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REGULAR ARTICLE

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Quantum chemical studies on structure activity relationship of natural product polyacetylenes

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Abstract An extract of the roots of *Levisticum officinale* L. (Apiaceae) exhibited significant antimycobacterial activity against *Mycobacterium fortuitum*, where diacetylene compounds were identified as the active components in this extract. In contrast, polyacetylenes isolated from different sources surprisingly exhibited no anti-mycobacterial activity. Additionally, a whole series of furanocoumarin ethers of the polyacetylene falcarindiol exhibited anti-proliferative properties. We have studied the relationship between the electronic properties and biological activity of these structurally related compounds and a good qualitative correlation between predicted lipophilic parameters and activity has been established.

Keywords Antimycobacterial activity · Polyacetylenes · Falcarindiol · log *P*

1 Introduction

Polyacetylene natural products are widespread in the plant families Asteraceae and Apiaceae exhibit many biological activities. Some of these compounds are highly poisonous such as cicutoxin from water hemlock (*Cicuta virosa*). Recently, the toxicity of these compounds has been shown to be due to their ability to bind to GABA-gated Cl^- channels of GABA receptors, and these channels play an important role in the acute toxicity of these compounds [1].

Certain diacetylenes have been found to have antimycobacterial [2], anti-staphylococcal [3], and anti-proliferative activities [4]. Polyacetylenes are also synthesized *de novo* as phytoalexins [5] and it is highly likely that these compounds are produced by plants as part of their chemical defense against microbes in their environment.

Our interest in this class of natural product started with the evaluation of members of the plant family Apiaceae with a view to discover and characterize new classes of antibacterial agents from plants [3,6,7]. Certain compounds, particularly falcarindiol display moderate bacterial, and mammalian cytotoxicity and are active against multi-drug resistant (MDR) strains of *Staphylococcus aureus* which is a continuing problem in the clinical setting. We have characterized a number of these agents to date and have noticed that simple changes to the polyacetylene core dramatically influence biological activity.

Significant antimycobacterial activity against *Mycobacterium fortuitum* was attributed to the active components 3(*R*)-falcarinol [3(*R*)-(-)-1,9-heptadecadien-4,6-diin-3-ol] (panaxynol) **1** and 3(*R*)-8(*S*)-falcarindiol [3(*R*)-8(*S*)-(+)-1,9-heptadecadien-4,6-diin-3,8-diol] **2** from an extract of the roots of *Levisticum officinale* L. (Apiaceae) [6]. 3(*R*),8(*R*)-dehydrofalcarindiol **3** and 1,3 *R*,8 *R*-trihydroxydec-9-en-4,6-yne **4** were inactive, while a whole series of polyacetylenes exhibited inhibitory activity against MK-1 cell growth (panaxynol **1**, (9Z)-l,9-heptadecadiene-4,6-diyne-3,8,11-triol **5**, 8-acetoxy-heptadeca-1,9-diene-4,6-diyne-8-ol **6**, and japoangelols A, B, C and D **7**, **8**, **9** and **10**, respectively) [8].

This paper describes an evaluation of the calculated molecular properties of a series of polyacetylenes (Scheme 1) in an attempt to explain differences in bioactivity, particularly as cytotoxic agents against bacterial and mammalian cell lines.

