RSC Advances

This is an *Accepted Manuscript*, which has been through the Royal Society of Chemistry peer review process and has been accepted for publication.

Accepted Manuscripts are published online shortly after acceptance, before technical editing, formatting and proof reading. Using this free service, authors can make their results available to the community, in citable form, before we publish the edited article. This *Accepted Manuscript* will be replaced by the edited, formatted and paginated article as soon as this is available.

You can find more information about *Accepted Manuscripts* in the Information for Authors.

Please note that technical editing may introduce minor changes to the text and/or graphics, which may alter content. The journal's standard Terms & Conditions and the Ethical guidelines still apply. In no event shall the Royal Society of Chemistry be held responsible for any errors or omissions in this *Accepted Manuscript* or any consequences arising from the use of any information it contains.

ROYAL SOCIETY OF CHEMISTRY

www.rsc.org/advances

New α-Glucosidase Inhibitors from a Marine Sponge-derived Fungus *Aspergillus* sp. OUCMDZ-1583

Fandong Kong,[†] Chengying Zhao,[†] Jiejie Hao,[†] Cong Wang,[†] Wei Wang,[†] Xiaolong Huang,[‡] Weiming Zhu[*,†]

[†] *Key Laboratory of Marine Drugs, Ministry of Education of China, School of Medicine and Pharmacy, Ocean University of China, Qingdao 266003, China;* [‡] *College of Agriculture, Hainan University, Haikou 570228, China*

RSC Advances Accepted Manuscript

[*] To whom correspondence should be addressed. Tel: +86-532-82031268. Fax: +86-532-82031268. E-mail: weimingzhu@ouc.edu.cn

Eighteen new compounds named aspergones A−Q (**1−17**) and 6-*O*-demethylmonocerin (**18**), along with five known analogues (**19−23**), were isolated from the fermentation broth of *Aspergillus* sp. OUCMDZ-1583 associated with an unidentified marine sponge XD10410 from the Xisha Islands of China. The structures including the absolute configurations were unambiguously elucidated by spectroscopic, X-ray crystallographic, chemical, and Mosher's methods along with quantum ECD calculation. Compounds **1**, **2**, **5**, **10**, **11**, **14–18**, and **21–23** showed α-glucosidase inhibitions with IC_{50} values of 2.36, 1.65, 1.30, 2.37, 2.70, 1.36, 1.54, 2.21, 2.26, 0.027, 1.65, 1.19 and 1.74 *m*M, respectively (acarbose as positive control, IC_{50} 0.95 *m*M), among which compound **18** is 35 times more potent than acarbose. In addition, compounds **18** and **21** exhibited inhibitory activity against the influenza A (H1N1) virus with IC_{50} values of 172.4 and 175.5 μM, respectively (ribavirin as positive control, IC_{50} 137.3 μM).

RSC Advances Accepted Manuscript

Microorganisms continue to play an important role in the search for novel and bioactive compounds for drug development.[1,2] However, with the deepening of research on the terrestrial microbial natural products, the discovery of new entities from these microorganisms is increasingly difficult due to chemical redundancy.[3] As a result, many natural product chemists turned their attention to marine counterparts especially marine fungi that are supposed to be a tremendous resource for drug discovery for their special niches.[4,5] With this trend in mind and as a continuation of our investigations on structurally new and bioactive natural products of marine fungal origin,[6–9] an endozoic fungus *Aspergillus* sp. OUCMDZ-1583 was isolated from an unidentified marine sponge XD10410 from the Xisha Islands of China. The EtOAc extract of the fermentation broth showed α–glucosidase inhibition with an IC_{50} value of 0.97 mg/mL while the IC_{50} value of acarbose (positive control) was 0.61 mg/mL. Chemical examination of the fermentation broth resulted in the isolation and identification of eighteen new compounds that we named aspergones A−Q (**1**−**17**) and 6-*O*-demethylmonocerin (**18**) as well as five known compounds, epoxyquinol (**19**),[10] 7-*O*-demethylmonocerin (**20**),[11] (+)-monocerin (**21**),[11,12] fusarentin 6-methyl ether (**22**),[11] and 6,7-*O*-dimethyl-4*R*-hydroxy-10-*epi*fusarentin (**23**)[13] (Table S1, Supporting Information).

Results and Discussion

Aspergone A **(1)** was obtained as a brown oil with the molecular formula of $C_{11}H_{14}O_3$ from the HRESIMS peak at *m/z* 195.1014 $[M + H]^+$. The IR spectrum of **1** showed the presence of a lactone carbonyl (1777 cm^{-1}), and a hydroxy group (3377 cm^{-1}). The ^1H and ^{13}C NMR data (Table 1) along with the HSQC spectrum showed the presence of one triplet methyl ($\delta_{C/H}$ 13.7/0.91), three

RSC Advances Accepted Manuscript

methylenes including an oxygenated one, six olefinic carbons with four protonated, and one lactone carbonyls (δ_C 170.1). These data were similar to the known 5-(*E*)-but-2-enylidene-3-propyl-5*H*-furan-2-one (**24**) [14] except for an oxygenated methylene ($\delta_{C/H}$ 63.3/4.24) replacement of the corresponding methyl signals, indicating hydroxylation of C-8 in **1**. This deduction was further evidenced by the COSY cross-peaks from H-5 (δ_H 5.67) to H_2-8 (δ_H 4.24) through H-6 (δ_H 6.74) and H-7 (δ_H 6.04) and from H_2-9 (δ_H 2.31) to H_3-11 (δ_H 0.91) through H_2-10 (δ_H 1.57) as well as the HMBC correlations of H-3 (δ_H 6.96) to C-1 (δ_C 170.1), C-2 (δ_C 134.4), C-4 (δ_C 148.0), and C-9 (δ_C 27.4), H-6 to C-4, H-5 to C-3 (δ_C 136.6) and C-4, and of H-9 to C-1, C-2, and C-3 (Figure 1). The *E*- and *Z*- geometries of the Δ^6- and Δ^4- double bonds could be deduced from the large $J_{H\text{-}6,\ H\text{-}7}$ value (16.3 Hz) (Table 1) and NOE difference experiment (Figure 2), respectively. The H-3 signal (δ_H 6.96) was significant enhanced when H-5 signal (δ_H 5.67) was irradiated.

Aspergone B (**2**) has the same molecular formula with **1** from the HRESIMS peak at *m/z* 217.0832 $[M + Na]^+$ and the similar ^{13}C NMR data (Table 1). The difference was the hydroxymethine ($\delta_{C/H}$ 66.3/4.04) and a methyl signals ($\delta_{C/H}$ 19.0/1.84) replaced the corresponding hydroxymethylene and methylene signals of **1**. HMBC correlations from H-3 (δ_H 7.15) to C-1 (δ_C 171.1), C-2 (δ_C 129.7) and C-4 (δ_C 146.2), H_2-9 (δ_H 2.46, 2.53) to C-1, C-2 and C-3 (δ_C 139.3), and from H_3-8 (δ_H 1.84) to C-6 (δ_C 124.9) and C-7 (δ_C 136.5) along with the ^1H-^1H COSY data of H_2-9/H-10 (δ_H 4.04)/H_3-11 (δ_H 1.21) suggested the hydroxy group located at C-10 in **2**. The *E*- and *Z*- geometries of the Δ^6- and Δ^4- double bonds could be deduced from the large $J_{H\text{-}6,\ H\text{-}7}$ value (15.6 Hz) (Table 1) and NOE difference experiment (Figure 2), respectively. The H-5 signal (δ_H 5.71) was significantly enhanced when H-3 signal (δ_H 7.15) was irradiated. And the absolute configuration of C-10 was determined by the modified Mosher's method.[15] The $\Delta\delta$ values between (*S*)-MTPA ester (**2b**) and (*R*)- MTPA ester (**2a**)

RSC Advances Accepted Manuscript

clearly indicated the 10*S*-configuration (Figure 3).

Aspergones C (**3**) and D (**4**) were initially obtained as a racemic mixture from the zero value of specific rotation and the same molecular formula of $C_{11}H_{16}O_3$ from a HRESIMS peak at *m/z* 219.0988 $[M + Na]^+$. Although the ^1H and ^{13}C NMR data revealed the presence of the same furan-2(5*H*)-one nucleus as **1**, the remaining portion was slightly different. ^1H-^1H COSY data (supporting information) and HMBC correlations of H-7 (δ_H 3.96) to C-5 (δ_C 110.4) and H-5 (δ_H 5.26) to C-3 (δ_C 137.0) and C-4 (δ_C 149.8) revealed that 3-hydroxybutylidene group in **3** and **4** replaced the corresponding 4-hydroxybutenylidene group in **1**. The *Z*-geometry of the Δ^4- double bond was deduced from the NOE enhancements of the H-5 signal after irradiation of the H-3 (δ_H 6.99) (Figure 2). Upon chiral chromatography over a CHIRAPAK IA HPLC column, optically pure **3** and **4** were obtained. The distribution of $\Delta\delta$ values between the (*S*)- and (*R*)-MTPA esters (**3a** and **3b)** indicated the 7*R*-configuration of **3** (Figure 3). Therefore, the absolute configuration of **4** was determined to be 7*S.*

The molecular formula of aspergone E (**5**) was established as $C_{11}H_{16}O_3$ by HRESIMS peak at *m/z* 219.0989 $[M + Na]^+$ with four degrees of unsaturation. Strong IR absorption at 1769 and 1645 cm^{-1} implied the presence of lactone carbonyl and double bond functional groups. ^1H-^1H COSY cross-peaks from H_2-8 (δ_H 5.30, 5.37) through H-7 (δ_H 6.68), H-6 (δ_H 6.31), H-5 (δ_H 5.41), H-4 (δ_H 4.98), H-3 (δ_H 3.97), H-2 (δ_H 2.62), H_2-9 (δ_H 1.84, 1.58) and H_2-10 (δ_H 1.57, 1.51) to H_3-11 (δ_H 0.96) and HMBC correlations from H-2 and H_2-9 to C-1 (δ_C 175.9) indicated a 4-(but-1,3-dienyl)-3-hydroxy-2-propylbutyrrolactone structure (figure 2). The *Z*- geometry of the Δ^5-double bond could be determined by the $J_{H\text{-}5,H\text{-}6}$ value (10.2 Hz, Table 1) and NOE enhancement of H-7 after irradiation of H-4. Further, the NOE enhancements of H-5 and H-9 were observed when

RSC Advances Accepted Manuscript

H-3 was irradiated while H-2 signal was enhanced after irradiation of H-4, indicating *trans*-orientations of both H-2 and H-4 with H-3 (Figure 2). The distribution of $\Delta\delta$ values between (*S*)- and (*R*)-MTPA esters (**5a** and **5b**) indicated the 3*S*-configuration (Figure 3). Thus, the absolute configuration of **5** was determined to be 2*S*, 3*S*, and 4*R*.

