INTRODUCTION

The rapid development of computational techniques finds its way to be very helpful for related sciences, e.g. for chemistry to predict physicochemical properties of compounds. According to quantum chemistry, a molecular system is described by a wavefunction which can be found by solving the Schrödinger equation:

$$\hat{H} \Psi = E \Psi$$

In the end, this equation will describe the positions of the nuclei and electrons in the system. Quantum chemistry should be applied for ‘small system’, which can be treated at a very high level when electronic properties are sought (electric moments, polarizabilities, shielding constants in NMR, etc) [1].

Energy is one of the most important parameters in science. All computational chemistry techniques define energy such that the system with the lowest energy is the most stable. In formulating a mathematical representation of molecules, it is necessary to define a reference system that is defined as having zero energy. This zero of energy for ab initio or density functional theory (DFT) corresponds to having all nuclei and electrons at an infinite distance from one another. Most empirical methods use a valence energy that corresponds to having the valence electrons removed and the resulting ions at an infinite distance. Most molecular mechanics methods use stainless molecule as zero energy [2].

In this work, we confirmed the accuracy of computational technique prediction on physicochemical properties (log P) and spectrum (ultraviolet-visible, 1H-NMR, 13C-NMR) of quercetin (fig. 1a), glucosamine (fig. 1b), and andrographolide (fig. 1c) by comparing it to laboratory analysis. Therefore, computational calculations could be used to simplify and shorten the long process of analytical works in the laboratory. These three compounds were selected to represent molecule with aromatic and carbonyl chromophores which have $\pi \to \pi^*$ and $n \to \pi^*$ electronic transitions (quercetine), a molecule which lacks of the chromophore (glucosamine), and molecule without aromatic chromophore (andrographolide). These three compounds have been proven to show anti-inflammatory activity in animals [3-8].

Fig. 1: Chemical structures of quercetine (a), glucosamine (b), PITC-glucosamine (c), and andrographolide (c)
MATERIALS AND METHODS

Methods

Hardware, programs, and instruments

A personal computer equipped with Intel® Core™ i5-450M (2.40 GHz, Cache 3 MB), Intel HM55, 4 GB DDR3 SODIMM PC-8500 of memory, ATI Mobility Radeon HD 5430 512MB, 500 GB Serial ATA 7200 RPM hard drive, was used for computational technique.

Programs used were: Hyper Chem, ChemBio Office®ultra 2010 (trial version) with pre-installed add Gamsess Client® Pro for ab initio calculation, CS MOPAC®Pro for semiempirical calculation, Mechanics®Pro for molecular mechanics calculation.

Instruments used for physicochemical analysis were: ultraviolet-visible spectrophotometer (SPECORD200-Analytic Jena), 1H-NMR spectroscopy (Agilent 500 MHz), 13C-NMR spectroscopy (Agilent 125 MHz).

Chemicals

Standards used were: quercetine (Sigma-Aldrich), glucosamine (DongCheng Biochemical), and andrographolide (Sigma-Aldrich). Chemicals for solvents and reagents were: methanol (Merck), ethanol (Merck), n-octanol (Merck), ether (Merck), sodium acetate (Merck), phenylisothiocyanate (PITC) (Merck).

Data Preparations

Structures of quercetin (http://www.chemspider.com/4444051), glucosamine (http://www.chemspider.com/388352), and andrographolide (Sigma-Aldrich) were downloaded from ChemSpider.

Geometry optimisation for ab initio was performed using Hartree-Fock (HF) with central field approximation. The function of coordination used was a linear combination of Slater exp (-x) with STO-3G basis set.

Geometry optimisation for the semiempirical method was performed using neglect of diatomic differential overlap (NDDO) approach with AM1 parameter. Geometry optimisation for molecular mechanics method was performed using MM2 force field.

Log P and spectrum were calculated and predicted using Hyper Chem, ChemBio Office®ultra 2010. Data obtained were compared with laboratory results and calculated their accuracy by employing mean absolute deviation (MAD), mean square error (MSE), mean forecast error (MFE), and mean absolute percentage error (MAPE) parameters.

Laboratory section

Log P was determined by dissolving the compounds in a mixture of n-octanol and water and measuring the absorbance of the compounds in each solvent.

RESULTS AND DISCUSSION

Geometry optimisation of quercetin, glucosamine, and andrographolide showed that the lowest energy value was obtained by ab initio method. According to the theory, it could be concluded that in this state the molecules are at their most stable conformations.

UV spectra of the compounds were determined using 10 ppm of each compound in n-octanol and water, and log P were determined by measuring the absorbance of the compounds in each solvent. According to Woodward-Fieser, \( \lambda_{\text{max}} \) could be predicted by determining the base value of the compound and adding the contribution value of its substituents [9].

For quercetin (fig. 1a), the base structure is 6-membered ring enone (215 nm) with –OH at α-position as the substituent (35 nm), hence its theoretical maximum is 250 nm. fig. 3 shows UV spectra of quercetin in n-octanol \( \lambda_{\text{max}} \) 255 nm and in water \( \lambda_{\text{max}} \) 260 nm. The difference of the maxima is caused by the solvent.

Due to its lack of chromophores, glucosamine (fig. 1b) was reacted with phenylisothiocyanate, prior to be spectrophotometrically measured. The reaction produces a PITC-glucosamine (fig. 1c) which according to the work of Tekko and colleagues gave maximum at 245 nm [10], compared to our work which is 240 nm (fig. 4), whereas the analytical work of Shen for PITC-glucosamine was set at 254 nm [11].

