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A Theoretical Study on the Chemopreventive Activity of Flavonoid Compounds

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Abstract

An electronic and structural characterization of some flavonoid compounds with chemopreventive activity show that structural effects are more relevant to explain the biological activity of these compounds than the electronic ones. The flavonoid compounds studied were classified as having a moderate electron-accepting and electron-donating character and no correlation was found between the electronic descriptors studied and the chemopreventive activity presented by the compounds.

Keywords. Flavonoids; chemopreventive activity; AM1.

1 INTRODUCTION

Flavonoids are compounds with varied chemical structures and are commonly found in fruits, vegetables, nuts and seeds. These compounds are found at relatively high concentrations in the human diet and comprise several classes of molecules including flavonols, flavonones, flavanols and flavans.

These compounds have received considerable attention in recent years. A variety of properties have been found on several recent reports such as metal binding capacity, antioxidant activity, ability to affect the endocrine system and ability to prevent the enzymic activation in carcinogenesis [1]. Some flavonoid compounds are able to protect rodents against chemically-induced carcinogenesis, such as the flavonol and flavan classes and the quercetin molecule [2,3] and many efforts have been made to summarize and elucidate the structure-activity relationship (SAR) of these compounds [4-6]. Nowadays, this kind of SAR studies have been proven to be helpful in the understanding of the influence of molecular properties on the biological activity presented by several kind of compounds.

The flavonoid compounds studied in this work were isolated from *Tephrosia toxicaria* (sw) Pers. (*T. toxicaria*) which is a tropical fish-poisoning plant growing in Sri Lanka and South America and

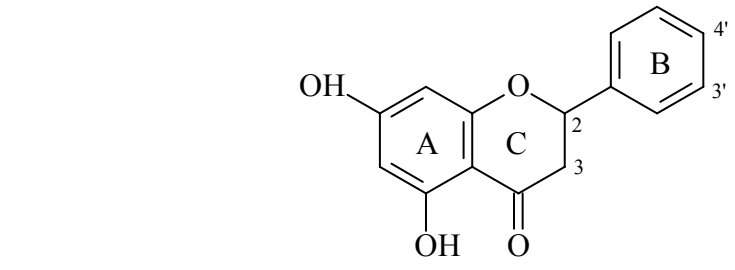
is well-know as a source of rotenoids including deguelin, sumatrol and toxicarol [5-9]. The stems of *T. toxicaria* were chosen as it is known in the literature that their ethyl acetate-soluble extract significantly induces the quinone reductase (QR) enzyme in cultured Hepa 1c1c7 (mouse hepatoma) cells [5]. Recent studies have been reported that the induction of the QR enzyme is considered a major mechanism of protection against tumor initiation [5,6].

The main purpose of this work is to study, theoretically, the molecular properties of some flavonoid compounds found in the stems of *T. toxicaria,* which were used in a bioassay based on the induction of the QR enzyme reported in the literature [5], in order to collaborate for the elucidation of the mechanism of action of these flavonoid compounds with a pronounced chemopreventive activity.

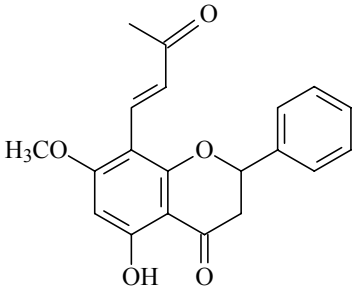
2 MATERIALS AND METHODS

2.1 Chemical Data

The flavonoid compounds studied in this work were isolated and characterized from the stems of the *T. toxicaria* [5]. The general structure of a flavonoid compound, as well as the chemical structure of the flavonoid molecules studied in this work are displayed in Figure 1.