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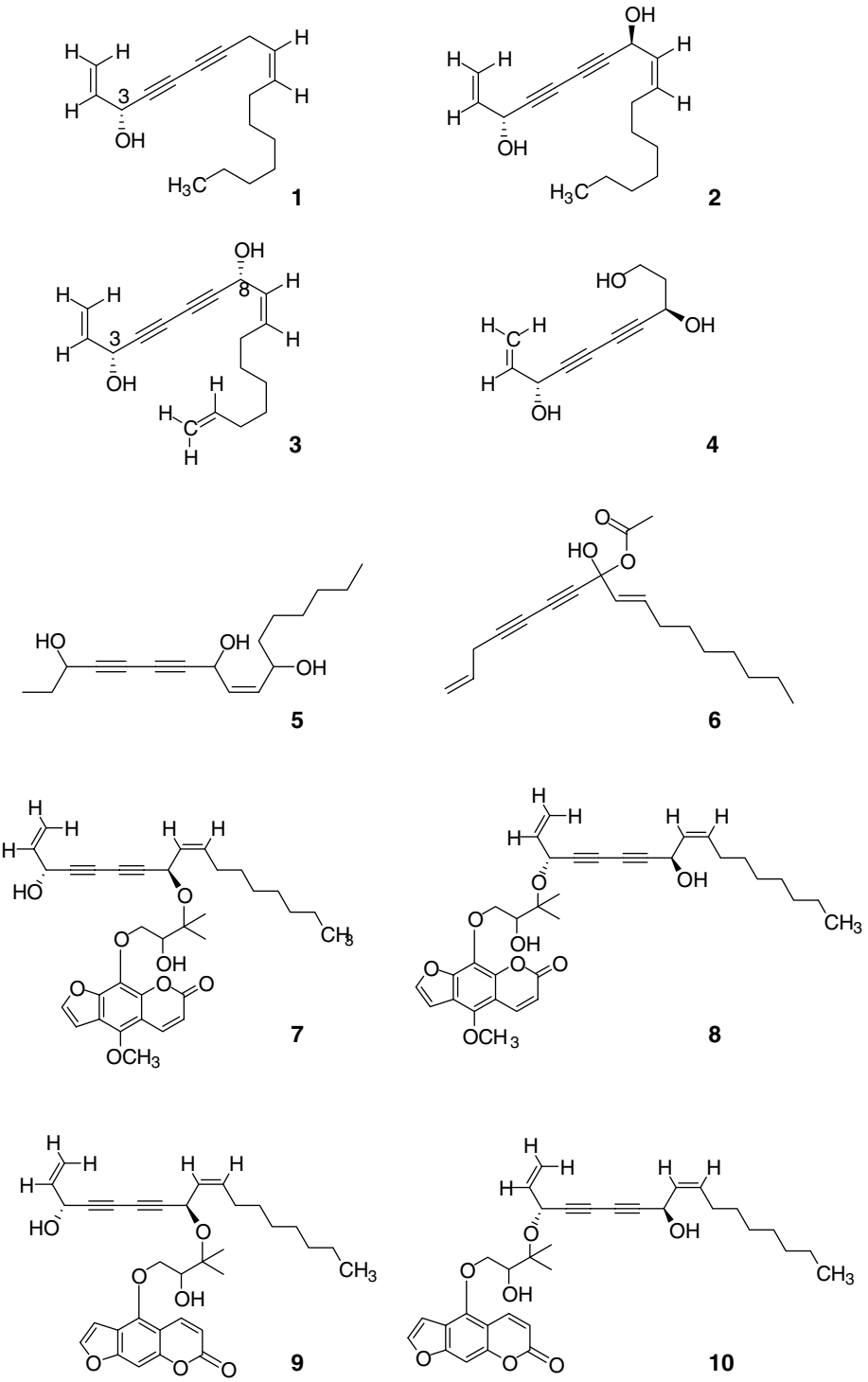

Scheme 1

2 Computational methods

2.1 Model building and conformational analysis

The initial structures of a series of polyacetylenes were built and saved as mol2 files by Chemoffice Ultra 7.0.0 [9]. These structures were imported into MacroModel [10], atom and bond types were adjusted and minimized with the MMFFs force field parameters [11]. The generalized Born/surface area (GB/SA) continuum solvent model for H_2O [12] implemented in MacroModel was used to simulate an aqueous environment, with a constant dielectric function ($\varepsilon = 1$). An extended non-bonded cutoff (van der Waals 8 Å; electrostatics 20 Å) was used.