The molecular formula of aspergone F (**6**) was established as $C_{11}H_{16}O_2$ by HRESIMS peak at *m/z* 203.1037 $[M + Na]^+$ with an oxygen less than in **5**. Comparison of ^{13}C NMR data between **6** and **5** (Table 1) revealed that an olefinic quaternary carbon, an olefinic methine and an oxygenated methylene signals in **6** replaced the corresponding sp^3-methine, sp^3-methylene and ester carbonyl signals in **5**. Two separated ^1H-^1H COSY systems of H-3/H-4/H-5/H-6/H-7/H-8 and H-9/H-10/H-11 (supporting information) were observed in **6**, indicating that the propylidene group replaced the propyl group in **5**. And the key HMBC data from H_2-1 (δ_H 4.27, 4.18) to C-2 (δ_C 140.2), C-4 (δ_C 86.6) and C-9 (δ_C 125.7) and from H-9 (δ_H 5.36) to C-1 (δ_C 70.1) confirmed the replacement of the carbonyl group in **5** by a –CH_2– group in **6**. The *E*-geometry of the Δ^5-double bond was deduced from the large $J_{H\text{-}5,H\text{-}6}$ value (15.7 Hz, Table 1) and the NOE enhancement of H-6 (δ_H 6.21) after irradiation of H-4 (δ_H 4.09) (Figure 2). The NOE enhancements of H-5 (δ_H 5.71) and H-9 after irradiation of H-3 (δ_H 4.22) (Figure 2) indicated the *trans*-orientation of H-3 and H-4 and *E*-geometry of $\Delta^{2(9)}$-double bond. The distribution of $\Delta\delta$ values between (*S*)- and (*R*)-MTPA esters (**6a** and **6b**) indicated the 3*S*-configuration (Figure 3). Thus, the absolute configuration of **6** was determined as 3*S* and 4*R*.

Aspergones G (**7**) and H (**8**) were initially isolated as a racemic mixture and the molecular formula was determined as $C_{11}H_{16}O_2$ on the basis of HRESIMS peak at *m/z* 203.1038 $[M + Na]^+$. The strong UV and IR absorptions at λ_{max} 223 nm and ν_{max} 1703 and 1614 cm^{-1} indicated the presence of a

RSC Advances Accepted Manuscript

conjugated enone moiety which was further supported by the HMBC correlations from an olefinic proton (δ_{H-2} 5.87) to a sp^2-quaternary carbon (δ_{C-3} 180.7) and a carbonyl carbon (δ_{C-1} 206.9). ^{1}H-^{1}H COSY (Figure 1) and HSQC data indicated the presence of two isolated spin systems, CH(4)-CH(5)-CH_2(6)-CH(7)-CH_2(8) and CH_3(11)-CH_2(10)-CH_2(9). The key HMBC correlations of H-4 (δ_H 4.45) to C-1, C-2 (δ_C 128.9), C-5 (δ_C 55.3), C-6 (δ_C 31.7) and C-9 (δ_C 32.9) linked these three moieties as 5-allyl-4-hydroxy-3-propylcyclopent-2-en-1-one. When H-4 was irradiated, the H_2-6 (δ_H 2.56, 2.21) signals were enhanced, indicating *trans*-orientation between H-4 and H-5 (δ_H 2.37). A chiral HPLC separation over a CHIRAPAK IA column afforded the optically pure *d*-isomer (**7**) and *l*-isomer (**8**). The distribution of $\Delta\delta$ values between (*S*)- and (*R*)-MTPA esters (**7a** and **7b**) indicated the 4*R*- configuration (Figure 3). Thus, the absolute configuration of **7** was determined to be 4*R* and 5*R*. As a consequence, the absolute configuration of **8** was determined to be 4*S* and 5*S*.

Aspergone I (**9**) showed a HRESIMS peak at *m/z* 205.1195 [M + Na]$^+$ corresponding to the molecular formula $C_{11}H_{18}O_2$. Its 1D NMR revealed two methyls (δ_C 17.9 & 18.1), an oxygenated methylene (δ_C 68.7), six olefinic methines and an oxygenated quaternary carbon (δ_C 75.0) (Table 1). ^{1}H-^{1}H COSY and HSQC data indicated the presence of CH_3(8)-CH(7)-CH(6)-CH(5)-CH(4)-CH_2(3) and CH(9)-CH(10)-CH_3(11) units (Figure 1). HMBC correlations (Figure 1) from H_2-1 (δ_H 3.45, 3.43) to C-2 (δ_C 75.0), C-3 (δ_C 41.0) and C-9 (δ_C 133.3) and from H_2-3 (δ_H 2.31, 2.32) to C-1 (δ_C 68.7), C-2 and C-9 connected these structural moieties as 2-propenylocta-4,6-diene-1,2-diol (Figure 1). The large *J* values of H-4/H-5 (14.5 Hz), H-6/H-7 (14.4 Hz) and H-9/H-10 (15.5 Hz) (Table 1) indicated all the $\Delta^{4,6,9}$- double bonds as *E*-geometry. The absolute configuration was assigned using the *in situ* dimolybdenum CD method.[16,17] After addition of $Mo_2(OAc)_4$ into DMSO solution of **9**, a metal complex auxiliary chromophore was generated. Because the contribution from the inherent CD was

RSC Advances Accepted Manuscript

subtracted, the Cotton effect observed in the induced CD spectrum originates solely from the chirality of the *vic*-diol moiety. The negative Cotton effect observed at 320 (Δε –0.64) and 400 (Δε –0.10) nm in the induced CD spectrum (Figure 5) revealed the 2*S*- configuration according to Snatzke's empirical rule.[18] The structure of **9** was therefore elucidated as (2*S*,4*E*,6*E*)-2-(*E*-propenyl)octa-4,6-diene-1,2-diol.

Aspergone J (**10**) has the same molecular formula $C_{11}H_{18}O_2$ as **9** from a HRESIMS peak at *m/z* 205.1196 $[M + Na]^+$. NMR comparison revealed that a methylene and an olefinic methylene of **10** replaced a methyl and an olefinic methine of **9**. ^1H-^1H COSY spectrum of **10** further indicated a hexatrienyl and a propyl other than the hexadienyl and propenyl in **9**, which were also supported by the key HMBC correlations from H-3 (δ_H 5.79) to C-1 (δ_C 69.0), C-2 (δ_C 75.2) and C-9 (δ_C 40.0). The *E*-geometries of $\Delta^{3,5}$- double bonds could be deduced from the *J* values of H-3/H-4 (14.8 Hz) and H-5/H-6 (14.8 Hz) (Table 1). And the same sign of $[\alpha]_D$ (–55.3) to that of **9** (–34.8) implied the same 2*R*- configuration, that is (2*R*,3*E*,5*E*)-2-propylocta-3,5,7-triene-1,2-diol.

Aspergone K (**11**) also has the molecular formula $C_{11}H_{18}O_2$ based on a HRESIMS peak at *m/z* 205.1195 $[M + Na]^+$. The NMR data and ^1H-^1H COSY and HMBC coupling modes along with $[\alpha]_D$ (–64.5) were very similar to those of **10**, indicating almost the same structure. The only difference performed in the *Z*-geometry of Δ^5- double bond from the relatively small $J_{H\text{-}5,H\text{-}6}$ value (11.3 Hz). Thus, structure of **11** was determined as (2*R*,3*E*,5*Z*)-2-propylocta-3,5,7-triene-1,2-diol.

The molecular formula of aspergone L (**12**) was also established as $C_{11}H_{18}O_2$ by a HRESIMS peak at *m/z* 205.1196 $[M + H]^+$. 1D NMR, ^1H-^1H COSY and HSQC data indicated an oxygenated methylene, a 5-hydroxyhex-2-en-1-ylidene (CH_3(8)-CH(7)-CH_2(6)-CH(5)-CH(4)-CH(3)), a propenyl (CH(9)-CH(10)-CH_3(11)), and an olefinic quaternary carbon. The key HMBC correlations of H-3 (δ_H

RSC Advances Accepted Manuscript

6.03) to C-1 (δ_C 62.4), C-2 (δ_C 135.7) and C-9 (δ_C 125.8) along with H_2-1 (δ_H 4.07) to C-2, C-3 (δ_C 126.7) and C-9 connected above structural moieties as 2-propenylocta-2,4-diene-1,7-diol (Figure 2). All the geometries of $\Delta^{2,4,9}$- double bonds were assigned as *E*- according to the large *J* value of H-4/H-5 (15.0 Hz) and H-9/H-10 (16.6 Hz) (Table 1) and the NOESY cross-peak between H-3 and H_2-1 (Figure 2). To determine the absolute configuration of **12**, the 1-*O*-*t*-butyldimethylsilyl (TBDMS) derivative (**12a**) and the (*R*)- and (*S*)-MTPA esters of **12a** were prepared. The distribution of $\Delta\delta$ values between (*S*)- and (*R*)-MTPA esters (**12aa** and **12ab**) indicated the 7*R*-configuration (Figure 3). Thus, compound **12** was determined to be (7*R*,2*E*,4*E*)-2-(*E*-propenyl)octa-2,4-diene-1,7-diol.

The molecular formula of aspergone M (**13**) was established as $C_{11}H_{20}O_2$ by a HRESIMS peak at *m/z* 207.1350 $[M + Na]^+$, equivalent to **12** with an additional H_2 unit. Comparison of ^1H and ^{13}C NMR data between **13** and **12** revealed that their structures were only different in the replacement of the propenyl group in **12** with a propyl group in **13**. This deduction was further supported by the ^1H-^1H COSY connectivity of H_3-11/H_2-10/H_2-9 and the key HMBC correlations of H_2-1 (δ_H 3.87) to C-2 (δ_C 140.9), C-3 (δ_C 124.3) and C-9 (δ_C 30.4). The *E*-geometry of both Δ^2- and Δ^4- double bonds were assigned from the large $J_{H\text{-}4,\,H\text{-}5}$ (15.4 Hz) and the NOESY cross-peak of H-3 (δ_H 5.97) with H_2-1 (Figure 2). The close specific rotation of **13** to **12** (–36.7 *vs* –30.5) implied the same 7*R*-configuration. Thus, compound **13** was elucidated as (7*R*,2*E*,4*E*)-2-propylocta-2,4-diene-1,7-diol.

Aspergone N (**14**) had a molecular formula $C_{10}H_{16}O_5$ from a HRESIMS peak at *m/z* 239.0888 $[M + Na]^+$ with three degrees of unsaturation. The IR spectrum of **14** showed the absorption of the double bond (1645 cm^{-1}) and hydroxy (3287 cm^{-1}). The ^1H and ^{13}C NMR along with the HSQC data revealed the presence of a doublet methyl, an oxygenated methylene, four oxygenated methines,

RSC Advances Accepted Manuscript

two vicinal olefinic methines, two olefinic quaternary carbons, and five hydroxy groups. This was accounting for two degrees of unsaturation, indicating a cyclic nucleus in **14**. ^{1}H-^{1}H COSY correlations reveal the presence of CH(-OH)(3)-CH(-OH)(4)-CH(-OH)(5)-CH(-OH)(6), CH_3(9)-CH(8)-CH(7), and CH_2(-OH)(10) moieties. HMBC correlations from H-7 (δ_H 6.34) to C-1 (δ_C 133.4), C-2 (δ_C 135.8) and C-6 (δ_C 67.9) and from H_2-10 (δ_H 4.18, 3.92) to C-1, C-2, and C-3 (δ_C 68.5) connected these moieties as 2-hydroxymethyl-1-propenyl-1-cyclohexene-3,4,5,6- tetraol. The large $J_{H-7,H-8}$ (15.7 Hz) suggested *E*-Δ^7-double bond. The relative configurations of C-3–C-6 and the geometry of Δ^7-double bond were supported by the x-ray single crystal diffraction (Figure 4). The absolute configurations of all the chiral centers were assigned as *R*- by ECD calculations of **14** and *ent*-**14** using the time-dependent density functional theory (TD-DFT) method at the B3LYP/6-31G(d) level.[19] The results showed that the measured CD curve is matched well with the calculated ECD for **14** and opposite to that of *ent*-**14** (Figure 6).