(a)



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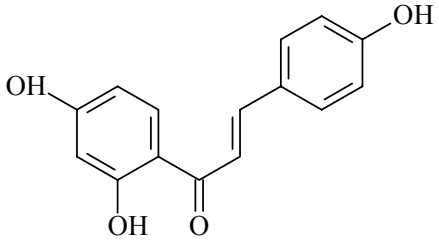

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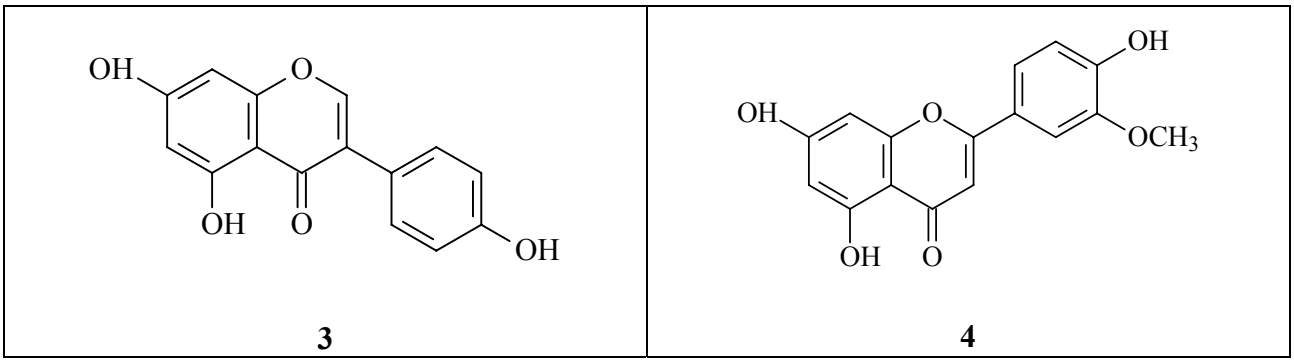

Figure 1. General structure of a flavonoid compound (a), and the chemical structure of the flavonoid molecules studied in this work (numbered as 1, 2, 3 and 4).

The compound 1 is classified as a butenyflavanone and it is named (2S)-5-hydroxy-7-methoxy-8-[(E)-3-oxo-1-butenyl]flavanone; the compound 2 is classified as a chalcone known as isoliquiritigenin; the compound 3 is named as genistein and it belongs to isoflavone class; the compound 4 is classified as a flavone and it is known as chrysoeriol. [5]. These compounds were used in a QR induction assay with the aim to assess their chemopreventive potential (the induction of QR is considered a major mechanism of protection against tumor initiation [5]). The biological data obtained from the QR induction assay are displayed in Table 1. From Table 1 we can see that compound 2 presents a high chemopreventive activity due to its low CD value (3.9 μM), while compound 3 can be considered the less active compound against the QR enzyme, as it presents a CD value >10 μg/mL [5].

Table 1. Biological data obtained for the flavonoid compounds studied by using a quinone reductase (QR) induction assay [5]

Compound	**QR**[a]		
	CD (μM)	IC_{50} (μM)	CI
1	10.7	>14.9	>1.4
2	3.9	36.3	10.1
3	22.9	45.2	2.0
4	11.7	36.0	3.1

[a] CD = concentration required to double QR activity; IC50 = concentration inhibiting cell growth by 50%; CI = chemoprevention index (=IC50/CD). Compounds with CD values <10 μg/mL are considered active.

2.2 Computer Software

2.2.1 Quantum chemistry software

The geometries of the compounds studied in this work were initially optimized by using the molecular mechanic method MM+ [10,11] and an additional reoptimization was carried out employing the AM1 semi-empirical method [12,13]. The calculations of the molecular properties used to describe the structural and electronic features of the compounds under study were performed with the AM1 method and the program Chemplus 1.5 [14]. The atomic charges and the molecular electrostatic potential (MEP) maps were obtained by employing the program Spartan 5.0 [15].

The molecular properties calculated by the AM1 method were: heat of formation (ΔH_f), total energy (E_T), energy of the highest occupied molecular orbital (E_{HOMO}) and energy of the lowest unoccupied molecular orbital (E_{LUMO}). In addiction, the steric properties surface area (A) and molecular volume (V) were calculated by using the Chemplus 1.5 program.

