Synthesis, Characterization and Antioxidant Activity of Co(II) and Cd(II) Complexes with Quercetin

SVETLANA TRIFUNSCHI*, DORINA ARDELEAN
Vasile Goldis Western University of Arad, Faculty of Pharmacy, Department of Pharmaceutical Science, 86 Liviu Rebreanu Str., 310045, Arad, Romania

Complexes of cobalt(II) and cadmium(II) with quercetin have been synthesized. The structure of compounds are confirmed by FTIR, and UV-Vis spectral data. The antioxidant activity of free quercetin and quercetin complexes have been evaluated by using 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging method. These complexes of flavonoid have high antioxidant activity than the free flavonoid which recommends them for further work in possible therapeutic applications.

Keywords: quercetin, flavonoid, antioxidant activity, Co(II)-quercetin complex, Cd(II)-quercetin complex

Transition metal ions participate in the initiation of free radical processes. The antioxidant action of flavonoids has been considered to be via two possible ways of action: metal chelation and radical scavenging. Radical scavenging aries by donation of hydrogen from the free hydroxyl groups on the flavonoids ring and other possible antioxidant mechanism for flavonoids is metal chelation through phenolic OH group, preventing metal-mediated generation of free radicals and may shelter the potential biological target from oxidative stress [1, 2].

Among the most dietary and biologically active flavonoids, quercetin was selected. Quercetin ($C_{15}H_{10}O_7$) (3,3',4',5,7-pentahydroxyflavone) is one of the most familiar major dietary flavonols found in nature. Because is a flavonoid of flavonol type that contain five hydroxyl group in 3,5,7,3' and 4' position and a carbonyl group in 4 position; quercetin easily forms complexes with many metals. Many studies demonstrate that quercetin is a strong antioxidant and has many beneficial effects on health, including cancer, cardiovascular protection, ulcer, and antiallergic, antiviral and anti-inflammatory properties [3,4].

The experimental data show that the chelate compound are more effective for scavenging free radicals that flavonoids ligands and play a role by protecting from oxidative stress and anti-microbial body. The complexes of rutin wit Zn (II) and Cu (II) are better antioxidant activity than simple ligands [5,6]; the complexes of Cd (II) with quercetin at *p*H 4.4 had less antioxidant activity compared to the free quercetin [7].

The interaction of quercetin with divalent cations changes the antioxidant properties and biological effects of free flavonoids.

Due to the importance of metal chelation in the antioxidant behaviour of flavonoids and continuing our interest in flavonoid-metal complexes, we report here the synthesis, characterization and antioxidant activity of the Co(II) and Cd(II) complexes with quercetin.

Experimental part

Materials

All chemichals were of the highest purity commercially avaible. Quercetin dihidrat was purchased from Fluka; $Co(NO_3)_2 \cdot 6H_2O$, $Cd(NO_3)_2$ and methanol were obtained from Sigma- Aldrich.

Synthesis of the complexes

The complexes were synthetized by Bukhari modified method [6].

Synthesis of cobalt (II)- quercetin complex (A)

In a bottomed flask equipped with an electromagnetic stirrer, quercetin ($C_{15}H_{10}O_7 \cdot 2H_2O$: 0.17 g / 0.0005 mol) dissolved in methanol (20 mL) within 10 min, the color of the solution was light yellow. Consequently, $Co(NO_3)_2 . 6H_2O$ (0.29g / 0.001.moli) was added quickly in the reaction mixture and the solution was stirred at room temperature for 2 hours. After stirring, the reaction mixture was filtered and the filtrate was evaporated slowly at room temperature. The resulting brownish yellow product was washed with t-butanol to remove the unreactive part of the reagent and dried in a vacuum desiccator.

Synthesis of cadmium (II)- quercetin complex (B)

In a bottomed flask equipped with an electromagnetic stirrer, quercetin ($C_{15}H_{10}O_7 \cdot 2H_2O$: 0.17 g / 0.0005 mol) dissolved in methanol (20 mL) within 10 min, the color of the solution was light yellow. Consequently, $Cd(NO_3)_2$ (0.30g / 0.001moli) was added quickly in the reaction mixture and the solution was stirred at room temperature for 2 h. After stirring, the reaction mixture was filtered and the filtrate was evaporated slowly at room temperature. The resulting green yellow product was washed with t-butanol to remove the unreactive part of the reagent and dried in a vacuum desiccator.