Aspergone O (**15**) was isolated as a colorless orthorhombic crystal with a molecular formula of $C_{10}H_{15}ClO_4$ on the basis of a HRESIMS peak at *m/z* 257.0548 $[M + Na]^+$, indicating the substitution of -Cl for –OH relative to **14**. In addition, we noted that there was one hydroxy signal less and ^{1}H-^{1}H COSY and HMBC coupling patterns were similar to **14** except the lack of COSY between 4-OH and 4-CH, supporting the substitution of 4-Cl in **15** for 4-OH in **14**. The large $J_{H-7, H-8}$ (16.2 Hz) indicated *E*-geometry of Δ^7-double bond. The similar CD data with **14** (Figure 6) indicated (3*R*,4*S*,5*S*,6*R*)-configuration of **15** that was further confirmed by single-crystal X-ray diffraction (Figure 4) with a small Flack parameter (0.05(7)) and a heavy chlorine atom in molecule.[20,21] Thus, aspergone O (**15**) was identified as (3*R*,4*S*,5*S*,6*R*,7*E*)-4-chloro-2-hydroxymethyl-1-propenylcyclohex-1-ene-3,5,6-triol.

RSC Advances Accepted Manuscript

Aspergone P (**16**) had the molecular formula of $C_{10}H_{14}O_4$ as determined from its HRESIMS peak at *m/z* 221.0780 $[M + Na]^+$, equivalent to **14** minus a H_2O unit. The absence of two hydroxy proton signals and the upfield oxygenated methine carbon signals (δ_C 54.5, 54.3) indicated a replacement of two hydroxy groups by an epoxy group. This epoxidation was deduced to occur at C-4 and C-5 according to the ^1H-^1H COSY of OH-3/H-3/H-4/H/5/H-6/OH-6 and the HMBC correlations from H-3 (δ_H 4.52) to C-1 (δ_C 131.2) along with H-6 (δ_H 4.45) to C-2 (δ_C 133.2), C-4 (δ_C 54.5) and C-7 (δ_C 128.3). The large $J_{H\text{-}7,\ H\text{-}8}$ (15.8 Hz) suggested *E*-geometry of Δ^7-double bond. The absolute configuration of **16** was defined by chemical transformation into **15**. After adding hydrochloric acid to a methanol solution of **16**, compound **15** was yielded as a major product (Figure S120) which showed the identical $[\alpha]_D$, NMR and MS data with those of natural one, indicating (3*R*,4*R*,5*S*,6*R*)-configuration of **16**. The result also indicated that compound **15** might be an artifact under acidic conditions formed in the fermentation process.

The molecular formula of aspergone Q (**17**) was established as $C_{11}H_{14}O_6$ by a HRESIMS peak at *m/z* 265.0680$[M + Na]^+$. The NMR data (Table 1) of **17** are similar to those of **14**, except for the absence of two hydroxy proton signals and the presence of an additional carbonate carbonyl signal (δ_C 154.8), suggesting that two hydroxy groups of **14** were esterified to form a cyclic carbonate. HMBC correlations of H-5 (δ_H 4.81) and H-6 (δ_H 5.60) to C-11 (δ_C 154.8) and H-7 (δ_H 6.42) to C-6 (δ_C 75.5) supported that the 5,6-dihydroxy was carbonated in **17** (Figure 2). Treatment of **17** in 1% aqueous sodium hydroxide yielded compound **14** which showed the identical $[\alpha]_D$, NMR and MS data with the natural one, indicating (3*R*,4*R*,5*S*,6*R*)- configuration of **17**.

Compound **18** was found to have the molecular formula $C_{15}H_{18}O_6$ by HRESIMS peak at *m/z* 295.1171 $[M + H]^+$. Apart from the phenyl motif, the ^1H and ^{13}C NMR data (Table 1) were nearly

RSC Advances Accepted Manuscript

identical to the known compound **20**,[11] indicating the similar structure isomeric in phenyl motif. The HSQC revealed the presence of five aromatic quaternary carbons, one aromatic methine ($\delta_{C/H}$ 108.1/6.60), three oxygenated methines, three methylenes, one methyl ($\delta_{C/H}$ 14.1/0.90), one methoxyl ($\delta_{C/H}$ 60.9/3.97), and one ester carbonyl (δ_C 168.2). The key HMBC correlations from a phenolic hydroxy proton ($\delta_{HO\text{-}8}$ 11.5) to three aromatic quaternary carbons ($\delta_{C\text{-}7,8,8a}$ 134.9, 155.6, 101.3) and the methoxy protons (δ_H 3.97) to a relative upfield oxygenated aromatic quaternary carbon ($\delta_{C\text{-}7}$ 134.9) revealed that *O*-methylation occurred at 7-OH in **18** rather than 6-OH in **20**. The relative configuration was established from the NOE data and the comparison of NMR data with **20**, especially vicinal coupling constants. The small $J_{H\text{-}3,H\text{-}4}$ (3.0 Hz) (Table 1) along with the NOESY correlations from H-10 (δ_H 4.10) to H-3 (δ_H 5.02) and H-4 (δ_H 4.49) indicated the same *cis*-orientation of H-3, H-4 and H-10 as in **20**. The similar CD spectra between **18** and **20** (Figure 5) revealed the same (3*R*,4*R*,10*S*)-configuration. Because the absolute configuration of **20** has been resolved by chemical synthesis[11] and further by Cu-Kα radiated x-ray single crystal diffraction (Figure S107) with a small Flack parameter (0.0(3)) in this paper, the structure of compound **18** could be clearly elucidated as 6-*O*-demethylmonocerin.

All the isolated compounds (**1–23**) were evaluated for antiviral activity against the H1N1 flu virus, as well as α-glucosidase inhibition by preciously described methods.[6,22] Only compounds **18** and **21** exhibited anti-H1N1 activities with IC_{50} values of 172.4 μM and 175.5 μM, respectively (ribavirin as positive control, IC_{50} 137.3 μM), while the other compounds were not active against H1N1. These data suggested that the tetrahydrofuran moiety and the methoxy group located at C-7 in the isocoumarins were required for the antiviral activity against the influenza A H1N1 virus. In addition, compounds **1**, **2**, **5**, **10**, **11**, **14–18**, and **21–23** showed α-glucosidase inhibition with IC_{50}

RSC Advances Accepted Manuscript

values of 2.36, 1.65, 1.30, 2.37, 2.70, 1.36, 1.54, 2.21, 2.26, 0.027, 1.65, 1.19 and 1.74 *m*M (acarbose as positive control, IC_{50} 0.95 *m*M) (Table 3). The result showed that compound **18** is 35 times more potent than the positive control, which supports that marine fungi might be a promising source for drug discovery. Compounds **1–23** were also tested for cytotoxicities against A549 and K562 tumor cells by MTT[23] and MCF-7 cells by CCK-8[24] methods, respectively. Their antimicrobial activities against *Escherichia coli*, *Enterobacter aerogenes*, *Bacillus subtilis*, *Pseudomonas aeruginosa* and *Candida albicans* were also evaluated by an agar dilution method.[25] However, none of the compounds showed any activities against the tested tumor cells and pathogens.

Experimental Section

General Experimental Procedures. Optical rotations were measured on a JASCO P-1020 digital polarimeter, and UV spectra were measured on a Beckman DU 640 spectrophotometer. CD data were collected using a JASCO J-715 spectropolarimeter. IR spectra were taken on a Nicolet Nexus 470 spectrophotometer as KBr discs. ^{1}H NMR, ^{13}C NMR, DEPT, HMQC, HSQC, HMBC, COSY and NOESY spectra were acquired using a JEOL JNM-ECP 600 spectrometer or an Agilent 500 MHz DD2 spectrometer using TMS as an internal standard or residual solvent signals for referencing. HR-ESI-MS spectra were determined using the Q-TOF ULTIMA GLOBAL GAA076 LC mass spectrometer. Semi-preparative HPLC was carried out using an ODS column (YMC-pack ODS-A, 10 × 250 mm, 5 μm, 4 mL/min) and a πNAP column (COSMOSIL-pack, 10 × 250 mm, 5 μm, 4 mL/min). Sea salt used was made from the evaporation of seawater collected in Laizhou Bay (Weifang Haisheng Chemical Factory). Thin layer chromatography (TLC) and column chromatography (CC) were performed on plates precoated with silica gel GF_{254} (10–40 μm, Qingdao Marine Chemical Factory) and Sephadex LH-20 (Amersham Biosciences), respectively. Vacuum-liquid chromatography (VLC) utilized silica gel H (Qingdao Marine Chemical Factory).

RSC Advances Accepted Manuscript

Fungal Material and Fermentation. The fungus *Aspergillus* sp. OUCMDZ-1583 was isolated from a piece of sponge XD10410 from the Xisha Islands of China in August 2010. After it was ground into powder, the sample (1 g) was diluted to 10^{-2} g/mL with sterile water, 100 μL of which was deposited on a PDA (200 g potato, 20 g glucose, 20 g agar per liter of tap water) plate containing chloramphenicol (100 μg/mL) as a bacterial inhibitor. A single colony was transferred onto another PDA plate and was identified according to its morphological characteristics and ITS gene sequences (GenBank accession No. KM056275, Supporting Information). A reference culture of *Aspergillus* sp. OUCMDZ-1583 maintained at –80 °C is deposited in our laboratory. The isolate was cultured on slants of PDA medium at 28 °C for 5 days. Plugs of agar supporting mycelium growth were cut and transferred aseptically to 200 × 1000 mL Erlenmeyer flasks each containing 300 mL of liquid medium (2% mannitol, 2% maltose, 1% glucose, 1% monosodium glutamate, 0.3% yeast extract, 0.05% corn meal, 0.05% KH_2PO_4, 0.03% $MgSO_4$, 1.75% $Na_2HPO_4 \cdot 2H_2O$, 1.05% $C_4H_2O_7 \cdot H_2O$, 3.3% sea salt, pH 7.0). The flasks were incubated at room temperature under static conditions for 30 days.