3 RESULTS AND DISCUSSION

The optimized structures of each compound studied are displayed in Figure 2; from them we calculated all of the molecular properties used in this work to be correlated with the chemopreventive activity presented by the flavonoid compounds under study.

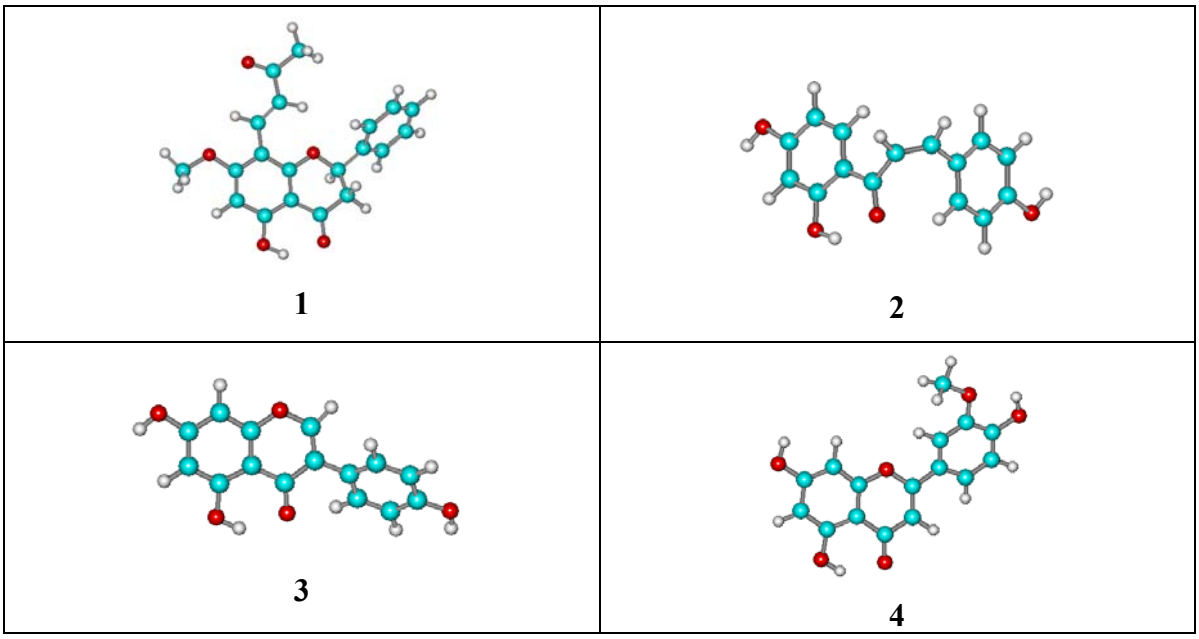

Figure 2. Optimized structure for the flavonoid compounds studied.

In Table 2 are shown the obtained values for the steric properties surface area (A) and molecular volume (V). From Table 2 we can notice that the compounds 2, 3 and 4 have similar values for A and V, while the compound 1 has the highest value for these steric properties. Now, when we compared the results of Table 2 with the biological data of Table 1, we can see that A and V (that are steric properties related to the whole molecule) have no connection with the level of chemopreventive activity presented by the flavonoid compounds studied. However, some regions of these molecules can be important for explaining the chemopreventive activity presented by these compounds.

Table 2. Molecular properties calculated for the flavonoid compounds studied

	A **(Å^2)**	**V** **(Å^3)**	**ΔH_f** **(kcal/mol)**	**E_T** **(eV)**	E_E **(eV)**	**E_{HOMO}** **(eV)**	**E_{LUMO}** **(eV)**
1	559.02	954.35	-123.658	-4409.1	-31724.6	-8.891	-0.721
2	454.61	742.21	-100.332	-3366.34	-19798.2	-8.958	-0.525
3	439.75	720.72	-130.396	-3659.01	-21577.6	-8.800	-0.699
4	486.50	802.01	-166.673	-4134.81	-25591.9	-9.058	-0.944

Analysing the calculated heat of formation (ΔH_f), total (E_T) and electronic (E_E) energies (displayed in Table 2) for the four flavonoid compounds studied, we can see that the compounds have similar values for these properties and, consequently, the optimized structures have similar chemical stability.

