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# Chemical constituents of gorgonian *Verrucella umbraculum* from the South China Sea

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## 1. Subject and source

Gorgonian coral of the genus *Verrucella* belongs to family Ellisellidae. The specimen of *Verrucella umbraculum* was collected off the coral reef of Weizhou island in the South China Sea, China, in September 2008, and was identified by Prof. Hui Huang, South China Sea Institute of Oceanology, Chinese Academy of Sciences, China. The voucher specimen (GX-WZ-2008003-3) was deposited in the Key Laboratory of Marine Drugs, Ministry of Education, Ocean University of China, Qingdao, China.

## 2. Previous work

To date, there have been no reports concerning the secondary metabolites of *V. umbraculum*. However, recently investigation on a species of the genus *Verrucella* revealed the presence of seven compounds, including junceellin, praelolide, cholesterol, (*E*)-*N*-2-(1,3-dihydroxy octadecan-4-en)-hexadecamide, thymine, thymidine and batyl alcohol (Wang et al., 2010).

## 3. Present study

The frozen specimen (2 kg, wet weight) was extracted with 95% EtOH three times (3000 mL × 3) at room temperature, and the solvent was evaporated *in vacuo*. The residue was partitioned in $H_2O$ and extracted with EtOAc three times. After removal

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of the solvent under reduced pressure, the EtOAc extract (9.0 g) was subjected to vacuum liquid chromatography (VLC) on silica gel and eluted with petroleum ether containing increasing amounts of EtOAc to afford 8 fractions (Fr.1–Fr.8). Fr.2 was isolated by column chromatography on silica gel eluted with petroleum ether–EtOAc (100:0 to 30:1), and then subjected to Sephadex LH-20 column eluted with petroleum ether–$CHCl_3$–MeOH (2:1:1) to obtain compounds **3** (29.0 mg), **5** (8.0 mg) and **6** (45.0 mg). Fr.3 was first subjected to repeated silica gel chromatography, and then purified by Sephadex LH-20 column ($CHCl_3$–MeOH 1:1) and reversed-phase silica gel chromatography to afford **1** (9.5 mg), **7** (8.3 mg) and **8** (10.0 mg). Fr.4 was applied to column chromatography on silica gel with petroleum ether–EtOAc (3:1), and further separated by Sephadex LH-20 column ($CHCl_3$–MeOH 1:1) to yield **2** (9.5 mg) and **4** (8.0 mg). Fr.5 was fractionated on a silica gel column, eluted with $CHCl_3$–MeOH (15:1), then purified by Sephadex LH-20 column (MeOH) and repeated reversed-phase silica gel chromatography to afford **9** (5.0 mg) and **10** (9.3 mg).

The structures of isolated compounds were identified on the basis of their $^1$H NMR, $^{13}$C NMR and ESI–MS spectra and by comparison with those reported literature data as suberoretisteroid A (**1**) (Zhang et al., 2005), suberoretisteroid B (**2**) (Zhang et al., 2005), suberoretisteroid C (**3**) (Zhang et al., 2005), 3,22,25-trihydroxy-16-24,20-24-bisepoxy-3$\beta$,16$\beta$,20*S*,22*R*,24*S*-cholest-5-ene (**4**) (Yang et al., 2005), reticulatic acid (**5**) (Yang et al., 2005), cholest-5-en-3$\beta$-ol (**6**) (Huang et al., 2004), (22*E*,24*S*)-5$\alpha$,8$\alpha$-epidioxy-24-methyl-cholesta-6,22-dien-3$\beta$-ol (**7**) (Ioannou et al., 2009), (22*E*,24*S*)-5$\alpha$,8$\alpha$-epidioxy-24-ethyl-cholesta-6,22-dien-3$\beta$-ol (**8**) (Ioannou et al., 2009), (24*R*)-24-methyl-5$\alpha$-cholest-7-ene-3$\beta$,5$\alpha$,6$\beta$-triol (**9**) (Madaio et al., 1989), and (22*E*,24*R*)-24-methyl-5$\alpha$-cholesta-7,22-dien-3$\beta$,5$\alpha$,6$\beta$-triol (**10**) (Liu et al., 2006) respectively (Fig. 1).

## 4. Chemotaxonomic significance

The present study reported the isolation and identification of ten known steroids. All the compounds were isolated from *V. umbraculum* for the first time. Of these secondary metabolites, compounds **1**–**5** were polyoxygenated steroids, and especially **1**–**4** were spiroketal steroids that possessed the rare 24-ketal function.

The polyoxygenated steroids **1**–**5** were characterized for the first time from the genus *Verrucella*. Interestingly, the occurrence of spiroketal steroids **2**–**4** in both *V. umbraculum* and *Gorgonella umbraculum* (Anjaneyulu et al., 2003, 2007; Subrahmanyam and Kumar, 2000), which belongs to the same family Ellisellidae, may indicate a close relationship on their chemical composition between the two genera and may be useful as chemotaxonomic markers for the family Ellisellidae. This finding confirms that the genera *Verrucella* and *Gorgonella* are closely related taxonomically. On the other hand, compounds **1** and **5** were obtained for the first time in the family Ellisellidae. Thus the finding of compounds **1**–**5** in the present investigation is a major contribution to chemotaxonomic studies of the family Ellisellidae.

