Chinese Chemical Letters 35 (2024) 109089

Contents lists available at ScienceDirect

Chinese Chemical Letters

journal homepage: www.elsevier.com/locate/cclet

# Convergent synthesis and immunological study of oligosaccharide derivatives related to galactomannan from *Antrodia cinnamomea*

Shuying Li[a,1], Weiwei ZhuGe[a,1], Xuan Sun[a,1], Chongzhen Sun[a], Zhaojun Liu[a], Chenghe Xiong[a], Min Xiao[a,b], Guofeng Gu[a,b,*]

[a] National Glycoengineering Research Center and Shandong Key Laboratory of Carbohydrate Chemistry and Glycobiology, Shandong University, Qingdao 266237, China
[b] NMPA Key Laboratory for Quality Research and Evaluation of Carbohydrate-based Medicine, Shandong University, Qingdao 266237, China

ARTICLE INFO

*Article history:*
Received 7 July 2023
Revised 7 September 2023
Accepted 11 September 2023
Available online 17 September 2023

*Keywords:*
*Antrodia cinnamomea*
Galactomannan
Oligosaccharide
Immunostimulatory propriety
Chemical synthesis

ABSTRACT

The galactomannan from *Antrodia cinnamomea* (AC) is characterized as one of the important bioactive components that exhibits potential immunostimulatory propriety. The biological function of its corresponding oligosaccharide fragments has not been revealed yet. In this study, we reported the first chemical synthesis of the series of oligosaccharide fragments related to AC galactomannan *via* the convergent glycosylation strategy. The preliminary immunological evaluation of these synthesized AC oligosaccharides disclosed that the backbone tetrasaccharide **1d** showed the best immunomodulatory ability on enhancing proliferation, phagocytosis and cytokines secretion of Raw264.7 macrophages *in vitro*, indicating its immense potential as an immunostimulant candidate.



Medicinal mushrooms have long been widely concerned because of their multiple pharmaceutical activities, including antibacterial, antiviral, antitumor, antioxidant, and immunomodulatory activities [1–3]. For example, *Antrodia cinnamomea* (AC), known as a medicinal fungus that is uniquely distributed in Taiwan region, has been attracted much attention owing to its extremely broad medicinal applications in treatment of various diseases, such as abdominal pain, drug poisoning, skin itching, hypertension, cancer and liver ailment, *etc.* [4,5]. Polysaccharides in AC have been identified as one of the main pharmacologically active components and showed great potential as immunostimulants or adjuvants in immunotherapy and vaccination development [6]. In 2017, Wu and co-workers reported the isolation and purification of a cold-water soluble galactomannan from AC, and disclosed that it exhibited significant immunostimulatory ability on the phagocytosis and bactericidal activity of J774A.1 macrophages [5]. Further studies revealed that the isolated AC galactomannan could extremely elicit tumor necrosis factor-$\alpha$ (TNF-$\alpha$) and interleukin-6 (IL-6) secretion in J774A.1 macrophages and human dendritic cells speculatively through activating protein kinase C-$\alpha$ (PKC-$\alpha$) and mitogen activated protein kinases (MAPK) signaling pathways after binding to the Toll-like receptor 4 (TLR4) on cell surface [6]. However, the underlying mechanism of its immunomodulation still remain poorly understood; therefore, the screening bioactive oligosaccharide epitope of AC galactomannan is worthwhile for exploration.