Extraction and Isolation. The cultures (50 L) were filtered through cheesecloth to separate the mycelial mass from the aqueous layer. The filtrate was then extracted three times by 3-fold volumes of EtOAc, while the mycelium was extracted by acetone. After removing acetone by evaporation under vacuum, the obtained aqueous acetone solution was extracted three times with equal volumes of EtOAc. The combined EtOAc extracts were dried under vacuum to produce 28.7 g of extract. The EtOAc extract was subjected to a silica gel VLC column, eluting with a stepwise gradient of 0%, 20%, 40%, 60%, 80% and 100% MeOH in CH_2Cl_2 (v/v), to give 20 fraction (fractions 1−20). Fraction 1 (1.2 g) was subjected to Sephadex LH-20 chromatography (5 × 200 cm) with CH_2Cl_2-MeOH (1:1) to afford two subfractions (1.1 and 1.2). Fraction 1.1 (106 mg) was further

RSC Advances Accepted Manuscript

purified by HPLC over a πNAP column (70% MeOH/H_2O, v/v) to give compounds **23** (t_R 11.8 min; 20 mg) and **21** (t_R 16.0 min; 24 mg). Fraction 1.2 (234 mg) was subjected to HPLC over ODS (55% MeOH/H_2O, v/v) to yield **20** (t_R 9.5 min, 16.3 mg), **2** (t_R 9.7 min, 10.2 mg), **1** (t_R 12.6 min, 7.6 mg) and **18** (t_R 13.8 min, 7.6 mg). The fraction 2 (3.4 g) eluted with petroleum CH_2Cl_2-MeOH (100:1) was subjected to reversed-phase C18 silica column eluting with a stepwise gradient of 30% to 100% MeOH in H_2O to obtain four subfractions (2.1−2.4). Fraction 2.1 (247 mg) was further separated into three subfractions (2.1.1−2.1.3) by Sephadex LH-20 chromatography (5 × 200 cm) eluting with CH_2Cl_2-MeOH (1:1, v/v). Fractions 2.1.1 (56 mg) was subjected to HPLC over ODS (55% MeOH/H_2O, v/v) to yield fractions 2.1.1.1, which was further purified by HPLC over a πNAP column (50% MeOH/H_2O, v/v) to give compound **5** (t_R 17.0 min; 8.2 mg) and racemic mixture of **7** and **8** (t_R 14.7 min; 16 mg) that was further subjected to HPLC over a CHIRAPAK IA column (75% n-C_6H_{14}/EtOH, v/v) to give the optically pure **7** (t_R 5.7 min; 7.1 mg) and **8** (t_R 5.9 min; 8.3 mg). Fraction 2.1.2 (78 mg) was subjected to HPLC over ODS (35% MeCN/H_2O, v/v) to yield racemic mixture of **3** and **4** (t_R 7.7 min; 24 mg) that was further purified by a CHIRAPAK IA column (75% n-C_6H_{14}/EtOH, v/v) to give optically pure **3** (t_R 6.9 min; 11.3 mg) and **4** (t_R 7.8 min; 10.9 mg). Fraction 2.1.3 (39 mg) was subjected to HPLC over ODS (50% MeOH/H_2O, v/v) to yield compound **6** (t_R 21.3 min; 11.3 mg). Fractions 15–20 (4.2 g) were combined and re-chromatographed over Sephadex LH-20 (5 × 200 cm, MeOH) to afford four subfractions (Fr. 15.1–Fr. 15.4). Fraction 15.4 (706 mg) was subjected to HPLC over ODS (25% MeOH/H_2O, v/v) to give **17** (t_R 6.2 min; 76 mg), **14** (t_R 4.0 min; 22 mg) and subfraction 15.4.1. Fraction 15.4.1 (215 mg) was fractionated by silica gel CC using petroleum ether−EtOAc (4:6, v/v) to afford **15** (35.5 mg), **16** (106.5 mg) and **19** (5.4 mg). Fractions 6 and 7 (1.3 g) were combined and chromatographed over Sephadex LH-20 (5 × 200

RSC Advances Accepted Manuscript

cm, MeOH) to afford three subfractions (Fr. 6.1–Fr.6.3). Fraction 6.1 (78 mg) was subjected to HPLC over ODS (55% MeOH/H_2O, v/v) to yield compounds **24** (t_R 6.7 min; 24.3 mg) and **22** (t_R 9.9 min; 14.7 mg). Fraction 6.2 (108 mg) was subjected to HPLC over ODS (50% MeOH/H_2O, v/v) to yield compounds **9** (t_R 26.6 min; 18.3 mg), **10** (t_R 27.9 min; 3.7 mg) and **11** (t_R 30.5 min; 4.5 mg). Fraction 6.3 (94 mg) was separated by HPLC over ODS (50% MeOH /H_2O, v/v) to yield compounds **12** (t_R 11.7 min; 18.3 mg) and **13** (t_R 13.9 min; 3.5 mg).

Aspergone A (**1**): brown oil; UV (MeOH) λ_{max} (log ε) 325 (3.67) and 208 (3.09) nm; IR (KBr) ν_{max} 3377, 2984, 1777, 1715 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 195.1014 $[M + H]^+$ (calcd for $C_{11}H_{15}O_3$, 195.1016).

Aspergone B (**2**): brown oil; $[\alpha]^{25}_D$ –27.8 (*c* 0.1, MeOH); UV (MeOH) λ_{max} (log ε) 324.8 (3.63) and 208.6 (3.11) nm; IR (KBr) ν_{max} 3416, 2972, 1766, 1641 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 217.0832 $[M + Na]^+$ (calcd for $C_{11}H_{14}O_3Na$, 217.0832).

Aspergone C (**3**): brown oil; $[\alpha]^{25}_D$ –20.6 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log ε) 275.2 (3.80) and 201.6 (3.10) nm; IR (KBr) ν_{max} 3420, 2918, 1676, 1645 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 219.0988 $[M + Na]^+$ (calcd for $C_{11}H_{16}O_3Na$, 219.0992).

Aspergone D (**4**): brown oil; $[\alpha]^{25}_D$ +21.8 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log ε) 275.2 (3.80) and 201.6 (3.10) nm; IR (KBr) ν_{max} 3420, 2918, 1676, 1645 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 219.0988 $[M + Na]^+$ (calcd for $C_{11}H_{16}O_3Na$, 219.0992).

Aspergone E (**5**): brown oil; $[\alpha]^{25}_D$ –16.8 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log ε) 225.2 (3.78) and 204.3 (3.17); CD (*c* 0.1, MeOH) λ_{max} ($\Delta\varepsilon$) 226.5 (+1.05) and 209 (+2.35); IR (KBr) ν_{max} 3420, 2966, 1769, 1645 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 219.0989 $[M + Na]^+$ (calcd for $C_{11}H_{16}O_3Na$, 219.0992).

RSC Advances Accepted Manuscript

Aspergone F (**6**): brown oil; $[\alpha]^{25}_{D}$ +18.7 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 225.5 (3.75) and 203.6 (3.50); CD (*c* 0.1, MeOH) λ_{max} ($\Delta\varepsilon$) 217.5 (+3.20) and 210.5 (–0.87); IR (KBr) ν_{max} 3443, 2966, 1680, 1645 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 203.1037 $[M + Na]^+$ (calcd for $C_{11}H_{16}O_2Na$, 203.1043).

Aspergone G (**7**): brown oil; $[\alpha]^{25}_{D}$ +10.2 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 223 (3.38) and 203.5 (2.99); CD (*c* 0.05, MeOH) λ_{max} ($\Delta\varepsilon$) 235 (+1.96) and 207 (–0.51); IR (KBr) ν_{max} 3404, 2960, 1703, 1614 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 203.1038 $[M + Na]^+$ (calcd for $C_{11}H_{16}O_2Na$, 203.1043).

Aspergone H (**8**): brown oil; $[\alpha]^{25}_{D}$ –9.1 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 223 (3.38) and 203.5 (2.99); CD (*c* 0.05, MeOH) λ_{max} ($\Delta\varepsilon$) 235 (–2.06) and 207 (+0.54); IR (KBr) ν_{max} 3404, 2960, 1703, 1614 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 203.1038 $[M + Na]^+$ (calcd for $C_{11}H_{16}O_2Na$, 203.1043).

Aspergone I (**9**): brown oil; $[\alpha]^{25}_{D}$ –34.8 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 229.5 (3.45) and 204 (2.94); CD (*c* 0.05, MeOH) λ_{max} ($\Delta\varepsilon$) 233.5 (+0.99) and 209 (+0.40); IR (KBr) ν_{max} 3420, 2918, 1676, 1645 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 205.1195 $[M + Na]^+$ (calcd for $C_{11}H_{18}O_2Na$, 205.1199).

Aspergone J (**10**): brown oil; $[\alpha]^{25}_{D}$ –55.3 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 253 (3.34), 263 (3.57) and 274 (3.38); CD (*c* 0.1, MeOH) λ_{max} ($\Delta\varepsilon$) 273 (+2.23) and 258.5 (–0.51); IR (KBr) ν_{max} 3420, 2949, 1680, 1641 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 205.1196 $[M + Na]^+$ (calcd for $C_{11}H_{18}O_2Na$, 205.1199).

Aspergone K (**11**): brown oil; $[\alpha]^{25}_{D}$ –64.5 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 253 (3.44), 263 (3.48) and 274 (3.38); CD (*c* 0.05, MeOH) λ_{max} ($\Delta\varepsilon$) 265 (+2.54) and 249 (–0.70); IR (KBr) ν_{max}

RSC Advances Accepted Manuscript

3392, 2964, 1676, 1645 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 205.1195 [M + Na]$^{+}$ (calcd for $C_{11}H_{18}O_2Na$, 205.1199).

Aspergone L (**12**): brown oil; $[\alpha]^{25}_{D}$ –30.5 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 258.5 (3.79), 269.5 (3.87) and 280 (3.77); CD (*c* 0.1, MeOH) λ_{max} (Δ*ε*) 276 (+2.43) and 261.5 (–1.10); IR (KBr) ν_{max} 3389, 2918, 1649, 1614 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 205.1196 [M + Na]$^{+}$ (calcd for $C_{11}H_{18}O_2Na$, 205.1199).

Aspergone M (**13**): brown oil; $[\alpha]^{25}_{D}$ –36.7 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 233 (3.72), 239 (3.74) and 248.5 (3.57); CD (*c* 0.1, MeOH) λ_{max} (Δ*ε*) 251.5 (+0.26) and 234 (–0.55); IR (KBr) ν_{max} 3389, 2918, 1649, 1614 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 207.1350 [M + Na]$^{+}$ (calcd for $C_{11}H_{20}O_2Na$, 207.1356).

Aspergone N (**14**): colorless orthorhombic crystal; mp 123−125; $[\alpha]^{25}_{D}$ –73.5 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 239.5 (3.39), 206 (3.03); CD (*c* 0.05, MeOH) λ_{max} (Δ*ε*) 230.5 (–1.02); IR (KBr) ν_{max} 3287, 2910, 1645, 1443 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 239.0888 [M + Na]$^{+}$ (calcd for $C_{10}H_{16}O_5Na$, 239.0890).

Aspergone O (**15**): colorless orthorhombic crystal; mp 117−119; $[\alpha]^{25}_{D}$ –12.7 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 238 (3.58), 206 (3.18); CD (*c* 0.05, MeOH) λ_{max} (Δ*ε*) 236.5 (–1.67); IR (KBr) ν_{max} 3392, 2925, 1657, 1443 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 257.0548 and 259.0518 [M + Na]$^{+}$ (calcd for $C_{10}H_{15}O_4ClNa$, 257.0551 and 257.0522).