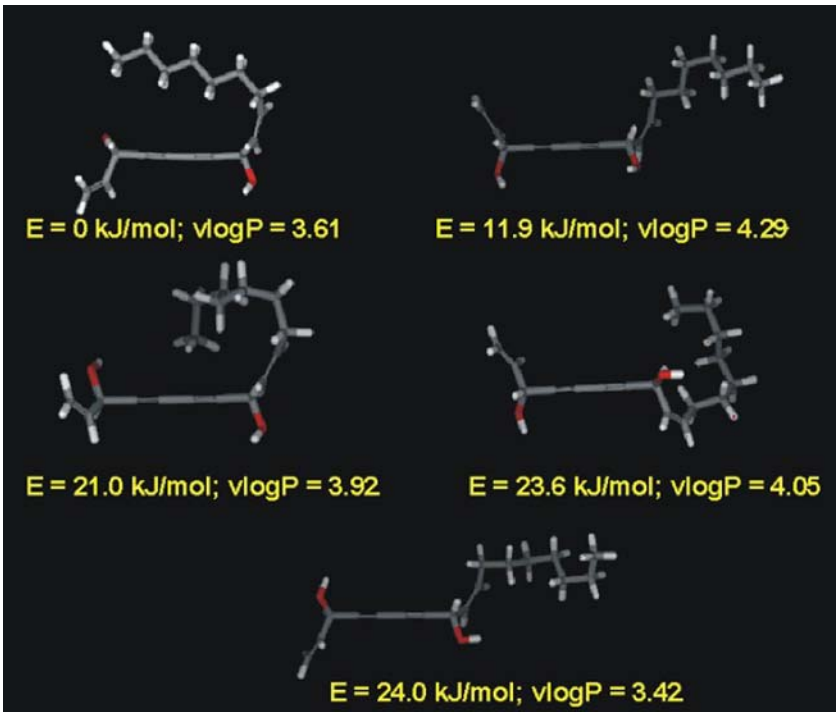

Fig. 1 The representative structures of **2** with the virtual log P (v log P) and relative energies (E) calculated using the HF/3-21G(d) basis set

Using the optimized structures, a systematic conformational search on each molecule was performed. All compounds used 500 step Monte Carlo conformational analysis, with the energy cut off generally set to $\Delta E = 10$ kJ/mol above the lowest energy conformation. The ensembles of generated structures were clustered and analyzed using the cluster analysis program Xclusterl [12]. Representative structures for each molecule were selected and subjected to *ab initio* calculation using Gamess US software and HF/3-21G(d) basis set [13].

2.2 Calculation of molecular properties

Molecular lipophilicity potential (MLP) is a structure–property descriptor that visualizes the lipophilic properties of the molecule on its three-dimensional (3D) surface and was calculated by projecting the Broto–Moreau lipophilicity atomic constants on the molecular surface [14]. The virtual log P, Broto log P and lipole, of all single molecules and complexes were evaluated by VegaZZ software [15, 16].

The correlations between different calculated values and activities were examined by the Gretl software package [17].

3 Results and discussions

The conformational, electronic, and molecular properties were studied to examine the possible relationship between structure and broad cytotoxic activity on several different targets, although the mechanism of action of polyacetylenes is still not known.

The conformational space of all compounds was examined using a Monte Carlo conformational search and five representative structures were fully optimized using an HF/3-31G(d) basis set. This produced a set of stable structures for each compound, with some conformations that could be adopted. The conformational flexibility of molecule **2** is shown in Fig. 1 as an example, and the energies calculated by the *ab initio* method are given relative to the most stable conformation. It was noticed that lipole and virtual log P depend on the conformation of a molecule, and the predicted values of virtual log P (v log P) from **2** are also depicted in Fig. 1. The virtual log P varies in a wide range from 3.42 to 4.29, and depends on the spatial arrangement of the terminal double bond and the hydrocarbon chain. The lowest value of virtual log P was observed when those two moieties are in a *trans* conformation, while the *cis* arrangement will result in high virtual log P values. The arrangement of other groups of the molecule will contribute to the variations of the virtual log P within extremes. Since it is not known which conformation is bioactive, the average values of lipole and virtual log P were used in further calculation. The most stable conformation was selected from five representative conformations of each molecule and used in calculation of other molecular descriptors used in this study.