Structurally, as depicted in Fig. 1A, the AC galactomannan has been chemically elucidated as the following structure: {→6)-$\alpha$-D-Man*p*-(1→2)-$\alpha$-D-Man*p*-(1→2)-[$\alpha$-D-Man*p*-(1→3)-$\alpha$-D-Man*p*-(1→2)-$\alpha$-D-Man*p*-(1→6)-$\alpha$-D-Gal*p*-(1→6)]-$\alpha$-D-Man*p*-(1→6)-$\alpha$-D-Gal*p*-(1→}$_n$ [5,6]. Its repeating unit is an octasaccharide that composed of a tetrasaccharide backbone chain and a tetrasaccharide side chain, in whose structure D-mannose and D-galactose residues were interconnected *via* $\alpha-1,2$, $\alpha-1,3$ and $\alpha-1,6$ glycosidic bonds, respectively. Thus far, there are no reports on the synthesis and biological activity of the oligosaccharide fragments related to this AC galactomannan. Accordingly, we reported herein the first chemical synthesis of intact repeating unit, octasaccharide **1a**, of AC galactomannan and its substructures, including two hexasaccharide fragments **1b** and **1c**, and three tetrasaccharide fragments **1d–1f**, for in-depth structure-activity relationship immunological study (Fig. 1B). With the structurally defined AC oligosaccharides, we also preliminarily investigated their immunostimulatory activity on viability, phagocytosis and cytokines secretion in Raw264.7 cell line *in vitro*.

Considering that the target molecules **1a–1f** were all even number in chain length, we planned to assembly them by the con-

* Corresponding author at: National Glycoengineering Research Center and Shandong Key Laboratory of Carbohydrate Chemistry and Glycobiology, Shandong University, Qingdao 266237, China.
*E-mail address:* guofenggu@sdu.edu.edu (G. Gu).
[1] These authors contributed equally in this work.

https://doi.org/10.1016/j.cclet.2023.109089
1001-8417/

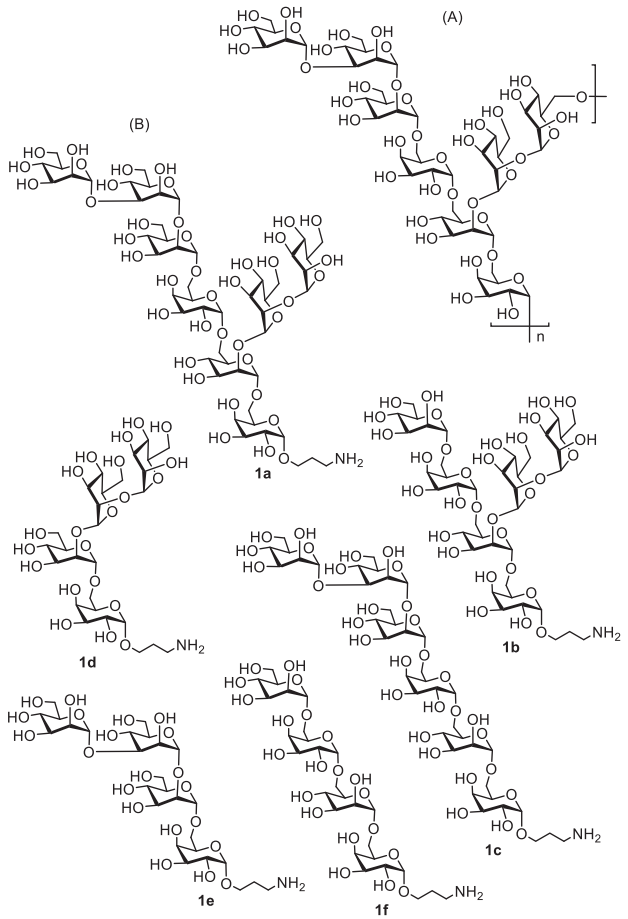
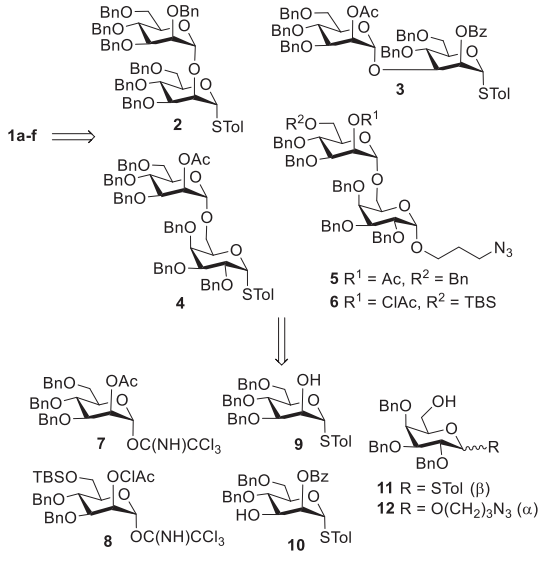