Aspergone P (**16**): white amorphous powder; $[\alpha]^{25}_{D}$ +81.0 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log *ε*) 238 (3.48), 206.5 (2.90); CD (*c* 0.1, MeOH) λ_{max} (Δ*ε*) 232 (–1.54); IR (KBr) ν_{max} 3350, 2921, 1660, 1446 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 221.0780 [M + Na]$^{+}$ (calcd for $C_{10}H_{14}O_4Na$, 221.0784).

RSC Advances Accepted Manuscript

Aspergone Q (**17**): colorless oil; $[\alpha]^{25}_{D}$ –11.6 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log ε) 238 (3.36), 205.5 (2.97); CD (*c* 0.05, MeOH) λ_{max} ($\Delta\varepsilon$) 231 (–1.29); IR (KBr) ν_{max} 3350, 2921, 1660, 1446 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 265.0680 $[M + Na]^+$ (calcd for $C_{11}H_{14}O_6Na$, 265.0683).

6-*O*-Demethylmonocerin (**18**): brown oil; $[\alpha]^{25}_{D}$ +23.3 (*c* 0.1, MeOH), UV (MeOH) λ_{max} (log ε) 219 (3.58), 232.5 (3.53), 274 (3.37), 309 (3.02); CD (*c* 0.1, MeOH) λ_{max} ($\Delta\varepsilon$) 274 (–2.23), 242 (+0.62), and 212.5 (+2.68); IR (KBr) ν_{max} 3335, 2956, 1660, 1468, 1376 cm^{-1}; ^{1}H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 295.1171 $[M + H]^+$ (calcd for $C_{15}H_{19}O_6$, 295.1176).

Preparation of 12-*O*-*tert*-butyldimethylsilylaspergone L (12a). *tert*-Butyldimethylsilyl chloride (TBDMSCl) (6.7 mg) was added to a mixture of compound **12** (4.1 mg) and imidazole (10 mg) in DMF (1.5mL). The reaction mixture was stirred at r.t. for 2 h. Then the reaction mixture was quenched with H_2O and extracted three times with EtOAc. The EtOAc layers were combined and separated by semipreparative HPLC on ODS (100% MeOH) to afford **12a** (t_R 4.4 min, 3.4 mg) as one product (Figure S119).

12-*O*-*tert*-Butyldimethylsilylaspergone L (12a): ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.59 (dd, J = 15.4, 11.8, 1H, H-4), 6.48 (d, J = 16.2, 1H, H-9), 6.14 (d, J = 11.2, 1H, H-3), 5.73 (m, 1H, H-10), 5.69 (m, 1H, H-5), 4.32 (s, 2H, H-1), 3.86 (m, 1H, H-7), 2.34 (m, 1H, H-6a), 2.25 (m, 1H, H-6b), 1.81 (d, J = 6.6, 3H, H-11), 1.21 (d, J = 6.3, 3H, H-8), 0.92 (s, 9H, H-TBDMS), 0.08 (s, 6H, H-TBDMS); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 135.1 (C, C-2), 130.3 (CH, C-5), 128.9 (CH, C-4), 125.7 (CH, C-9), 125.6 (CH, C-10), 124.5 (CH, C-3), 67.3 (CH, C-7), 63.8 (CH_2, C-1), 43.0 (CH_2, C-6), 26.0 ($3\times CH_3$, C-TBDMS), 22.8 (CH_3, C-8), 19.0 (CH_3, C-11), 18.4 (C, C-TBDMS), −5.3 ($2\times CH_3$, C-TBDMS); ESIMS *m/z* 297.2 $[M + H]^+$.

RSC Advances Accepted Manuscript

Preparation of MTPA Esters. Compound **2** (1 mg for each) was reacted with either *R*-(–)- or *S*-(+)-MTPA chloride (10 μL) in anhydrous pyridine (500 μL) for 6 h. The reaction mixture was quenched with H_2O and extracted three times with CH_2Cl_2. The organic layers were combined and separated by semipreparative HPLC on ODS (80% MeOH/H_2O, v/v) to afford the *S*-MTPA ester **2a** (1.2 mg, t_R 10.2 min) and *R*-MTPA ester **2b** (0.8 mg, t_R 10.3 min), respectively. By the same procedures, the *S*-MTPA esters **3a**, **5a**, **6a**, **7a**, and **12aa** and *R*-MTPA esters **3b**, **5b**, **6b**, **7b**, and **12ab** were prepared, respectively.

Compound 2a: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.93 (s, 1H, H-3), 6.57 (m, 1H, H-6), 6.04 (m, 1H, H-7), 5.65 (d, *J* =11.3 Hz, 1H, H-5), 5.38 (m, 1H, H-10), 2.74 (dd, *J* = 16.0, 8.3 Hz, 1H, H_a-9), 2.67 (dd, *J* = 16.0, 4.8 Hz, 1H, H_b-9), 1.90 (d, *J* =7.2 Hz, 3H, H-8), 1.35 (d, *J* =6.7 Hz, 3H, H-11); ESIMS *m/z* 411.1 $[M + H]^+$.

Compound 2b: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.62 (s, 1H, H-3), 6.54 (m, 1H, H-6), 6.03 (m, 1H, H-7), 5.53 (d, *J* =11.3 Hz, 1H, H-5), 5.37 (m, 1H, H-10), 2.67 (dd, *J* = 16.2, 8.5 Hz, 1H, H_a-9), 2.62 (dd, *J* = 16.2, 5.4 Hz, 1H, H_b-9), 1.89 (d, *J* =7.0 Hz, 3H, H-8), 1.40 (d, *J* =6.3 Hz, 3H, H-11); ESIMS *m/z* 411.1 $[M + H]^+$.

Compound 3a: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.85 (s, 1H, H-3), 5.26 (m, 1H, H-7), 4.89 (t, *J* =10.2 Hz, H-5), 2.67 (m, 1H, H_a-6), 2.64 (m, 1H, H_b-6), 2.32 (t, *J* =9.0 Hz, 2H, H-9), 1.56 (m, 2H, H-10), 1.38 (d, *J* =8.3, 3H, H-8), 0.97 (t, *J* =9.2 Hz, 3H, H-11); ESIMS *m/z* 413.1 $[M + H]^+$.

Compound 3b: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.94 (s, 1H, H-3), 5.28 (m, 1H, H-7), 5.07 (t, *J* =10.3 Hz, H-5), 2.74 (m, 1H, H_a-6), 2.71 (m, 1H, H_b-6), 2.34 (t, *J* =9.0 Hz, 2H, H-9), 1.60 (m, 2H, H-10), 1.31 (d, *J* =8.3, 3H, H-8), 0.97 (t, *J* =9.2 Hz, 3H, H-11); ESIMS *m/z* 413.1 $[M + H]^+$.

Compound 5a: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.57 (m, 1H, H-7), 6.35 (t, *J* = 11.0 Hz, 1H, H-6),

RSC Advances Accepted Manuscript

5.45 (t, J =10.7 Hz, H-5), 5.42 (d, J =16.7 Hz, 1H, H_a-8), 5.34 (d, J =9.8 Hz, 1H, H_b-8), 5.32 (m, 1H, H-4), 5.18 (dd, J =9.0, 6.8 Hz, 1H, H-3), 2.70 (m, 1H, H-2), 1.79 (m, 1H, H_a-9), 1.66 (m, 1H, H_b-9), 1.61 (m, 1H, H_a-10), 1.40 (m, 1H, H_b-10), 0.91 (t, J = 7.2 Hz, 3H, H-11); ESIMS m/z 571.1 $[M + H]^+$.

Compund 5b: ^1H NMR ($CDCl_3$, 500 MHz) δ 6.45 (m, 1H, H-7), 6.29 (t, J = 11.0 Hz, 1H, H-6), 5.43 (t, J =10.1 Hz, H-5), 5.37 (d, J =16.7 Hz, 1H, H_a-8), 5.29 (d, J =9.8 Hz, 1H, H_b-8), 5.29 (m, 1H, H-4), 5.18 (dd, J =9.2, 5.8 Hz, 1H, H-3), 2.78 (m, 1H, H-2), 1.84 (m,1H, H_a-9), 1.72 (m, 1H, H_b-9), 1.63 (m, 1H, H_a-10), 1.48 (m, 1H, H_b-10), 0.95 (t, J = 7.2 Hz, 3H, H-11); ESIMS m/z 571.1 $[M + H]^+$.

Compound 6a: ^1H NMR ($CDCl_3$, 500 MHz) δ 6.38 (m, 1H, H-7), 6.35 (m, 1H, H-6), 5.74 (dd, J = 14.5, 5.9 Hz, 1H, H-5), 5.69 (s, H-3), 5.60 (m, 1H, H-9), 5.29 (d, J = 15.7 Hz, 1H, H_a-8), 5.17 (d, J = 9.0 Hz, 1H, H_b-8), 4.66 (d, J = 6.9 Hz, 1H, H-4), 4.50 (d, J = 12.6 Hz, 1H, H_a-1), 4.38 (d, J = 12.6 Hz, 1H, H_b-1), 2.34 (m, 1H, H_a-10), 2.06 (m,1H, H_b-10), 0.87 (t, J = 7.7 Hz, 3H, H-11); ESIMS m/z 397.1 $[M + H]^+$.

Compound 6b: ^1H NMR ($CDCl_3$, 500 MHz) δ 6.37 (m, 1H, H-7), 6.32 (m, 1H, H-6), 5.76 (s, H-3), 5.73 (dd, J = 14.5, 6.3 Hz, 1H, H-5), 5.66 (m, 1H, H-9), 5.28 (d, J = 15.7 Hz, 1H, H_a-8), 5.18 (d, J = 9.0 Hz, 1H, H_b-8), 4.50 (d, J = 6.3 Hz ,1H, H-4), 4.50 (d, J = 12.3 Hz, 1H, H_a-1), 4.39 (d, J = 12.3 Hz, 1H, H_b-1), 2.34 (m, 1H, H_a-10), 2.16 (m,1H, H_b-10), 1.0 (t, J = 7.7 Hz, 3H, H-11); ESIMS m/z 397.1 $[M + H]^+$.

Compound 7a: ^1H NMR ($CDCl_3$, 500 MHz) δ 6.07 (s, 1H, H-2), 5.90 (s, 1H, H-4), 5.69 (m, H-7), 5.25 m, 1H, H_a-8), 5.12 (m, 1H, H_b-8), 2.63 (m, 1H, H_a-6), 2.53 (s, 1H, H-5), 2.52 (m, 1H, H_b-6), 2.14 (m,1H, H_a-9), 2.11 (m,1H, H_b-9), 1.56 (m,1H, H_a-10), 1.48 (m,1H, H_b-10), 0.90 (t, J = 6.5 Hz,

RSC Advances Accepted Manuscript

3H, H-11); ESIMS *m/z* 397.1 $[M + H]^+$.

Compound 7b: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.10 (s, 1H, H-2), 5.87 (s, 1H, H-4), 5.67 (m, H-7), 5.23 (m, 1H, H_a-8), 5.10 (m, 1H, H_b-8), 2.61 (m, 1H, H_a-6), 2.47 (m, 1H, H_b-6), 2.42 (s, 1H, H-5), 2.34 (m,1H, H_a-9), 2.27 (m,1H, H_b-9), 1.64 (m,1H, H_a-10), 1.58 (m,1H, H_b-10), 0.97 (t, J = 6.5 Hz, 3H, H-11); ESIMS *m/z* 397.1 $[M + H]^+$.

