IC_{50}, µM
Quercetin (3,3',4,5,6-pentahydroxyflavon) [1]	61.86±2.51
Bis(2,4,6-trinitrophenolate) de bis[2,2'-piridin-2,6-diil-kN)-bis-1H-benz-imidazol]- copper (II) bis(N,N-dimethylphormamid) solvate [3]	0.99±0.09
Nitrato-[2-({2-[(etylsulphanil)(prop-2-en-1-il)carbonoimidoil]-hydrazy-niliden}methyl)phenolato]aquacopper	0.86±0.15

The established property of the above-mentioned nitrate-[2-({2-[(ethylsulphanyl) (prop-2-en-1-yl) carbononoimidoyl]-hydrazinylidene} methyl) phenolato] aquacopper is new, because its use as an inhibitor of superoxide radical has not been described yet.

Comparative analysis of nitrato- [2 - ({2 - [(ethylsulphanyl) (prop-2-en-1-yl) carbon-imidooyl] hydrazinylidene} methyl) phenolate] aqua with bis (2,4,6-trinitrophenolate) compound of bis [2,2'-pyridine-2,6-diyl-kN) -bis-1H-benzimidazole] -copper (II) bis (N, N-dimethylformamide) solvate demonstrate that they differ in that they belong to different classes of coordination compounds of copper (II) and in this compound a new combination of already known chemical bonds was made.

Due to the high reactivity, the superoxide radical O_2^- (which is formed by capturing an electron upon O_2 activation) is responsible for multiple harmful actions in the body, such as inflammation, reperfusion injury, radiation damage, metabolic disorders, cell aging, atherosclerosis and carcinogenesis [6]. Therefore, therapeutic inhibition of superoxide radical is a new contribution, because compounds with superoxide antiradical activity show a strong curative effect, preventing the development of cellular and tissue damage [4]. The detected properties of nitrato- [2 - ({2 - [(ethylsulphanyl) (prop-2-en-1-yl) carbon-imidooyl] hydrazinylidene} methyl) phenolate] aquacopper are of interest in medicine in terms of expanding the arsenal of synthetic inhibitors of superoxide radical. This complex can inhibit the exacerbation of processes affecting organic molecules with superoxide radical in the body. Due to these properties, it can find application in medicine as an inhibitor of superoxide radical in the body, thus preventing the development of cellular and tissue damage, characteristic of MFDs, such as chronic inflammatory processes, atherosclerosis and carcinogenesis. Therefore, we believe that the obtained data signify a beginning that opens the perspectives of elaborations, which will diversify the arsenal of remedies for combating different pathological processes.

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