3.1 Antibacterial activity of polyacetylenes

The molecular properties of the set of molecules with known antibacterial activity are shown in the Table 1. Although this set is small, a trend was observed in which the antibacterial

Table 1 Calculated molecular descriptors log *P*, HOMO, LUMO, their difference, dipole moment, and antibacterial activity against *Mycobacterium fortuitum* from [1,2]

Molecule	log *P*	lipole	Virtual logp *P*	HOMO(eV)	LUMO(eV)	Difference(eV)	Dipole(Db)	LogMIC(exp)	LogMIC (calc)
1	4.762	6.530	5.180	−9.2286	3.550	12.779	2.044	4.80	4.59
2	3.409	4.206	3.860	−9.3744	3.318	12.693	1.731	4.19	4.41
3	03.172	4.717	3.736	−9.3204	3.413	12.733	4.529	<3.30	3.37
4	−0.249	2.073	0.232	−9.6066	3.208	12.814	3.319	<3.15	3.06

MIC is defined as a molar minimum inhibitory concentration

Table 2 Calculated molecular descriptors log *P*, HOMO, LUMO, their difference, dipole moment, and inhibitory activity against MK-1 cell growth from [3]

Molecule	log *P*	lipole	Virtual logp *P*	HOMO(eV)	LUMO(eV)	Difference(eV)	Dipole(Db)	LogMIC(exp)	LogMIC (calc)
1	4.762	6.530	5.180	−9.2286	3.550	12.779	2.044	5.92	5.71
5	2.293	3.337	3.049	−9.5526	3.1185	12.671	2.198	5.10	4.98
6	3.672	3.809	4.804	−9.2961	3.4209	12.717	1.836	4.97	5.07
2	3.409	4.206	3.860	−9.3744	3.3183	12.693	1.731	4.91	5.17
7	8.705	2.010	7.330	−8.5428	1.7469	10.290	6.068	4.83	4.75
8	8.705	1.843	6.528	−8.1135	1.9953	10.109	11.075	4.90	4.69
9	8.564	3.323	8.090	−8.7156	1.6902	10.406	7.465	4.87	5.07
10	8.564	2.389	7.251	−8.6886	1.7199	10.409	8.493	4.81	4.84

MIC is defined as a molar minimum inhibitory concentration

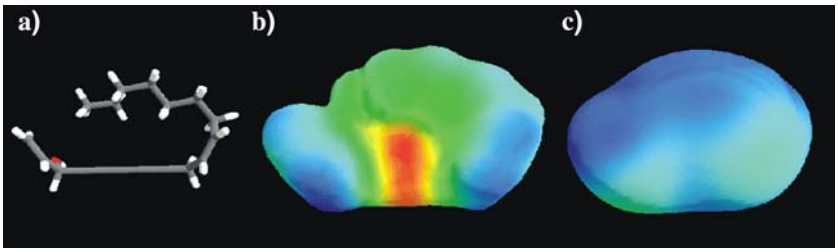

Fig. 2 **a** Lowest energy conformation of **1**, **b** map of electrostatic potential, **c** map of lipophilicity potential

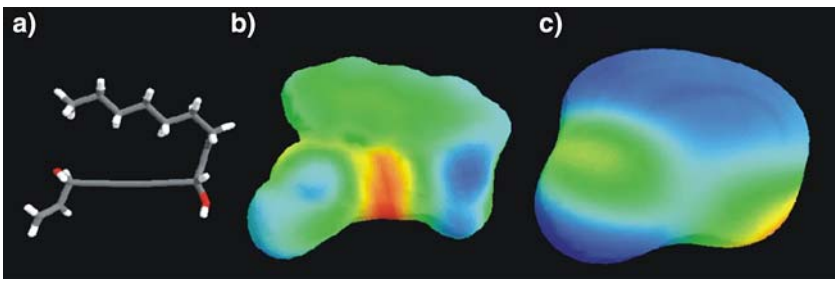

Fig. 3 **a** Lowest energy conformation of **2**, **b** map of electrostatic potential, **c** map of lipophilicity potential

activity decreases with the decreasing log *P*. This is expected since it is known that the log *P* is usually correlated with a biological activity [18]. It was also found that biological activity could be correlated to predict dipole moments. The correlations were tested using the Mixed Approach method [19] and regression analysis. Results of the regression analysis are shown in Eq. (1).