Compound 12aa: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.49 (dd, J = 15.5, 11.2 Hz,1H, H-4), 6.41 (d, J =16.0 Hz, 1H, H-9), 6.05 (d, J =11.7 Hz, H-3), 5.71 (m, 1H, H-10), 5.52 (m, 1H, H-5), 5.22 (m, 1H, H-7), 4.30 (s, 2H, H-1), 2.45 (m, 1H, H_a-6), 2.40 (m, 1H, H_b-6), 1.80 (d, J =6.6 Hz, 3H, H-11), 1.35 (d, J =6.2 Hz, 3H, H-8), 0.92 (s, 9H, H-TBDMS), 0.08 (s, 6H, H-TBDMS); ESIMS *m/z* 513.1 $[M + H]^+$.

Compound 12ab: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.57 (dd, J = 15.5, 11.2 Hz,1H, H-4), 6.44 (d, J =16.1 Hz, 1H, H-9), 6.11 (d, J =11.7 Hz, H-3), 5.71 (m, 1H, H-10), 5.63 (m, 1H, H-5), 5.21 (m, 1H, H-7), 4.32 (s, 2H, H-1), 2.51 (m, 1H, H_a-6), 2.44 (m, 1H, H_b-6), 1.80 (d, J =6.6 Hz, 3H, H-11), 1.28 (d, J =6.2 Hz, 3H, H-8), 0.92 (s, 9H, H-TBDMS), 0.08 (s, 6H, H-TBDMS); ESIMS *m/z* 513.1 $[M + H]^+$.

Chemical transformation of 16 to 15. To compound **16** (2 mg) in MeOH (1 mL) was added 15 μL concentrated HCl (37%). After stirred at r.t for 1 h, the reaction mixture was subjected to HPLC separation over ODS (10% MeOH/H_2O, v/v) to yield **15** (t_R 15.5 min, 1.2 mg).

Chemical transformation of 17 to 14. To compound **17** (2 mg) in methanol (1 ml) was added 500 μL NaOH solution (2%). After stirred at r.t. for 5 h, the reaction was neutralized to pH 7 by 1*M* HCl. The obtained mixture was concentrated and then was added to 1.5 mL H_2O. The H_2O layer was extracted twice with 3 mL EtOAc and the combined EtOAc extracts were concentrated and

RSC Advances Accepted Manuscript

purified by HPLC over ODS (20% $MeOH/H_2O$, v/v) to yield **14** (t_R 4.6 min, 1.5 mg).

X-ray crystal data for 14 and 15: Colorless crystals of **14** and **15** were obtained in $MeOH-H_2O$ (1:1, v/v). Crystal data of **14** were obtained on a Bruker APEX DUO area detector diffractometer with graphite monochromatic Cu-Kα radiation (λ = 1.54178 Å). Crystal data of **15** were obtained on a Bruker Smart CCD area detector diffractometer with graphite monochromatic Mo-Kα radiation (λ = 0.71073 Å). Crystallographic data for **14** and **15** have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication numbers CCDC 995364 and 995361, respectively. These data can be obtained free of charge from the Cambridge Crystallographic Data Centre *via* www.ccdc.cam.ac.uk/data_request/cif.

Crystal Data for Aspergone N (14). Monoclinic, $C_{10}H_{16}O_5$ H_2O; space group $P2(1)$ with a = 15.2823(12) Å, b = 4.5964(3) Å, c = 16.3515(14) Å, V = 1143.39(15) $Å^3$, Z = 4, D_{calcd} = 1.361 Mg/m^3, μ = 0.957 mm^{-1}, and $F(000)$ = 504; unique cell angle (β) = 95.4540(10); crystal size: 0.21 × 0.14 × 0.08 mm^3. T = 293(2) K. The structure was solved by direct methods (SHELXS-97) and expanded using Fourier techniques (SHELXL-97). The final cycle of full-matrix least-squares refinement was based on 3891 unique reflections ($2\theta < 50°$) and 292 variable parameters and converged with unweighted and weighted agreement factors of R1 = 0.1339, wR2 = 0.3268, and R = 0.2463 for I > $2\sigma(I)$ data. Absolute structure parameter is 0.0(12) that could not be used for determined the absolute configuration.

Crystal Data for Aspergone O (15). Trigonal, $C_{10}H_{15}ClO_4$; space group $R3$ with a = 24.752(2) Å, b = 24.752(2) Å, c = 5.0591(6) Å, V = 2684.3(4) $Å^3$, Z = 1, D_{calcd} = 1.307 Mg/m^3, μ = 0.313 mm^{-1}, and $F(000)$ = 1116; crystal size: 0.48 × 0.37 × 0.35 mm^3. T = 298(2) K. The structure was solved by direct methods (SHELXS-97) and expanded using Fourier techniques (SHELXL-97). The final cycle

RSC Advances Accepted Manuscript

of full-matrix least-squares refinement was based on 2066 unique reflections ($2\theta < 50°$) and 137 variable parameters and converged with unweighted and weighted agreement factors of R1 = 0.0378, wR2 = 0.0787, and R = 0.0532 for $I > 2\sigma(I)$ data. Absolute structure parameter is 0.05(7).

Conclusion

In conclusion, eighteen new compounds (**1−18**) were isolated from marine sponge-derived *Aspergillus* sp. strain OUCMDZ-1583. Compounds **5**, **14** and **18** displayed comparable or stronger α-glucosidase inhibition to acarbose (IC_{50} 0.95 *m*M) with the IC_{50} values of 1.30, 1.37 and 0.027 *m*M, respectively. In addition, the α-glucosidase inhibition of fusarentin 6-methyl ether (**22**) was reported here for the first time with an IC_{50} value of 1.19 *m*M.

Acknowledgment. This work was supported by grants from NSFC (Nos. 21172204, 41376148 & 81373298), from 863 programs of China (Nos. 2012AA092104 & 2013AA092901), from NSFC-Shandong Joint Fund for Marine Science Research Centers (No. U1406402), and from the Special Fund for Marine Scientific Research in the Public Interest of China (No. 201405038).

Supporting Information Available: Bioassay protocols used, the NMR spectra of compounds **19−23**, the ITS gene sequences of *Aspergillus* sp. OUCMDZ-1583. These materials are available free of charge via the Internet.

References and Notes

(1) F. Pietra, *Nat. Prod. Rep.*, 1997, **14**, 453–464.

(2) J. W. Blunt, B. R. Copp, R. A. Keyzers, M. H. Munro and M. R. Prinsep, *Nat. Prod. Rep.*, 2013, **30**, 237–323.

RSC Advances Accepted Manuscript

(3) R. K. Pettit, *Appl. Microbiol. Biotechnol.*, 2009, **83**, 19–25.

(4) M. E. Rateb and R. R. Ebel, *Nat. Prod. Rep.*, 2011, **28**, 290–344.

(5) M. Saleem, M. S. Ali, S. Hussain, A. Jabbar, M. Ashraf and Y. S. Lee, *Nat. Prod. Rep.*, 2007, **24**, 1142–1152.

(6) Y. Q. Fan, Y. Wang, P. P. Liu, P. Fu, T. H. Zhu, W. Wang and W. M. Zhu, *J. Nat. Prod.*, 2013, **76**, 1328–1336.

(7) J. K. Zheng, Z. H. Xu, Y. Wang, K. Hong, P. P. Liu and W. M. Zhu, *J. Nat. Prod.*, 2010, **73**, 1133–1137.

(8) Y. B. Zhuang, X. C. Teng, Y. Wang, P. P. Liu, G. Q. Li and W. M. Zhu, *Org. Lett.*, 2011, **13**, 1130–1133.

(9) F. Kong, Y. Wang, P. Liu, T. Dong and W. M. Zhu, *J. Nat. Prod.*, 2014, **77**, 132–137.

(10) C. Li and J. A. Porco, *J. Org. Chem.*, 2005, **70**, 6053–6065.

(11) B. Fang, X. Xie, C. Zhao, P. Jing, H. Li, Z. Wang, J. Gu and X. She, *J. Org. Chem.*, 2013, **78**, 6338–6343.

(12) H. K. Kwon, Y. E. Lee and E. Lee, *Org. Lett.*, 2008, **10**, 2995–2996.

(13) W. Zhang, K.Krohn, S. Draeger and B. Schulz, *J. Nat. Prod.*, 2008, **71**, 1078–1081.

(14) M. Johansson, B. Ä. R. B. E. L. Köpcke, H. Anke and O. Sterner, *J. Antibiot.*, 2002, **55**, 104–106.

(15) R. Ueoka, Y. Nakao, S. Kawatsu, J. Yaegashi, Y. Matsumoto, S. Matsunaga, K. Furihata, R. W. van Soest and N. Fusetani, *J. Org. Chem.*, 2009, **74**, 4203−4207.

(16) L. Di Bari, G. Pescitelli, C. Pratelli, D. Pini and P. Salvadori, *J. Org. Chem.*, 2001, **66**, 4819−4825.

RSC Advances Accepted Manuscript

(17) M. Górecki, E. Jabłońska, A. Kruszewska, A. Suszczyńska, Z. Urbańczyk-Lipkowska, M. Gerards, J. W. Morzycki, W.J. Szczepek and J. Frelek, *J. Org. Chem.*, 2007, **72**, 2906−2916.

(18) S. Chen, F. Ren, S. Niu, X. Liu and Y. Che, *J. Nat. Prod.* 2014, **77**, 9−14.

(19) P. J. Stephens, J. J. Pan and K. J. Krohn, *J. Org. Chem.*, 2007, **72**, 7641–7649.

(20) J. A. Beutler, K. L. McCall, K. Herbert, D. L. Herald, G.R. Pettit, T. Johnson, R. H. Shoemaker and M. R. Boyd, *J. Nat. Prod.*, 2000, **63**, 657−661.

(21) H. D. Flack, *Acta Crystallogr., Sect. A*, 1983, **39**, 876−881.

