



Antifungal potential of extracts, fractions and compounds from *Uvaria comperei* (Annonaceae) and *Oxyanthus unilocularis* (Rubiaceae)

Maurice Taboula Kayo , Marguerite Kamdem Simo , Maurice Tagatsing Fotsing , Emmanuel Talla , Sophie Laurent , Luce Vander Elst , Céline Henoumont , Emmanuel Yankep , Tamfu Alfred Ngenge , Rodrigue Keumoe , Alex de Theodore Atchade , Elisabeth Menkem Zeukóo , Modeste Lambert Sameza , Alain Roch , Robert Muller , Fabrice Fekam Boyom & Joseph Tanyi Mbafor

To cite this article: Maurice Taboula Kayo , Marguerite Kamdem Simo , Maurice Tagatsing Fotsing , Emmanuel Talla , Sophie Laurent , Luce Vander Elst , Céline Henoumont , Emmanuel Yankep , Tamfu Alfred Ngenge , Rodrigue Keumoe , Alex de Theodore Atchade , Elisabeth Menkem Zeukóo , Modeste Lambert Sameza , Alain Roch , Robert Muller , Fabrice Fekam Boyom & Joseph Tanyi Mbafor (2020): Antifungal potential of extracts, fractions and compounds from *Uvaria comperei* (Annonaceae) and *Oxyanthus unilocularis* (Rubiaceae), Natural Product Research, DOI: [10.1080/14786419.2020.1828409](https://doi.org/10.1080/14786419.2020.1828409)

To link to this article: <https://doi.org/10.1080/14786419.2020.1828409>



View supplementary material [↗](#)



Published online: 27 Nov 2020.



Submit your article to this journal [↗](#)



Article views: 8



View related articles [↗](#)



View Crossmark data [↗](#)



SHORT COMMUNICATION



Antifungal potential of extracts, fractions and compounds from *Uvaria comperei* (Annonaceae) and *Oxyanthus unilocularis* (Rubiaceae)

Maurice Taboula Kayo^{a,b}, Marguerite Kamdem Simo^{c,d}, Maurice Tagatsing Fotsing^a, Emmanuel Talla^e, Sophie Laurent^f, Luce Vander Elst^f, Céline Henoumont^f, Emmanuel Yankep^a, Tamfu Alfred Ngenge^e , Rodrigue Keumoe^c, Alex de Theodore Atchade^a, Elisabeth Menkem Zeukóo^c, Modeste Lambert Sameza^g, Alain Roch^f, Robert Muller^f, Fabrice Fekam Boyom^c and Joseph Tanyi Mbafor^a

^aLaboratory No. 3 of Organic Chemistry, Department of Organic Chemistry, Faculty of Science, University of Yaoundé I, Yaoundé, Cameroon; ^bDepartment of Chemistry, Higher Teacher Training College, University of Bamenda, Bambili, Cameroon; ^cLaboratory for Phytobiochemistry and Medicinal Plants Studies, Department of Biochemistry, Faculty of Science, University of Yaoundé I, Yaoundé, Cameroon; ^dDepartment of Biological Sciences, Faculty of Science, University of Maroua, Maroua, Cameroon; ^eDepartment of Materials Engineering/Chemical Engineering, School of Chemical Engineering and Mineral Industries, University of Ngaoundéré, Ngaoundéré, Cameroon; ^fNMR and Molecular Imaging Laboratory, Department of General, Organic and Biomedical Chemistry, Faculty of Medicine and Pharmacy, University of Mons, Mons, Belgium; ^gDepartment of Biochemistry, Faculty of Science, University of Douala, Douala, Cameroon

ABSTRACT

Phytochemical study of *Uvaria comperei* afforded an alkaloid, 8,9-dimethoxy-5H-phenanthridin-6-one (**1**), isolated and characterised (assignment of ¹H and ¹³C NMR) for the first time from a natural source along with two flavonoids, (2S)-5-hydroxy-7,8-dimethoxy-flavanone (**2**) and (2S)-7-hydroxy-5-methoxy-6,8-dimethylflavone (**3**). Clethric acid (**4**), oleanolic acid (**5**), β-sitosterol 3-O-β-D-glucopyranoside (**9**), β-sitosterol palmitate (**6**) and a mixture of stigmasterol (**7**) and β-sitosterol (**8**) were isolated from *Oxyanthus unilocularis*. The structures of these compounds were elucidated using modern spectroscopic techniques including 1D and 2D Nuclear Magnetic Resonance (NMR) Spectroscopy (¹H, ¹³C, ¹H-¹H COSY, HSQC, HMBC) and Mass Spectrometry. Some fractions and compounds from *Uvaria comperei* exhibited good antifungal activity against clinical isolates and standard strains of yeast species of *Candida* and *Cryptococcus* genera while extracts from *Oxyanthus unilocularis* displayed weak antifungal activity. The results obtained show that *Uvaria comperei* could be a potential source of antifungal drugs.