Fig. 2: Potential energy against time during geometry optimisation

Fig. 3: UV spectra of quercetine in n-octanol (a) and in water (b)
Fig. 4: UV spectrum of derivates of PITC-glucosamine

The base structure of andrographolide (Fig. 1d) is 5-membered ring enone (202 nm), with a ring residue at α-position as the substituent (10 nm) and an alkyl substituent at β-position (12 nm), that add up to 230 nm, theoretically.

Fig. 5: UV spectra of andrographolide in n-octanol (a) and in water (b)

Fig. 5 shows UV spectra of andrographolide in n-octanol λ_max 220 nm and in water λ_max 224 nm. The difference of the maxima is caused by the solvent.

Table 1: Determination of Log P

| Compounds      | Experimental result (A_t) | Prediction (F_t) | Error prediction test | Correlation test |
|----------------|---------------------------|------------------|-----------------------|------------------|
|                |                           | I^P               | II^P                  | III^P            | MAD  | MSE  | MFE  | MAPE (%) | r-value | p-value |
| Quercetine     | 1.45                      | 1.50             | 1.50                  | 1.50             | 0.19  | 0.06 | 0.16 | 8.62      | 0.995   | 0.05    |
| Glucosamine    | -2.04                     | -2.18            | -2.18                 | -2.18            | 2.12  | 2.12 | 2.12 | 2.12      | 2.12    |         |
| Andrographolide| 2.51                      | 2.12             | 2.12                  | 2.12             | 2.12  | 2.12 | 2.12 | 2.12      | 2.12    |         |

^1: ab initio; ^2: semiempirical; ^3: molecular mechanics

Where:

\[ M_A = \frac{1}{n} \sum \left( A_t - F_t \right) \]

\[ M_S = \frac{1}{n} \sum \left( A_t - F_t \right)^2 \]

\[ M_F = \frac{1}{n} \sum \left( A_t - F_t \right) \]

Determination of log P showed there is no difference of log P value calculated by the three methods; furthermore, it was proven that there is a good accuracy and correlation between computational calculation and the experimental result (Table 1).

Determination of λ_max showed there is a good accuracy and correlation between computational calculation (ab initio and semiempirical) with experimental results (Table 2). NMR spectra predicted, and experiments were showed in Fig. 6-8 and Table 3, 4.

Table 2: Determination of λ_max

| Method          | Compound      | Experimental result (nm) | Computational calculation (nm) | Error prediction test | Correlation test |
|-----------------|---------------|--------------------------|-------------------------------|-----------------------|------------------|
|                 |               |                          |                               | MAD                   | MSE  | MFE  | MAPE (%) | r-value | p-value |
| Ab initio       | Quercetine    | 256                      | 253                           | 2.67                  | 8.67 | 2.67 | 1.10      | 0.997   | 0.044  |
|                 | Glucosamine   | 240                      | 236                           |                       |     |     |           |         |         |
|                 | Andrographolide| 222                      | 221                           |                       |     |     |           |         |         |
| Semi empirical  | Quercetine    | 256                      | 250                           | 6.67                  | 45.33| 6.67 | 2.79      | 0.997   | 0.043  |
|                 | Glucosamine   | 240                      | 232                           |                       |     |     |           |         |         |
|                 | Andrographolide| 222                      | 216                           |                       |     |     |           |         |         |
| Molecular       | Quercetine    | 256                      | 224                           | 28.67                 | 830  | 28.67| 11.99     | 0.979   | 0.129  |
| mechanics       | Glucosamine   | 240                      | 215                           |                       |     |     |           |         |         |
|                 | Andrographolide| 222                      | 193                           |                       |     |     |           |         |         |
Table 3: 1H-NMR spectra

| Compound       | Error prediction test | Correlation test |
|----------------|-----------------------|------------------|
|                | MAD \(^a\)  | MSE \(^b\)  | MFE \(^c\)  | MAPE \(^d\) | r-value | p-value |
| Quercetine     | 0.86        | 1.93       | -0.35      | 10.15\%    | 0.940    | 0.001   |
| Glucosamine    | 0.79        | 1.09       | 0.66       | 26.71\%    | 0.919    | 0.001   |
| Andrographolide | 0.55        | 0.43       | 0.50       | 18.20\%    | 0.966    | 0.001   |

Table 4: 13C-NMR spectra

| Compound       | Error prediction test | Correlation test |
|----------------|-----------------------|------------------|
|                | MAD \(^a\)  | MSE \(^b\)  | MFE \(^c\)  | MAPE \(^d\) | r-value | p-value |
| Quercetine     | 0.63        | 0.60       | 0.02       | 0.48\%     | 0.999    | 0.001   |
| Glucosamine    | 3.20        | 19.32      | -1.53      | 4.32\%     | 0.961    | 0.002   |
| Andrographolide | 0.92        | 2.31       | -0.57      | 3.23\%     | 0.999    | 0.001   |

Fig. 6: 1H-NMR (a) and 13C-NMR (b) spectra of quercetin by computational prediction (upper) and laboratory experiment (lower)

Fig. 7: 1H-NMR (a) and 13C-NMR (b) spectra of glucosamine by computational prediction (upper) and laboratory experiment (lower)
CONCLUSION

There is a positive correlation between computational \textit{ab initio} calculation method with experimental results in predicting log P and spectrum of quercetin, glucosamine, and andrographolide. This approach can be adapted to a wider range of compounds.

CONFLICTS OF INTERESTS

Declared none

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How to cite this article

• Sandra Megantara, Mutakin Mutakin, Jutti Levita. Prediction of log P and spectrum of quercetin, glucosamine, and andrographolide and its correlation with laboratory analysis. Int J Pharm Pharm Sci 2016;8(11):33-37.