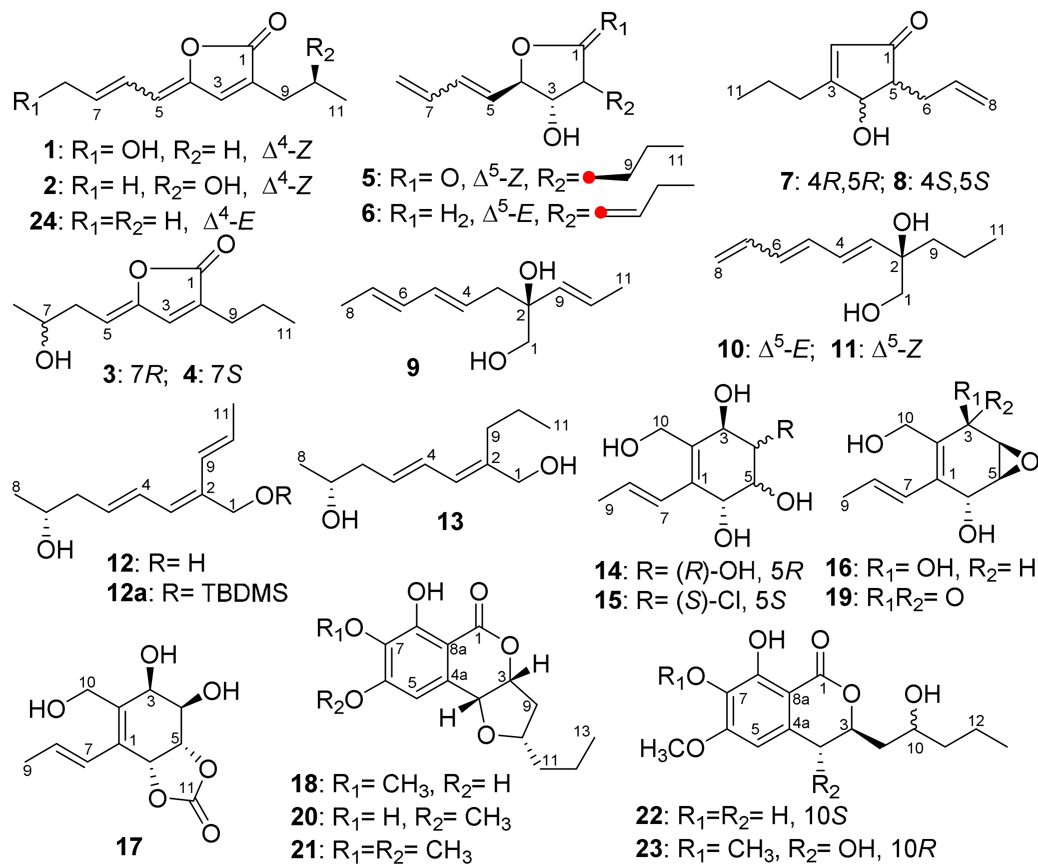
(22) S.V. Nampoothiri, A. Prathapan, O.L. Cherian, K.G. Raghu, V.V. Venugopalan and A. Sundaresan, *Food Chem. Toxicol.*, 2011, **49**, 125–131.

(23) T. Mosmann, *Immunol. Methods*, 1983, **65**, 55–63.

(24) T. Xiong, X.Q. Chen, H. Wei and H. Xiao, *Arch. Med. Sci.*, 2015, **11**, 301–306.

(25) L. L. Zaika, *J. Food Safety*, 1988, **9**, 97–118.

RSC Advances Accepted Manuscript



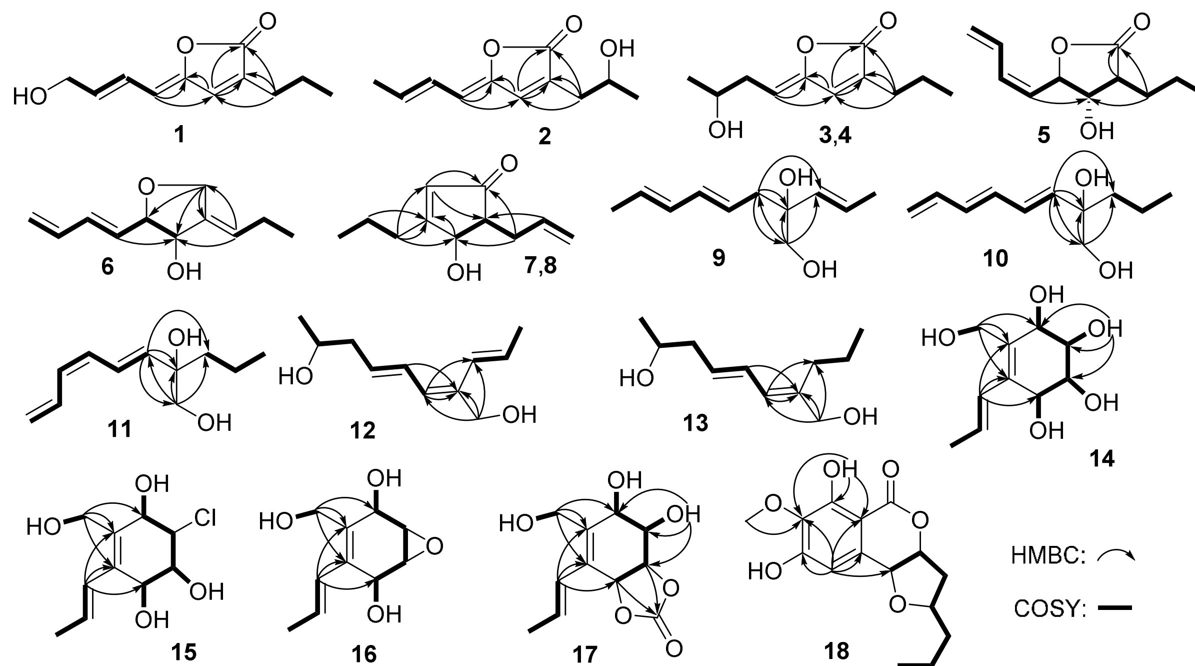


Figure 1. Key COSY and HMBC correlations of **1**−**18**.

RSC Advances Accepted Manuscript

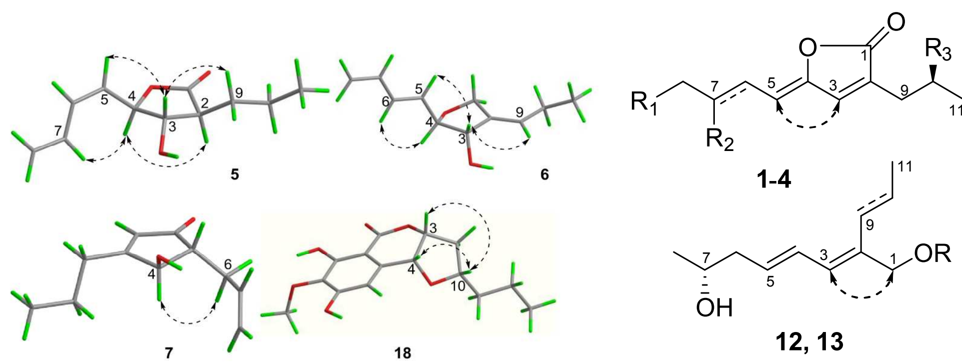


Figure 2. Key NOE correlations of compounds **1−7**, **12**, **13** and **18**.

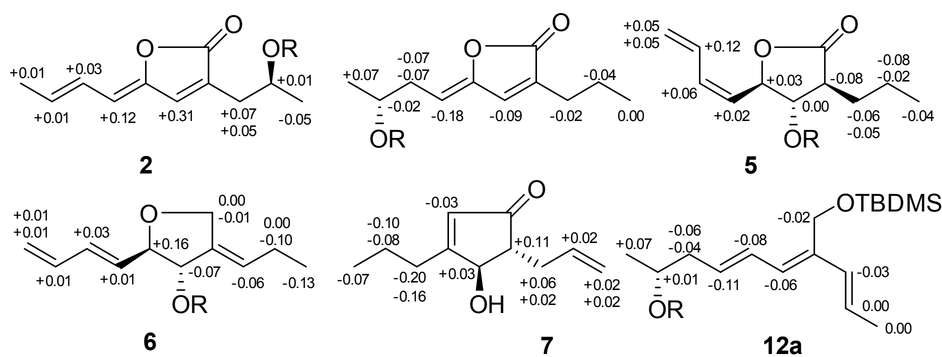


Figure 3. *Δδ* (=$\delta_S-\delta_R$) values for (*S*)- and (*R*)-MTPA esters of **2**, **3**, **5–7** and **12a**.

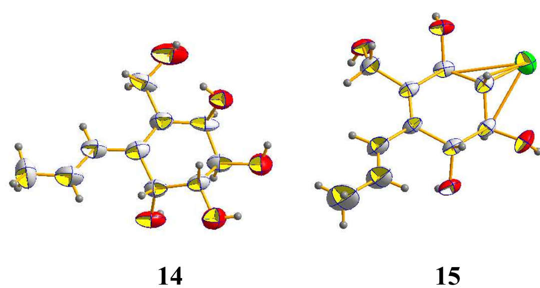


Figure 4. ORTEP drawings of **14** and **15**.

RSC Advances Accepted Manuscript

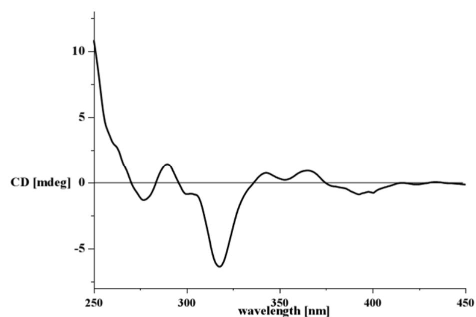


Figure 5. CD curve for the complex of **9** with $Mo_2(OAc)_4$ subtracted from the inherent CD.

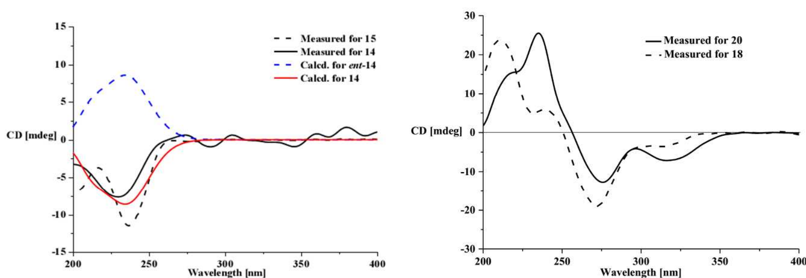


Figure 6. CD curves of **14**, **15**, **18**, **20** and calculated ECD of **14** and *ent*-**14**.