$$\text{LogMIC} = 4.3(\pm 0.6) + 0.21(\pm 0.09) \times \log P - 0.35(\pm 0.15) \times \text{Dipole} \quad (1)$$

$$n = 4,\ r = 0.94,\ F = 8.45,\ s = 0.77,$$

where n is number of molecules, r is correlation, F is Fisher's significance factor and s is the standard deviation. It has to be considered that the data set is small and this result might not be statistically significant. Although the fit is good (Table 1), these correlations should be treated with caution, especially since the small difference in log *P* between **2** and **3** does not fully explain the different activity. Therefore maps of electrostatic and lipophilicity potential were compared for

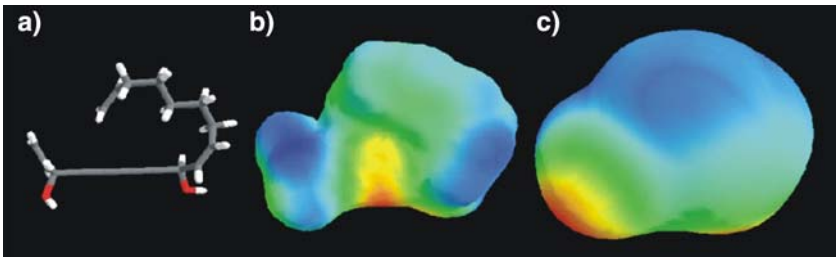

Fig. 4 **a** Lowest energy conformation of **3**, **b** map of electrostatic potential, **c** map of lipophilicity potential

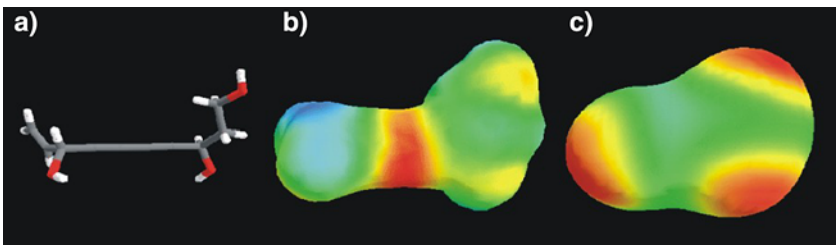

Fig. 5 **a** Lowest energy conformation of **4**, **b** map of electrostatic potential, **c** map of lipophilicity potential

molecules **1**–**4** (Figs. 2, 3, 4, 5, respectively). It is noticeable that the polarity and the lipophilicity of the substituents on the diacetylene moiety are different for all molecules. The higher polarity of the surface of the substituent may explain the lower antimicrobial activity of the molecule. Since the mechanism of cytotoxicity of the polyacetylene class is not known, we can hypothesize that the hydrophobic interaction between substituents on the polyacetylene structure and the target in the cell could play an important role for antibacterial activity.

3.2 Anti-proliferative properties

A separate study on a series of polyacetylene compounds was carried out to determine their inhibitory activity against MK-1 cell growth [8]. Molecular properties were calculated for each and correlated with their anti-proliferative activity. A correlation was observed between activity and calculated lipole. This suggests that the activity might depend on drug influx into cells [20]. Additionally, a correlation was observed between activity and LUMO energy. A similar correlation between the anti-proliferative activity and the E_{LUMO} was observed for bispyridinium compounds in the inhibition of choline kinase and the equation for predicting activity has given excellent results [21]. The mechanism of action was explained by the occurrence of either charge transfer or bipolar interactions. We believe that a similar mechanism could be involved in inhibition of MK-1 cell growth, since a correlation between activity and LUMO energies was observed. These correlations were tested using regression analysis and the results are given as Eq. (2).

$$\mathrm{LogMIC} = 4.4(\pm 0.25) + 0.24(\pm 0.083) \times \mathrm{lipole} - 0.06(\pm 0.15) \times \mathrm{LUMO}$$

$$n = 8,\ r = 0.76,\ F = 7.92,\ s = 0.22.$$

These findings do not have statistical significance and there is a problem to accurately calculate activities of most molecules, however it has predicted the activity of the highly potent molecule **1**. Therefore, these correlations should be used as qualitative indicators, rather than quantitative predictions of anti-proliferative activities.