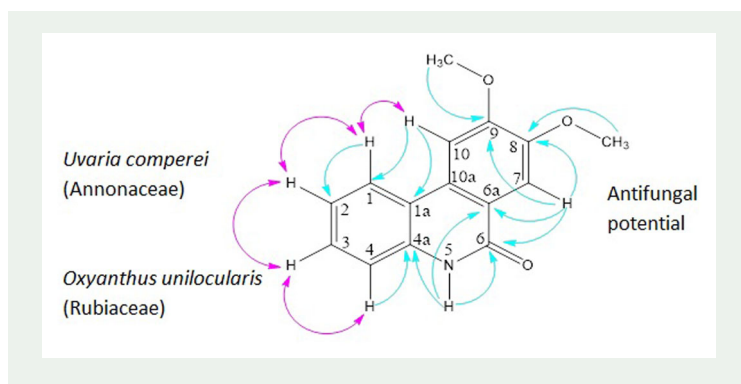
ARTICLE HISTORY

Received 20 July 2020

Accepted 12 September 2020

KEYWORDS

Uvaria comperei; *Oxyanthus unilocularis*; 8,9-dimethoxy-5H-phenanthridin-6-one; antifungal activity



1. Introduction

Microbial infections pose a health problem throughout the world, due to the resistance of bacteria against manufactured drugs, and plants can be used as possible sources of antimicrobial agents (Adenisa et al. 2000). Medicinal plants contain active principles which can be used as an alternative to cheap and effective herbal drugs against common bacterial, fungal and viral infections (Adesina et al. 2000; Kareru et al. 2008). *Oxyanthus unilocularis* and *Uvaria comperei* are two medicinal plants used to manage infectious diseases in Cameroon. *Oxyanthus* is a genus of plants belonging to Rubiaceae family best known in West Africa, plant of this genus occupy a prominent position in African traditional medicine (Tigoufack et al. 2012). It comprises 53 species, subspecies, varieties, forms and cultivars (Sonke and Cheek 1999). The leaves, bark or roots of *Oxyanthus unilocularis* are used for the treatment of fever, arthritis, swelling, pain, mild to moderate depression as well as for cutaneous and subcutaneous parasitic infestations (Nkeoua and Boundzana 1999; Nkeh-Chungag et al. 2010). The analgesic and anti-inflammatory properties of methanol extracts of the leaves and the bark of *O. unilocularis* have been reported (Nkeh-Chungag et al. 2010). The genus *Uvaria* belongs to the Annonaceae family, one of the biggest plant family comprising about 130 genera and species belonging to this genus are used in traditional medicine for the treatment of dysentery, wound, abdominal ache, malaria (Gina et al. 2014). The free-radical scavenging and antifungal activity of *U. comperei* extracts were reported (Simo et al. 2018). However, fractions and compounds responsible for the antifungal activity of this plant were not known.

This involves the evaluation of antifungal activity of fractions and compounds from *U. comperei* as well as chemical constituents from *O. unilocularis* and antifungal potential of its crude extracts.

2. Results and discussion

The synthesis of 8,9-dimethoxy-5H-phenanthridin-6-one was reported (Rigby and Laurent 1998; Chen et al. 2014) but without proper spectral assignments. The isolation and spectral characterisation of 8,9-dimethoxy-5H-phenanthridin-6-one (**1**) from a

natural source is reported here for the first time alongside (2S)-5-hydroxy-7,8-dimethoxyflavanone (**2**) and (2S)-7-hydroxy-5-methoxy-6,8-dimethylflavone (**3**) from the stem bark of *Uvaria comperei*. From the stem bark of *Oxyanthus unilocularis*, clethric acid (**4**), oleanolic acid (**5**), β -sitosterol 3-O- β -D-glucopyranoside (**9**), β -sitosterol palmitate (**6**) with a mixture of stigmasterol (**7**) and β -sitosterol (**8**) were isolated and characterised.