The steroids **6**–**10** are common occurrence in corals. Compound **6** had been reported from the genus *Verrucella* (Wang et al., 2010), and **7** and **10** had been isolated from the genus *Junceella* of the same family Ellisellidae (Liaw et al., 2008; Anjaneyulu and Rao, 1997; Qi et al., 2004). Compounds **8** and **9**, however, are the first report in the genus *Verrucella* as well as in the family Ellisellidae.

To date, all previous studies (Anjaneyulu et al., 2003, 2007; Subrahmanyam and Kumar, 2000; Liaw et al., 2008; Anjaneyulu and Rao, 1997; Qi et al., 2004) and our investigations have revealed the dominant presence of steroids in the family Ellisellidae. These results indicate that polyoxygenated steroids, especially spiroketal steroids, could be characteristic constituents of the family Ellisellidae. According to the above results, the existence of ten steroids could partly provide a chemotaxonomic evidence to support the morphological classification and molecular systematics' research. The chemical investigation of *V. umbraculum* may be used as foundation for further chemotaxonomic studies on the genus *Verrucella* and even on the family Ellisellidae.

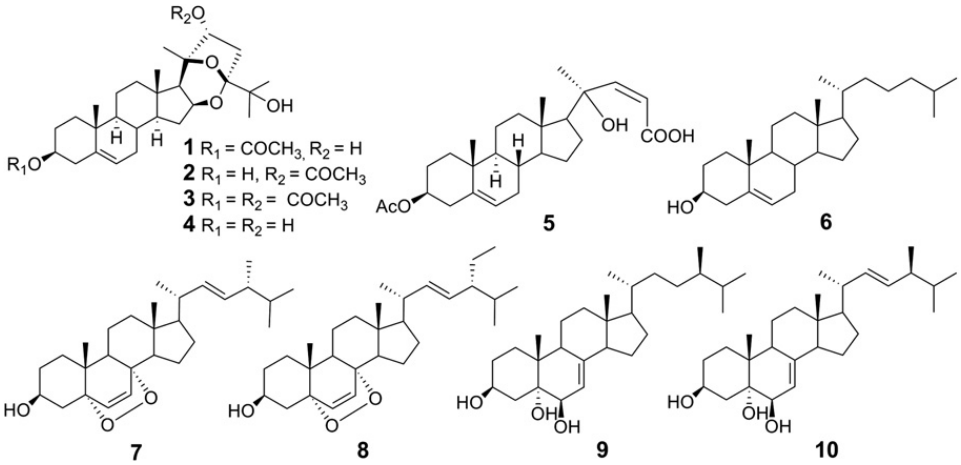


**Fig. 1.** Structures of compounds **1**–**10**.

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## References

Anjaneyulu, A.S.R., Rao, N.S.K., 1997. J. Chem. Soc., Perkin Trans. I 6, 959.
Anjaneyulu, A.S.R., Rao, V.L., Sastry, V.G., 2003. Nat. Prod. Rep. 17, 149.
Anjaneyulu, A.S.R., Rao, V.L., Sastry, V.G., Rao, D.V., Laatsch, H., 2007. Nat. Prod. Commun. 2, 131.
Huang, J.S., Li, Q.X., Wu, J., Zhang, S., 2004. Chin. Tradit. Herbal Drugs 35, 485.
Ioannou, E., Abdel-Razik, A.F., Zervou, M., Christofidis, D., Alexi, X., Vagias, C., Alexis, M.N., Roussis, V., 2009. Steroids 74, 73.
Liaw, C.C., Shen, Y.C., Lin, Y.S., Hwang, T.L., Kuo, Y.H., Khalil, A.T., 2008. J. Nat. Prod. 71, 1551.
Liu, J.Y., Huang, L.L., Ye, Y.H., Zou, W.X., Guo, Z.J., Tan, R.X., 2006. J. Appl. Microbiol. 100, 195.
Madaio, A., Piccialli, V., Sica, D., 1989. J. Nat. Prod. 52, 952.
Qi, S.H., Zhang, S., Xiao, Z.H., Huang, J.S., Wu, J., Li, Q.X., 2004. Chem. Pharm. Bull. 52, 1476.
Subrahmanyam, C., Kumar, S.R., 2000. J. Chem. Res. (S) 4, 182.
Wang, W., Xu, S.H., Liao, X.J., 2010. Chin. Pharm. J. 45, 172.
Yang, J., Qi, S.H., Zhang, S., Wu, J., Xiao, Z.H., 2005. Chin. J. Chem. 23, 1218.
Zhang, W., Guo, Y.W., Gavagnin, M., Mollo, E., Cimino, G., 2005. Helv. Chim. Acta 88, 87.