From the E_{HOMO} and E_{LUMO} values we can have an idea on the electron-donating and electron-accepting character of a compound and, consequently, an idea on the probability of a charge transfer complex (CTC) be formed [16]. Using the scale obtained for the electron-donating and electron-accepting character of biomolecules [16], we tried to classify the flavonoid compounds studied into two classes: electron donor or electron acceptor molecules. Based on the E_{HOMO} and E_{LUMO} values displayed in Table 2, we can see that all flavonoid compounds have similar E_{HOMO} and E_{LUMO} values, and comparing these values with the ones presented in the electron-donating and electron-accepting scale of Ref. [16], we verified that the compounds studied have moderate electron-accepting and electron-donating character. From these results, we can conclude that the interaction between the flavonoid compounds studied and the biological receptor would not occur through a charge transfer mechanism.

In order to have a insight on the main atomic contributions for the HOMO and LUMO, we decided to investigate the HOMO and LUMO plots (which are presented in Figure 3) for the compounds under study.

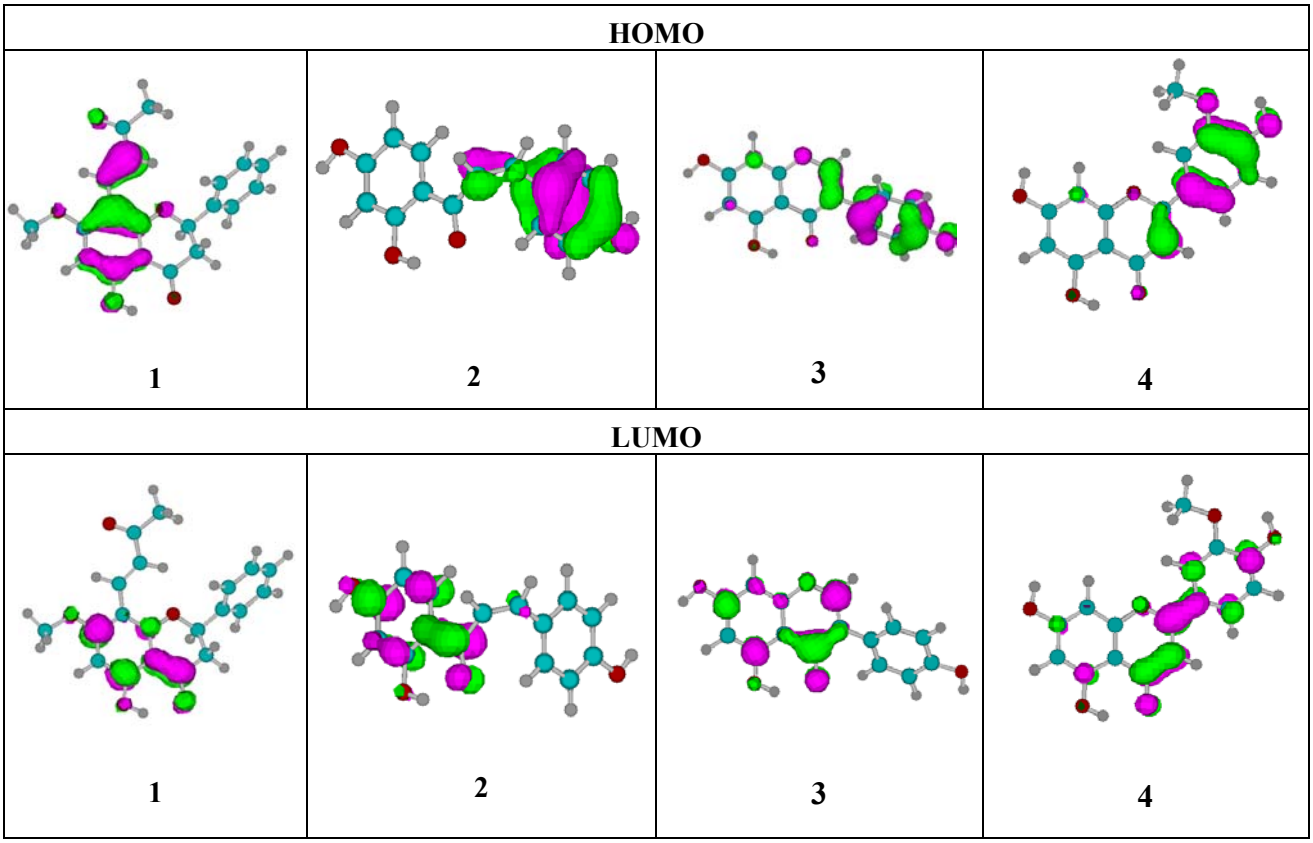

Figure 3. HOMO and LUMO plots for the flavonoid compounds studied.

From Figure 3 we can see that the atomic contributions for HOMO are very similar in the compounds 2, 3 and 4, i.e. their main contributions are localized at the substituents of ring C and along the bond between the atoms 2 and 3 (see Figure 1). The only exception is compound 1, as its main contributions for HOMO are localized at ring A. Regarding the atomic contributions for LUMO, we can see that the compounds 1, 2 and 3 have their main contributions localized at ring A and in the C=O bond, while the main contributions in compound 4 are localized at ring C.

The atomic charges were also calculated with the aim to investigate some influence of this electronic property in the chemopreventive activity of the flavonoid compounds studied. Figure 4 displays the net charges at each atom for the four compounds under study. From the atomic charges showed in Figure 4, we can see that is hard to find a correlation between the calculated atomic charges and the biological activity presented by the compounds.