**Fig. 1.** The chemical structures of the octasaccharide repeating unit (A) of AC galactomannan and the target oligosaccharide derivatives **1a–1f** (B) in this study.

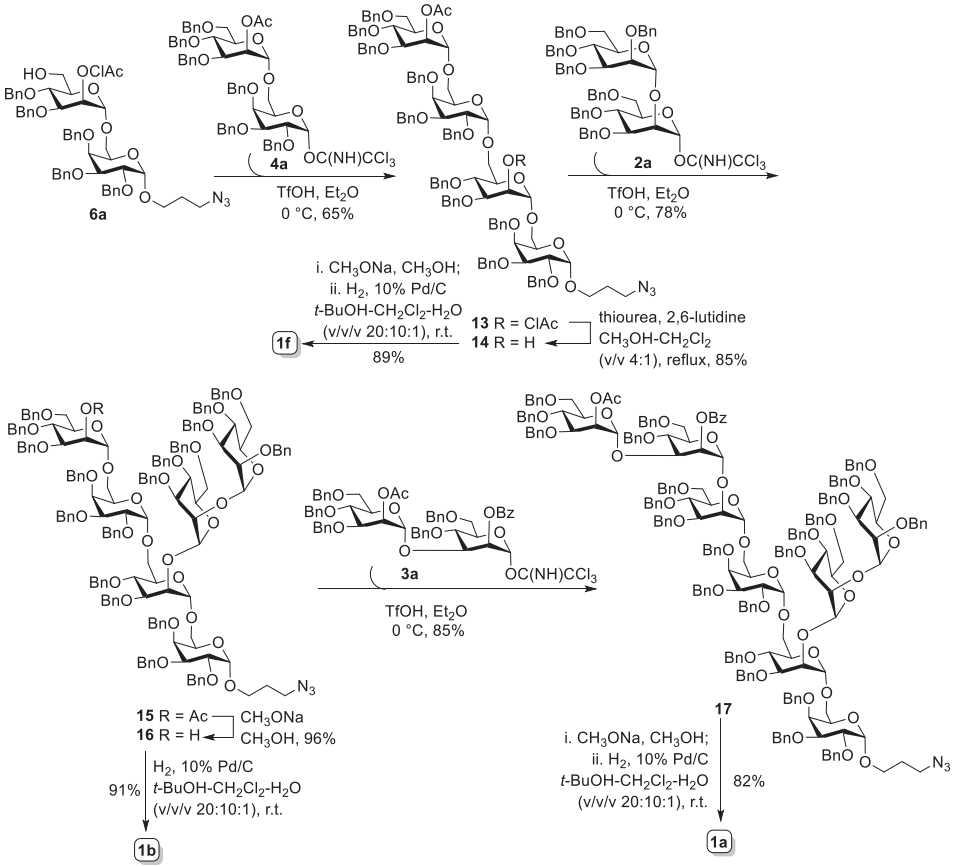



**Scheme 1.** Retrosynthesis of target AC oligosaccharides **1a-1f**.

vergent [2 + 2], [2 + 2 + 2] or [2 + 2 + 2 + 2] glycosylation strategy and employ the acyl-mediated neighboring group participation effect (NGPE) [7] and the ether-assisted solvent effect [8] to efficiently control the dominating formation of $\alpha$-mannosyl and $\alpha$-galactosyl bonds. Accordingly, as shown in Scheme 1, retrosynthetic disconnection of **1a–1f** issued in five disaccharides **2–6** as the key synthetic intermediates. Among them, thioglycosides **2**, **3**, and **4**, which were utilized as glycosyl donors in subsequent oligosaccharide assembly, could be easily prepared from mannosyl imidate donor **7** [9], mannosyl acceptors **9** [10,11], and **10** [12], and galactosyl acceptor **11** [13,14], whereas disaccharides **5** and **6** could be readily assembled from mannosyl imidate donors **7** and **8** with galactosyl acceptors **11** and **12**, and were then served as glycosyl acceptors later after selective removal of the acetyl (Ac) group at O-2 position or *tert*–butyldimethylsilyl (TBS) group at O-6 position of D-mannose residue. Additionally, selective deacetylation of disaccharide **4** would afford latent glycosyl acceptor used in construction of linear hexasaccharide **1c** in our design. The detailed synthetic procedures for monosaccharide intermediates **7–12** were outlined Schemes S1 and S2 (Supporting information).

In our design, as outlined in Scheme S3 (Supporting information), mannosyl trichloroacetimidate **7** acted as a universal glycosyl donor to react with different glycosyl acceptors, *i.e.*, **9–12**, to produce the key disaccharide intermediates **2–5**. For example, the coupling reaction between imidate **7** (1.1 equiv.) and **9** by the catalytic amount of trimethylsilyl triflate (TMSOTf) smoothly generated the desired disaccharide product followed by the conversion of protecting group (Ac→Bn) at mannosyl 2-O position, affording disaccharide **2** in overall 73% yield (3 steps). Thiomannoside **10** was glycosylated with **7** under the promotion of TMSOTf (0.1 equiv.) to yield disaccharide **3** in 87% yield. Likewise, condensation of thiogalactoside acceptor **11** and **7** with TMSOTf as catalyst reposefully furnished disaccharide **4** in excellent yield (92%). Galactoside acceptor **12** was reacted with **7** in activation