Compound **1**, was obtained as a yellow solid and melted between 295–297 °C. It gave a positive Dragendorff test suggesting that it is an alkaloid. The TOF-ESI-MS⁺ showed pseudo-molecular ion pic and dimer ion pic at *m/z* 256.3 and 511.3 attributable to $[M + H]^+$ and $[2M + H]^+$, respectively which is in accordance with the molecular formula $C_{15}H_{13}O_3N$, with 10 degrees of unsaturation. In the 1H NMR spectrum of compound **1** (see Table S1) in DMSO-*d*₆, 6 aromatic proton signals were observed and could be assigned to a 1,2,4,5-tetrasubstituted benzene and a 1,2-disubstituted benzene rings on the basis of 1H - 1H COSY spectrum and coupling patterns. The 1H NMR spectrum displayed two signals at δ_H 7.16 (1H, s, H-10) and 7.86 (1H, s, H-7) characteristic of a 1,2,4,5-tetrasubstituted benzene nucleus. It also exhibited a coupling pattern of 4 aromatic protons at δ_H 7.57 (1H, td, *J* = 7.63, 1.36, H-2), 7.61 (1H, td, *J* = 7.63, 1.36, H-3), 7.86 (1H, s, H-7), 7.99 (1H, dd, *J* = 7.63, 1.36, H-1) and 9.15 (1H, dd, *J* = 7.63, 1.36, H-4), confirming the presence of an *O*-disubstituted benzene ring. The remaining signals in the 1H NMR spectrum were protons of two methoxy groups at higher field [δ_H 4.04 (3H, s, H-OMe-9) and 4.06 (3H, s, H-OMe-8)] and a singlet at δ_H 10.86 showing no correlation with any carbons in HSQC spectrum that could be ascribed to an D₂O exchangeable proton of an NH group. The ^{13}C NMR spectrum (BB and DEPT) showed 13 carbon signals among which 2 methyls, 6 methines, and 7 quaternary carbons. At the low-field region of the ^{13}C NMR spectrum the presence of the quaternary carbon signal at δ_C 168.4 (C-6) was assigned to the carbonyl of an amide. The signals at δ_C 104.6 (CH, C-10), 109.6 (CH, C-7), 121.6 (C, C-10a), 123.3 (C, C-6a), 125.5 (CH, C-2), 125.9 (C, C-1a), 126.8 (CH, C-4), 127.5 (CH, C-3), 129.0 (CH, C-1), 135.1 (C, C-4a), 150.4 (C, C-9) and 154.2 (C, C-8) were attributed to aromatic carbons. At the up-field region the two deshielded methyl signals observed at δ_C 56.8 and 60.0 are characteristic of two methoxy groups. In the HMBC spectrum, the H-1 at δ_H 7.99 revealed a 2J correlation with C-2 (δ_C 125.5). The H-4 at δ_H 9.15 showed a 2J correlation with C-4a (δ_C 135.1). The amide proton (NH, δ_H 10.86) showed 2J correlations with C-4a (δ_C 135.1) and C-6 (δ_C 168.4); and a 3J correlation with C-6a (δ_C 123.3). The H-7 at δ_H 7.86 showed 2J correlations with C-6a (δ_C 123.3) and C-8 (δ_C 154.2); and 3J correlations with C-6 (δ_C 168.4) and C-9 (δ_C 150.4). The H-10 at δ_H 7.16 showed a 3J correlation with C-1a (δ_C 125.9) and a 4J correlation with C-1 (δ_C 129.0). The methoxy proton H-OMe-8 at δ_H 4.06 showed a 3J correlation with C-8 (δ_C 154.2). The methoxy proton H-OMe-9 at δ_H 4.04 showed a 3J correlation with C-9 (δ_C 150.4). On the basis of these data, the structure of compound **1** was established as 8,9-dimethoxy-5*H*-phenantridin-6-one. The 1H - 1H COSY spectrum of compound **1** showed the presence of a spin system due to a 1,4-disubstituted benzene ring. In fact, this spectrum exhibited correlations between H-1 and H-2, H-2 and H-3; and H-3 and H-2. Furthermore, 1H - 1H COSY spectrum of compound **1** showed the presence of a spin system due to a long-range correlation between the

proton H-10 at δ_{H} 7.16 and the proton H-1 at δ_{H} 7.99. Details of the spectra described here are summarised on Table S1.

2.1. Antifungal activity of extracts, fractions and compounds isolated

The results of the antifungal activity of fractions and compounds from *Uvaria comperei*, 8,9-dimethoxy-5*H*-phenanthridin-6-one (**1**), (2*S*)-5-hydroxy-7,8-dimethoxyflavanone (**2**) and (2*S*)-7-Hydroxy-5-methoxy-6,8-dimethylflavone (**3**) as well as the crude extracts from *O. unilocularis* are shown in Table S2. That table presents the MIC of fractions on yeasts. It indicated that the activity of fractions changes in function of yeast strain. The most active fractions with low MIC values were fraction 10–11, 12, 13–14, 8, 7, 19, 25–34 and 35 this in function of lower values of MIC, whereas, in function of spectrum of activity the most active fraction was fraction 7 followed by fractions 8, 19, 25–34, 10–11, 12, 13–14 and 35. Fractions 8, 10–11, 12, 19, 25–34, 7, 13–14 and 35 have the same statistically activity whereas it is significantly different from fractions 15–18 and 36–39 ($p \leq 0.05$). Non-polar fractions showed higher activity than polar fractions. The antifungal activity of fractions is higher than that of pure compounds. However, fractions 19, 25–34 and 35 afforded compounds **1**, **2** and **3** respectively. Interesting, each compound had same activity with the corresponding fraction from which it results. The ethyl acetate extract of *Oxyanthus unilocularis* showed better activity than other extracts from the plant.