Chemical and spectral analysis

UV-Vis spectra of the quercetin and its metal complexes were recorded on a Metertech UV-VIS spectrophotometer SP- 8001 using standard of 1.00 cm quarz cells and methanol as a solvent in the range of 250-800 nm

FTIR spectra were recorder on a Perkin Elmer FTIR spectrometer: Spectrum Two.

Determination of the antioxidant activity

The antioxidant activity of free quercetin and of the obtained complexes (Co(II)-quercetin and Cd(II)-quercetin) was determined by method using the stable 2,2-diphenyl-1-picrylhydrazyl radical (DPPH·) [8].

Methanol solution (0.1 mL) containing different concentration of quercetin (4, 8, 12, 16 and 40 μM) was

* email: trifunschi.svetlana@uvvg.ro; Phone: (+40)724496322

added to 3.9 mL of freshly prepared (57.65μM) DPPH in methanol. The reduction of the DPPH was followed by monitoring the decrease in absorbance at 517 nm in each 5 min for about 30 min ($A_{t=30}$). As a control, the absorbance of blank solution of DPPH (4 mL) was also determined at 517 nm ($A_{t=0}$).

The decreanse in absorbance was converted in to percentage by using formula:

$$AA\% = (A_{t=0}-A_{t=30}) / A_{t=0}$$

Results and discussions

UV-Vis study of the complexes

The changes in UV-Vis absorbtion of quercetin in the presence of Co (II), respectivelly Cd (II) were examined in the methanol solution.

Quercetin, like most flavonoids display two main absorbtion bands, in the ultraviolet-visible region, at 372 nm (band I) showing B ring absorbtion (cinamoyl system) and 256 nm (band II) is considered to be associated with the absorbtion involving the A ring (benzoyl system) [9] (fig. 1).

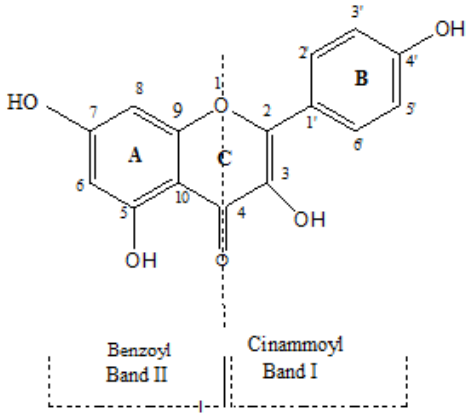

Fig. 1. Structure of quercetin and related UV-Vis absorbtion bands

The UV-Visible spectra given information about the coordination sites of quercetin; for example, the interactions of Metal (II) ions with quercetin at 2:1 metal/ quercetin ratio produces bathochromic shift in the absorbance of both bands (I and II): 262 nm and 375 nm for Co(II)- quercetin complex; 258 nm and 428 nm for Cd(II)-quercetin complex. The results indicated formation of complexes between quercetin and Co (II), respectivelly quercetin and Cd (II). Because the hydroxyl group of 3 position has a more acidic proton, therefore the 3-OH and oxygen of carbonyl group at position 4 are the first sites to be involved in the complexation process. The hydroxyl groups of 3' and 4' position bind a second metal ion [10].

FTIR spectra

The spectral data showed the evidence for the coordination between the metals and quercetin. Some features of the spectra are discussed bellow. The broad bands of O-H vibration frequency located at 3262 cm^{-1}, 3432 cm^{-1}, 3240 cm^{-1} indicates the existence of water in the complex and free quercetin. The C=O stretching of free quercetin occurs at 1658 cm^{-1} and due the interaction of quercetin with metals, the absorbtion band has been shifted to 1651 cm^{-1} for Co(II)-quercetin complex and to 1658 cm^{-1} for Cd(II)-quercetin complex. The bands located at 1351 and 1314 cm^{-1} for quercetin, 1554 cm^{-1} for Co(II)-quercetin complex and 1247 cm^{-1} were related to C-OH deformations vibrations. The bond related to the C-O-C