RSC Advances Accepted Manuscript

Table 1. ^{13}C NMR Data for **1−18** (δ_C ppm)

position	**1** [bc]	**2** [bd]	**3** and **4**[bc]	**5** [bd]	**6** [ad]	**7** and **8** [bc]	**9** [ac]	**10** [ac]	**11** [ac]	**12** [ac]	**13** [ac]	**14** [ac]	**15** [ad]	**16** [ac]	**17** [ac]	**18** [bc]
1	170.1, C	171.1, C	170.8, C	175.9, C	70.1, CH_2	206.9, C	68.7, CH_2	69.0, CH_2	68.9, CH_2	62.4, CH_2	65.0, CH_2	133.4, C	134.8, C	131,2, C	126.0, C	168.2, C
2	134.4, C	129.7, C	134.1, C	47.6, CH	140.2, C	128.9, CH	75.0, C	75.2, C	75.3, C	135.7, C	140.9, C	135.8, C	135.4, C	133.2, C	140.2, C	
3	136.6, CH	139.3, CH	137.0, CH	78.4, CH	74.1, CH	180.7, C	41.0, CH_2	140.8, CH	141.6, CH	126.7, CH	124.3, CH	68.5, CH	73.3, CH	65.5, CH	67.4, CH	81.5, CH
4	148.0, C	146.2, C	149.8, C	79.4, CH	86.6, CH	76.7, CH	125.1, CH	132.2, CH	123.6, CH	127.6, CH	128.1, CH	68.7, CH	69.0, CH	54.5, CH	69.8, CH	74.3, CH
5	111.2, CH	113.8, CH	110.4, CH	126.0, CH	132.9, CH	55.3, CH	134.7, CH	134.2, CH	128.8, CH	132.6, CH	130.8, CH	68.3, CH	75.6, CH	54.3, CH	77.9, CH	108.1, CH
6	123.8, CH	124.9, CH	35.8, CH_2	135.0, CH	131.8, CH	31.7, CH_2	131.3, CH	128.3, CH	130.8, CH	43.1, CH_2	43.2, CH_2	67.9, CH	73.4, CH	62.7, CH	75.5, CH	155.4, C
7	137.1, CH	136.5, CH	67.6, CH	131.1, CH	136.9, CH	135.3, CH	128.6, CH	137.8, CH	132.9, CH	66.6, CH	66.7, CH	127.7, CH	126.9, CH	128.3, CH	126.0, CH	134.9, C
8	63.3, CH_2	19.0, CH_3	23.4, CH_3	121.7, CH_2	118.2, CH_2	117.4, CH_2	18.1, CH_3	117.4, CH_2	118.5, CH_2	23.6, CH_3	23.6, CH_3	128.0, CH	129.6, CH	127.0, CH	130.2, CH	155.6, C
9	27.4, CH_2	35.0, CH_2	27.2, CH_2	30.4, CH_2	125.7, CH	32.9, CH_2	133.3, CH	40.0, CH_2	40.0, CH_2	125.8, CH	30.4, CH_2	19.3, CH_3	19.3, CH_3	19.2, CH_3	19.2, CH_3	39.1, CH_2
10	21.0, CH_2	66.3, CH	21.0, CH_2	19.9, CH_2	21.9, CH_2	20.2, CH_2	135.2, CH	16.8, CH_2	16.8, CH_2	125.7, CH	22.1, CH_2	58.1, CH_2	57.5, CH_2	56.8, CH_2	58.2, CH_2	78.7, CH
11	13.7, CH_3	23.3, CH_3	13.8, CH_3	13.9, CH_3	14.6, CH_3	14.0, CH_3	17.9, CH_3	15.2, CH_3	15.2, CH_3	19.3, CH_3	14.6, CH_3				154.8, C	38.1, CH_2

[a] recorded in DMSO-d_6. [b] recorded in $CDCl_3$. [c] measured on a JEOL JNM-ECP 600 spectrometer and δ_C of C-4a, C-8a, C-12, C-13 and 7-OMe for **18** were 131.4 (C), 101.3 C), 19.2 (CH_2), 14.1 (CH_3) and 60.9 (CH_3), respectively. [d] measured on an Agilent 500 MHz DD2 spectrometer.

RSC Advances Accepted Manuscript

Table 2. ^{1}H NMR Data for **1−18** (δ_H ppm, J in Hz)

position	**1**[bc]	**2**[bd]	**3 and 4**[bc]	**5**[bd]	**6**[ad]	**7 and 8**[bc]	**9**[ac]	**10**[ac]	**11**[ac]	**12**[ac]	**13**[ac]	**14**[ac]	**15**[ad]	**16**[ac]	**17**[ac]	**18**[bc]
1					4.27, d (12.5) 4.18, d (12.5)		3.45, d (11.7) 3.43, d (11.7)	3.25, d(12.0); 3.21, d (12.0)	3.26, d (12.5); 3.22, d (12.5)	4.07, s	3.87, s					
2				2.62, m		5.87, s										
3	6.96, s	7.15, s	6.99, s	3.97 dd (7.8, 8.9)	4.22, dd (5.5, 5.4)		2.31, m; 2.32, m	5.79, d (14.8)	5.82, d (15.0)	6.03, d (11.6)	5.97, d (11.0)	4.17, dd (5.3, 4.9)	4.24, dd (10.6, 7.5)	4.52, dd (5.1, 3.2)	4.24, dd (4.8, 3.6)	5.02, m
4				4.98, dd (7.8, 7.8)	4.09, dd (5.4, 5.4)	4.45, d (3.0)	5.48, dt (14.5, 7.4)	6.23, dd (14.8, 10.7)	6.68, dd (15.0, 12.4)	6.51, dd (15.0, 11.2)	6.26, dd (15.4, 11.0)	3.60, m	3.66, dd (10.6, 10.6)	3.35, dd (3.2, 4.0)	3.55, m	4.49, d (3.0)
5	5.67, d (11.6)	5.71, d (11.3)	5.26, t (8.2)	5.41, dd (10.2, 7.8)	5.71, dd (15.7, 6.7)	2.37, m	6.08, dd (14.5, 11.0)	6.28, dd (14.8, 10.7)	6.03, dd (11.3, 12.4)	5.70, m	5.62, dt (15.4, 7.5)	3.55, m	3.34, m	3.28, dd (4.0, 1.9)	4.81, dd (9.6, 7.6)	6.60, s
6	6.74, dd (16.3, 11.6)	6.52, dd (15.6, 11.3)	2.55, m; 2.49, m	6.31, dd (10.2, 11.3)	6.21, dd (15.7, 10.2)	2.56,ddd (14.6, 6.0, 6.2) 2.21, ddd (14.6, 7.9, 7.4)	6.02, dd (14.4, 10.2)	6.19, dd (14.8, 10.2)	5.93, dd (11.2, 11.3)	2.24, m; 2.14, m	2.19, m; 2.10, m	4.23, dd (4.0, 4.0)	3.99, dd (6.5, 6.5)	4.45, dd (7.7, 1.9)	5.60, d (7.6)	
7	6.04, dt (16.3, 4.9)	5.99, dq (15.6, 6.8)	3.96, m	6.68, ddd (16.6, 11.3, 10.3)	6.32, ddd (17.0, 10.2, 10.2)	5.75, m	5.62, dq (14.4, 6.9)	6.36, ddd, (17.1, 10.2, 10.2)	6.82, ddd (16.7, 11.2, 10.4)	3.65, m	3.62, m	6.34, dd (15.7, 1.6)	6.15, d (16.2)	6.36, d (15.8)	6.42, br.d (16.3)	
8	4.24, d (4.9)	1.84, d (7.1)	1.23, d (6.1)	5.37, br.d (16.6); 5.30, br.d (10.3)	5.21 br.d (17.0); 5.07 br.d (10.2)	5.11, br.d (17.0); 5.03, br.d (10.5)	1.73, d (6.9)	5.20, d(17.1); 5.06, d (10.2)	5.24, d (16.7); 5.15, (10.4)	1.03, d (6.1)	1.02, d (6.2)	5.94, dq (15.7, 6.7)	5.82, m	5.91, dq (15.8, 6.7)	5.93, dq (16.3, 6.8)	
9	2.31, t (7.5)	2.53, dd (15.0, 7.9) ; 2.46, dd (15.0, 3.9)	2.32, t (7.6)	1.84, m; 1.58, m	5.36, t (7.3)	2.49, m; 2.36, m	5.44, d (15.5)	1.41, m; 1.42, m	1.43, m; 1.41, m	6.49, dd (16.6, 1.5)	2.07, t (7.3)	1.77, dd (1.6, 6.7)	1.73,d (5.5)	1.76, d (6.7)	1.80, dd (6.6, 1.5)	2.56, ddd (14.6, 8.7, 6.4); 2.13, ddd (14.6, 6.0, 1.4)
10	1.57, m	4.04, m	1.59, m	1.57, m; 1.51, m	2.12, m	1.56, m; 1.62, m	5.73, dq (15.5, 6.6)	1.30, m; 1.18, m	1.30, m; 1.18, m	5.72, m	1.37, m	4.18, dd (12.0, 5.8); 3.92, dd (12.0, 5.8)	4.11, s	4.16, dd (6.0,11.7); 4.13, dd (6.0,11.7)	4.31, dd (12.4, 4.7); 3.95, dd (12.4, 4.7)	4.10, m
11	0.91, t (7.6)	1.21, d (6.4)	0.95, t (7.8)	0.96, t (7.3)	0.91, t (7.7)	0.95, t (7.4)	1.72, d (6.6)	0.83, t (7.3)	0.83, t (6.7)	1.76, dd (6.8, 1.5)	0.86, t (7.6)					1.68, m; 1.55, m
3-OH					5.27, d (5.5)							4.51, d (5.3)	5.39, d (7.5)	5.13, d (5.1)	5.14, d (4.8)	
4-OH												4.27, d (5.9)			5.47, d (5.9)	
5-OH												4.41, d (5.4)	5.41, d (5.6)			
6-OH												4.44, d (4.0)	5.09, d (6.5)	5.16, d (7.7)		
10-OH												4.64, t (5.8)	4.37, t (5.4)	4.35, t (6.)	4.91, t (4.9)	

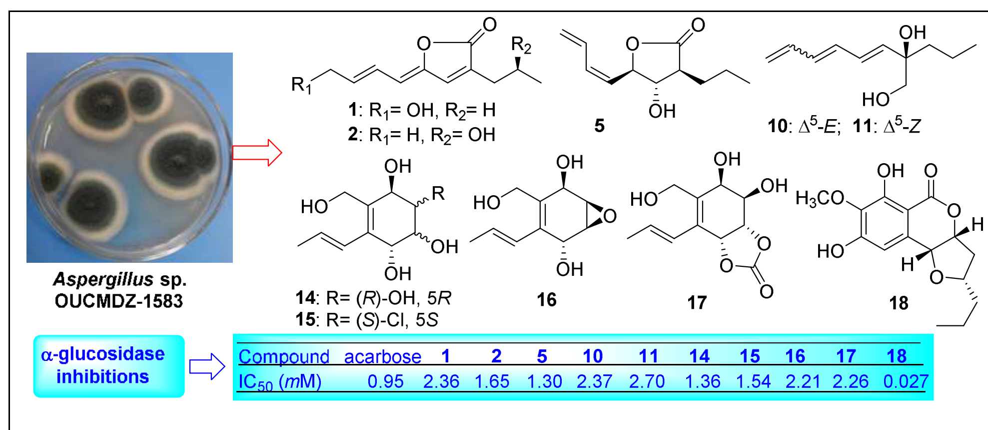
[a] recorded in DMSO-d_6. [b] recorded in $CDCl_3$. [c] measured on a JEOL JNM-ECP 600 spectrometer and δ_H of H-12, H-13, HO-8 and 7-OMe for **18** were 1.41 (m)/1.33 (m), 0.90 (t, J = 7.5 Hz, 3H), 11.5 (s) and 3.97 (s, 3H), respectively. [d] measured on an Agilent 500 MHz DD2 spectrometer.

RSC Advances Accepted Manuscript

Table 3. α-Glucosidase inhibitions of compounds **1**, **2**, **5**, **10**, **11**, **14–18**, and **21–23**.

Compound	Acarbose	**1**	**2**	**5**	**10**	**11**	**14**	**15**	**16**	**17**	**18**	**21**	**22**	**23**
IC_{50} (*m*M)	0.95	2.36	1.65	1.30	2.37	2.70	1.36	1.54	2.21	2.26	0.027	1.65	1.19	1.74

Table of Contents Graphic



RSC Advances Accepted Manuscript