After 48 h, MFC were determined and values were indicated in Table S3. Fraction 7 to fraction 35 were fungicidal on almost all yeast with MFC of 1.04 to 4.17 mg/mL. Fractions 56–62, 75–85, 155–163 were fungistatic (Table S4).

3. Conclusion

Fungal infections constitute a global health problem and many plants have been used in traditional medicine to manage this situation. *Uvaria comperei* and *Oxyanthus unilocularis* are two plants that are usually employed in traditional medicine to fight fungal and microbial infections. The chemical compounds contained in these two plants have been isolated and characterized in this study. Some of the compounds and fractions from *Uvaria comperei* as well as the extracts *Oxyanthus unilocularis* were tested on various fungal clinical isolates and standard strains and the results showed appreciable activity especially for fractions and compounds from *Uvaria comperei*. This study supports the ethnopharmacology application of *Uvaria comperei* as remedy for fungal diseases.

Acknowledgements

The authors are grateful to the *Commission Universitaire pour le Développement (CUD)* for providing a grant to Kayo Taboula Maurice to work at the Free University of Brussels, Belgium and University of Mons, Belgium in 2009, 2010 and 2011. We also thank Prof Pierre DUEZ and the Laboratory of Pharmacognosy, Bromatology and Human Nutrition of the Faculty of Pharmacy of the Free University of Brussels for hosting Kayo Taboula Maurice during his internships in Belgium.

Disclosure statement

No potential conflict of interest was reported by the authors.

ORCID

Tamfu Alfred Ngeenge  <http://orcid.org/0000-0001-6683-3337>

References

- Adesina SK, Idowu O, Ogundaini AO, Oladimeji H, Olugbade TA, Onawunmi GO, Pais M. 2000. Antimicrobial constituents of the leaves of *Acalypha hispida*. *Phytother Res*. 14(5):371–374.
- Chen YF, Wu YS, Jhan YH, Hsieh JC. 2014. An efficient synthesis of (NH)-phenanthridinones via ligand-free copper-catalyzed annulation. Supplementary information. Electronic Supplementary Material (ESI). *Org Chem Front*. 1:253–257.
- Gina F, Renata B, Souza L, Ari-de-Freitas H, Maas P, Pohlit A. 2014. Plants of the annonaceae traditionally used as antimalarials: a review. *Rev Bras Frutic*. 36(spe1):315–336.
- Kareru PG, Gachanja AN, Keriko JM, Kenji GM. 2008. Antimicrobial activity of some medicinal plants used by herbalists in Eastern Province of Kenya. *Afr J Trad Comp Altern Med*. 5:51–55.
- Nkeh-Chungag NB, Bekwa PCM, Ndebia JE, Kayo M, Mbafor TJ, Iputo EJ. 2010. Analgesic and anti-inflammatory properties of *Oxyanthus unilocularis*. *J Med Plants Res*. 4(10):932–939.
- Nkeoua G, Boundzana GC. 1999. Données sur les produits forestières non ligneux en Republique du Projet GCP/INT/679/EC Programme de partenariat. EC - FAO; pp. 95–96.
- Rigby JH, Laurent S. 1998. Addition of alkyl and aryl isocyanides to benzyne. *J Org Chem*. 63(19):6742–6744.
- Simo KM, Dongmo NMD, Sameza ML, Toghueo RK, Fekam FB, Foldi G. 2018. Cameroonian medicinal plants belonging to Annonaceae family: radical scavenging and antifungal activities. *Nat Prod Res*. 32(17):2092–2095.
- Sonke B, Cheek M. 1999. A new species of *Oxyanthus* (Rubiaceae-Gardeniinae) from Western Cameroon. *Kew Bull*. 55(4):889–893.
- Tigoufack BI, Ngnokam D, Tapondjou LA, Harakat D, Voutquenne L. 2012. Isolation and characterisation of sodium monocarboxylate ixoside salt from the stem bark of *Oxyanthus pallidus*. *Int J Biol Chem Sci*. 6(6):7007–7012.