of TMSOTf to produce disaccharide **5** in high yield of 87%. With the similar protocol established above, disaccharide **6** was smoothly prepared in a moderate yield of 65% from the glycosylation reaction of **12** with 2,6-orthogonally protected imidate donor **8**. The new $\alpha$-mannosyl bonds formed in disaccharides **2–6** were well guaranteed due to the acyl (Ac or ClAc) group-mediated NGPE. Moreover, disaccharyl thioglycosides **2–4** were further converted into more active trichloroacetimidate donors **2a–4a** for purpose of efficient construction of large oligosaccharide chain later. This transformation reaction was achieved through the following two steps: (i) hydrolysis of the reducing thioglycoside with NIS-AgOTf cocatalysis in wet solvent [15], and (ii) subsequent activation of the resultant hemiacetal with trichloroacetonitrile ($Cl_3CCN$) and 1,8-diazabicyclo[5.4.0]undec–7-ene (DBU) [16]. Furthermore, deacetylation of **5** under Zempén condition gave disaccharide acceptor **5a** in 98% yield, which was expected to subsequently assembly target tetrasaccharides **1d** and **1e**. Alternatively, selective cleavage of TBS group in **6** was carried out with $BF_3{\cdot}Et_2O$ in chloroform [17], successfully providing disaccharide acceptor **6a** in excellent yield, which would be used for construction of target molecules **1a-1c** and **1f** in following study.

The octasaccharide **1a**, as shown in Scheme 2, was designedly assembled *via* [2 + 2 + 2 + 2] strategy that required disaccharide building blocks **2a**, **3a**, **4a**, and **6a** involved. Given that the construction of $\alpha$-galactosyl bond would be the most difficult task in the synthesis, we commenced with the synthesis of tetrasaccharide **13** first (Scheme 2). Unfortunately, direct glycosylation of **6a** and thioglycoside **4** under different catalysts and reaction conditions (Table S1 in Supporting information) proceeded inefficiently, which merely generated the desired **13** in 10%−20% yields. However, when changing to disaccharyl trichloroacetimidate **4a** as glycosyl donor, the above glycosylation reaction produced **13** with a satisfying yield. Up to 65% yield of **13** could be achieved with triflic acid (TfOH) as the promotor in $Et_2O$ solvent. The newly formed $\alpha$-galactosyl bond in **13** was readily judged from the small coupling constant ($^3J_{1,2} = 3.0$ Hz) of H-1$^{Gal}$ signal at $\delta$ 5.10 ppm in $^1$H NMR spectrum. It should be noted here that $\beta$-isomeric product was also observed in above [2 + 2] reaction but inseparable from