Table 1

ASSIGNMENT OF THE MAIN FTIR BANDS OF COMPLEXES

Quercetin	Co(II)-quercetin complex	Cd(II)-Quercetin complex	Assignments
3292	3432	3240	ν O-H
1658	1624	1651	ν C=O
1611	1605	1610	ν inel A sau B
1520	1564	1554	ν C=C
1351	1367	1312	ν C-OH, ν inel B
1241	1271	1247	ν C-O-C
596, 465	603	456	γ C-HK

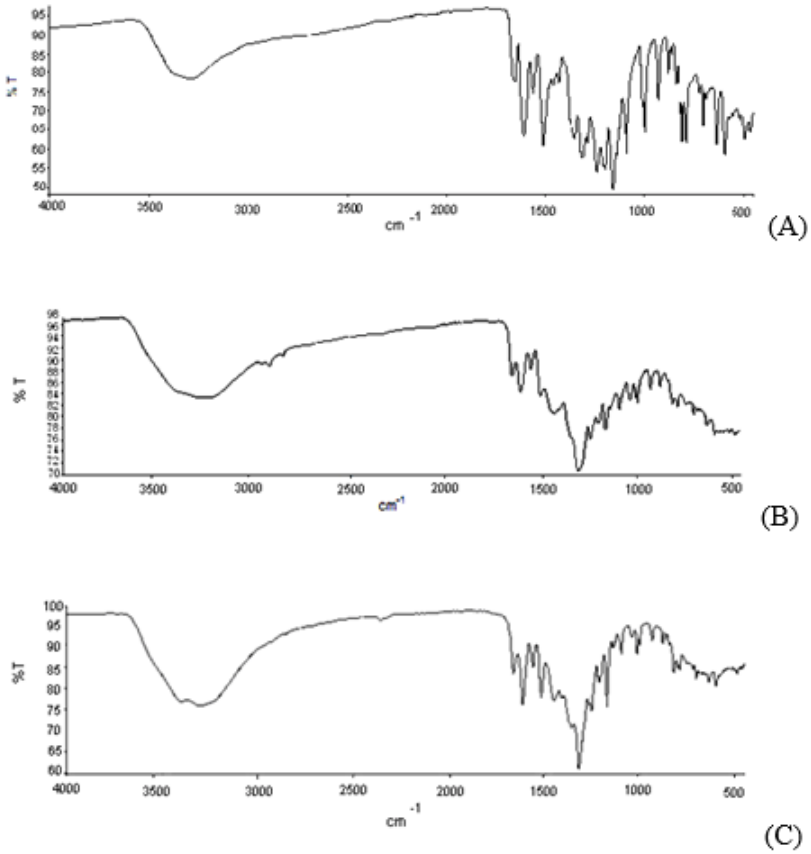

Fig. 2. The FTIR spectra of : (A)- ligand quercetin; (B) Co(II)-quercetin complex; (C) Cd(II)-quercetin complex

indicating that the ring oxygen is not involved in complexation. The aperance of peak at 603 cm^{-1} indicates the existence of O-Co bond in the complex and peak at 456 cm^{-1} indicates the existence of O-Cd bond.

The vibration at 1241 cm^{-1} for quercetin, 1271 cm^{-1} for Co(II)-quercetin complex and 1247 cm^{-1} for Cd(II)-quercetin complex indicating is not involved in the complexation [11].

FTIR spectra of quercetin, Co(II)-quercetin complex and Cd(II)-quercetin complex were summarized in Table 1 and spectra is shown in figure 2.