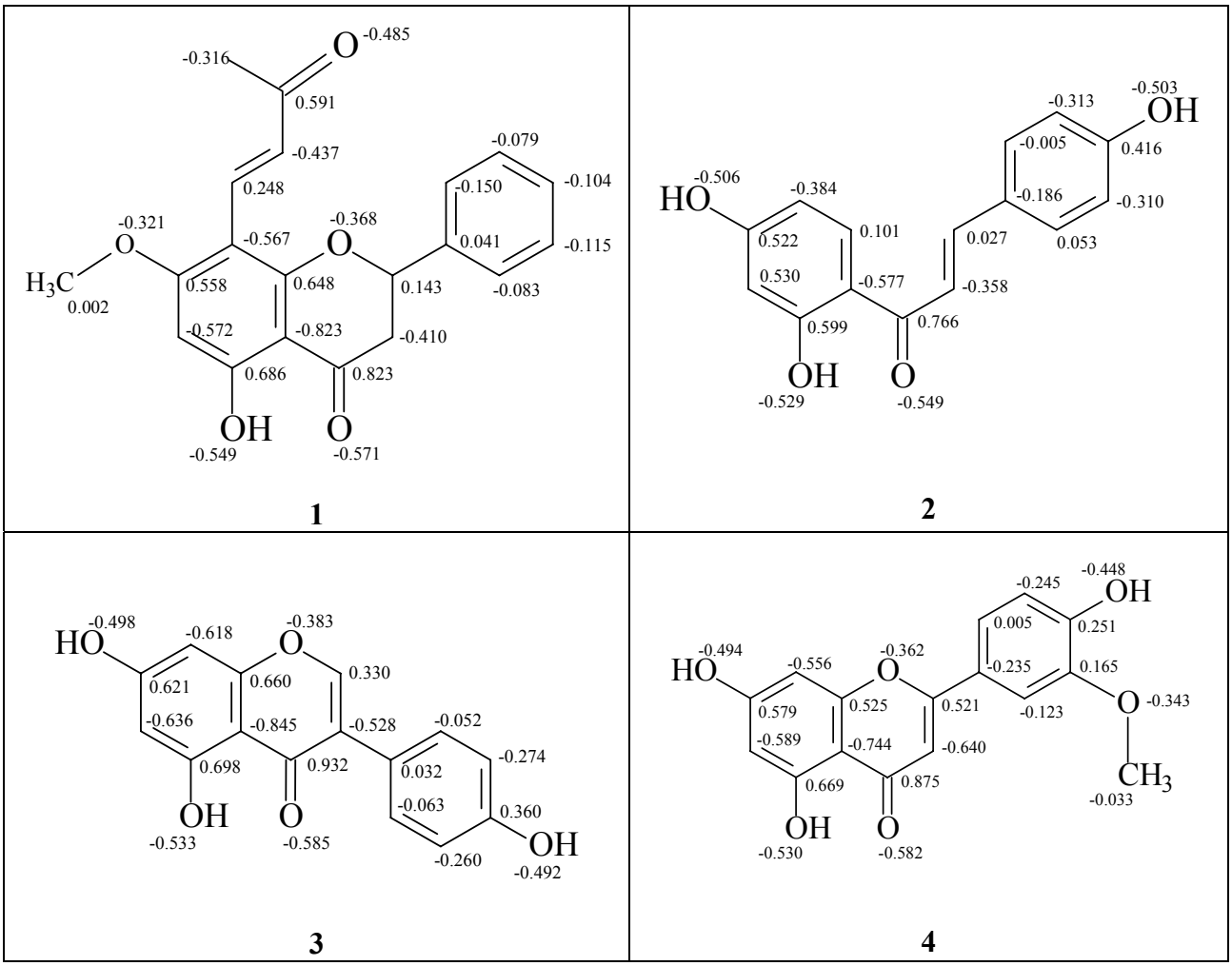

Figure 4. Atomic charges for the flavonoid compounds studied.

In fact, is hard to connect some specific charge variations in the compounds (see Figure 4) with their chemopreventive activity. From the calculated atomic charges showed in Figure 4, we can also construct the molecular electrostatic potential (MEP) maps (which give us a better overview of the electronic density of the whole molecule), then we decided to obtain these MEP maps in order to see if with them we could find some differences among the flavonoid compounds studied that could be correlated with their biological activity.

The MEP maps are displayed in Figure 5, and from them we can see that there are negative potential regions around all oxygen atoms of the compounds, indicating an excess of negative charge in these regions. Therefore, interactions with a positively charged region of a molecule (receptor) can occur with these negatively charged regions of the compounds; but from our HOMO and LUMO analysis, this possibility is not so strong. So, from the HOMO and LUMO plots, the calculated atomic charge values and from the MEP maps, we can conclude that the chemopreventive activity presented by the flavonoid compounds studied has probably no

correlation with the electronic properties of these compounds.