Although it has been found that compound **3** is not active against bacteria, the log P and the LUMO energy value is similar to those of anti-proliferative compounds. Therefore, we have decided on the basis of this study to evaluate the anti-proliferative activity of **3**, for which we are awaiting results.

4 Conclusions

Molecular modeling analysis of a series of the polyacetylene compounds has allowed us to correlate molecular properties with experimental antibacterial and anti-proliferative data. Although the datasets are small for a full QSAR study, based on the reasoning described above, we have concluded that antimicrobial activity correlates well with the calculated log P and with the presence of a hydrophobic group on the substituent of the polyacetylene moiety. The anti-proliferative activity increases with increasing lipole in a series of polyacetylene compounds. A good correlation was also observed between activity and LUMO energies, suggesting that charge transfer might be involved in the mechanism of action. The preliminary QSAR study indicated that it is not possible to quantitatively predict activities, but a fairly good qualitative tendency between experimental activities and calculated values for molecular properties was found for this series of polyacetylenic natural products.

References

1. Uwai K, Ohashi K, Takaya Y, Ohta T, Tadano T, Kisara K, Shibusawa K, Sakakibara R, Oshima Y (2000) J Med Chem 43:4508–4515
2. Kobaisy M, Abramowski Z, Lermer L, Saxena G, Hancock REW, Towers GHN, Doxsee D, Stokes RW (1997) J Nat Prod 60:1210
3. Lechner D, Stavri M, Oluwatuyi M, Pereda-Miranda R, Gibbons S (2004) Phytochemistry 64:331
4. Bernart MW, Cardellina JH II, Balaschak MS, Alexander MR, Shoemaker RH, Boyd MR (1996) J Nat Prod 59:748
5. Elgersma DM, Liem JI (1989) Physiol Mol Plant P 34:545
6. Schinkovitz A, Stavri M, Gibbons S, Bucar F (2005) Fitoterapia (submitted)
7. Stavri M, Mathew KT, Gibson T, Williamson RT, Gibbons S (2004) J Nat Prod 67:892
8. Furumi K, Fujioka T, Fujii H, Okabe H, Nakano Y, Matsunaga H, Katano M, Mori M, Mihashi K (1998) Bioorg Med Chem Lett 8:93
9. ChemOffice Ultra 2002 (2001) nCambridgeSoft
10. Mohamadi F, Richards N, Guida W, Liskamp R, Lipton M, Caulfield C, Chang G, Hendrickson T, Still W (1990) J Comput Chem 11:440
11. Cramer CJ, Truhlar DG (1995) In: Lipkowitz KB, Boyd DB (eds) Rev Comp Chem, VCH, New York 6:1
12. Still WC, Tempczyk A, Hawley RC, Hendrickson T (1990) J Am Chem Soc 112:6127
13. Schmidt MW, Baldridge KK, Boatz JA, Elbert ST, Gordon MS, Jensen JH, Koseki S, Matsunaga N, Nguyen KA, Su SJ, Windus TL, Dupuis M, Montgomery JA (1993) J Comput Chem 14:1347
14. Gaillard P, Carrupt PA, Testa B, Boudon A (1994) J CAMD 8:83
15. Pedretti A, Villa L, Vistoli G (2002) J Mol Graph 21:47
16. Pedretti A, Villa L, Vistoli G (2003) Theor Chem Acc 109:229
17. Gretl 1.3.2 "GNU regression, econometrics and time-series library"
18. Cubini H (1995) In: Burger's medicinal chemistry and drug discovery, vol 1. Interscience, New York
19. Buchwald P, Bodor N (1998) Curr Med Chem 5:353
20. Mälkiä A, Murtomäki L, Urtti A, Kontturi K (2004) Eur J Pharm Sci 23:13
21. Campos J, del Carmen Núñez M, Rodríguez V, Entrena A, Hernández-Alcoceba R, Fernández F, Lacal JC, Gallo MA, Espinosa A (2001) Eur J Med Chem 36:215