Table 2

ANTIOXIDANT ACTIVITY (%) ON DPPH RADICALS OF QUERCETIN COMPLEXES (A AND B) AND STANDARD QUERCETIN (0.01M) IN METHANOL

Compound	Antioxidant activity, AA (%)
A: Quercetin-Co(II) complex	74.20
B: Quercetin-Cd(II) complex	82.31
Quercetin	48.43

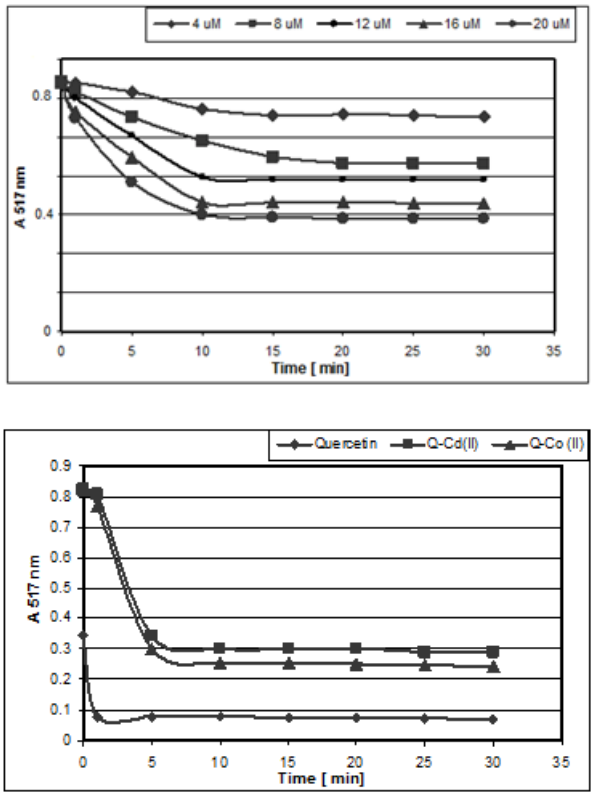

Fig. 3. Decreace in absorbance at λ 517 nm of DPPH (methanolic solution) in the presence of different concentrations of quercetin (a) and quercetin-metal(II) (b)

DPPH radical scavenging analysis

The antioxidant activity of quercetin and its complexes Co(II)-quercetin and Cd(II)-quercetin was measured in terms of their hydrogen donating or radical scavenging ability by UV-Vis spectrophotometer using the stable DPPH.

The reaction between quercetin and DPPH occurs in two steps: (1) DPPH absorbance decays quickly (tipic time, 60-120s) and (2) DPPH absorbance decays slowly in ~1 h to reach a constant value [6].

The complexation with metal ions decreased the oxidation potential of the flavonoid. The complexed flavonoid is relatively more readily oxidized by the free radical then the uncomplexed once. The metal complex can be oxidized to a Me(II)- semiquinone complex by free radical, which is oxidized by another free radical to form the Me(II) -semiquinone complex. (fig.3)

Figure 3- (a) show kinetic behaviour in the presence of different concentration of quercetin (4 - 20μM). and its estimated that the antioxidant activity is a function of time. The kinetic behavior in the presence of quercetin and of the metal-quercetin complexes is shown in figure 3- (b).

The antioxidant activity of methanolic solution of complexes are presented in table 2 and compared with the ligand- quercetin. The antioxidant activity in percentage are expressed as the ratio of absorbance decrease at 517 nm. The complexes, compounds A and B showed excellent antioxidant activity (74.20 %, respectively 82.31 %).

The antioxidant activity of the complexes was greater than the ligand quercetin. This suggests that the metal ions Co(II) or Cd(II) significantly change the chemical properties of the ligand quercetin.

Conclusions

This study described the synthesis, characterization and antioxidant activity of quercetin- metal complexes as potential antioxidants. The UV-Vis spectra of quercetin complexes presents a band to the benzoyl system – with maximum at 262 nm (quercetin –Co(II) complex) and 258 nm (quercetin –Cd(II) complex) and another band to the cynnamoyl system with maximum at 375 nm (quercetin –Co(II) complex) and 428 nm (quercetin – Cd(II) complex).

The coordination sites and the binding properties of quercetin were determined by using FTIR spectroscopy. The spectroscopic data show the importance of the 3-OH group which is coordination site of the ligand. Because the hydroxyl group of position 3 has a more acidic proton, therefore the 3-OH and oxygen of carbonyl group at position 4 are the first sites to be involved in the complexation process, and the hydroxyl groups of 3' and 4' position bind a second metal ion.

The antioxidant activity of flavonoid depends on the number and position of OH groups presents in the flavonoid structure. The compounds assayed showed excellent antioxidant activity than quercetin, which recommends them for further possible therapeutic applications.

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