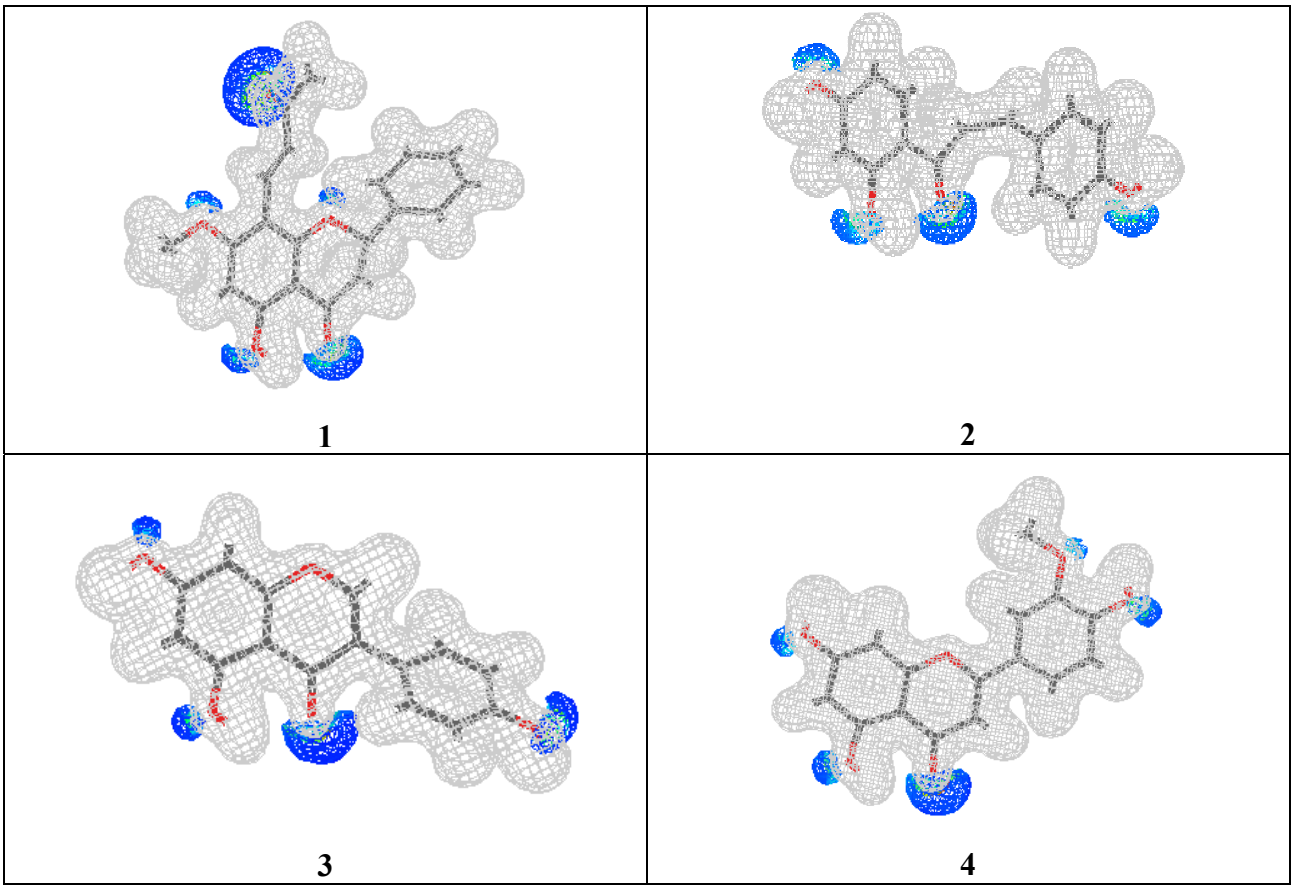

Figure 5. MEP maps for the flavonoid compounds studied. The blue surface corresponds to -50 kcal/mol (negative charge density) and the gray one corresponds to 50 kcal/mol (positive charge density).

On the other hand, it is also interesting to notice that the most active compound (compound 2) do not have the C ring, while the other compounds have it. From this fact we can say that the formation of the C ring probably causes the decrease of the chemopreventive activity presented by the compounds. Another important structural feature observed was the position of the B ring, as this ring appears in the position 2 or in position 3 in the compounds 1, 3 and 4, and this variation seems to influence the chemopreventive activity, as compounds with the C ring in the position 2 (compounds 1 and 4) have similar CD values, while the compound 3 has the C ring in the position 3 and its CD value is the highest one (see Table 1). So, we can conclude that structural aspects can be the more relevant effects (instead of the electronic ones) to be correlated with the chemopreventive activity presented by the flavonoid compounds studied in this work.

4 CONCLUSIONS

A theoretical study on the chemopreventive activity of some flavonoid compounds shows that this biological activity has probably no correlation with the electronic properties of these compounds.

The flavonoid compounds studied were classified as having a moderate electron-accepting and electron-donating character, and a comparison between the chemical structures of the compounds and their biological test data shows that structural aspects can be the more relevant effects (instead of the electronic ones) to be correlated with the chemopreventive activity presented by the flavonoid compounds studied in this work.

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