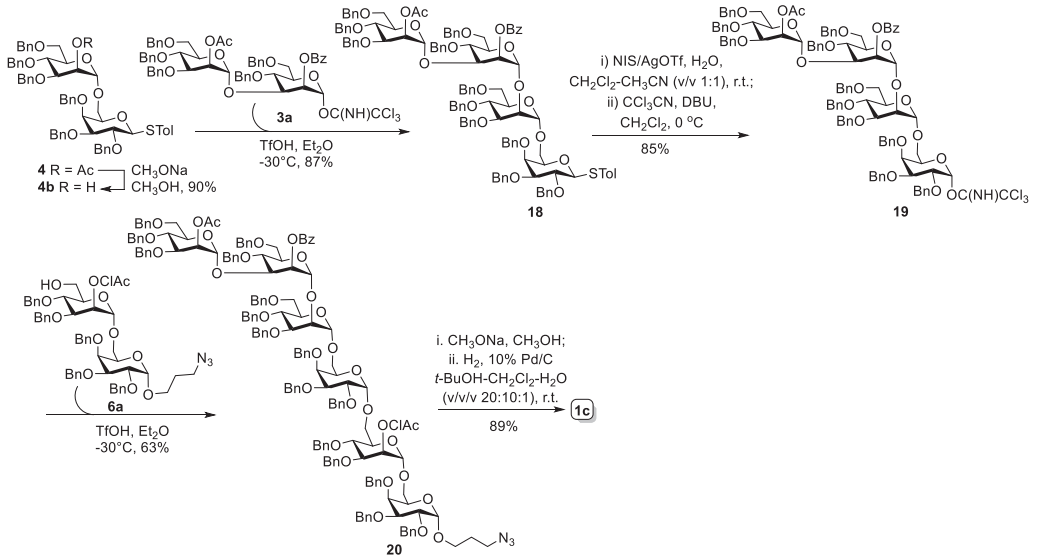

**Scheme 2.** Synthesis of the octasaccharide **1a** of the galactomannan intact repeating unit and its hexa- and tetra-saccharide fragments, **1b** and **1f**.

**13**. Thereafter, thiourea-promoted cleavage of chloroacetyl group (ClAc) [18] on the mannosyl 2-O-postion furnished tetrasaccharide acceptor **14** in 85% yield, which was then glycosylated with imidate donor **2a** (1.2 equiv.) in presence of catalytic amount of TfOH in $Et_2O$ successfully afforded hexasaccharide **15** in 78% yield. All $\alpha$-glycosidic bonds in **15** were undoubtedly confirmed from the $^1J_{C\text{-}1,\,H\text{-}1}$ coupling constants (>169 Hz) between C-1s and H-1s in its $^1$H-coupled HSQC spectrum [19,20]. Similarly, trace amount of inseparable $\beta$-isomer generated during the [4 + 2] glycosidation reaction. Next, treatment of **15** with $CH_3ONa$ in $CH_3OH$ smoothly provided 2-OH hexasaccharide acceptor **16** in excellent yield. The more active disaccharide imidate **3a** was preferentially chosen here for further glycosylation. As expected, condensation of **16** and **3a** (1.2 equiv.) under the promotion of TfOH accomplished perfectly the synthesis of fully protected octasaccharide **17** (85% yield). Again, the $^1J_{C\text{-}1,\,H\text{-}1}$ coupling constants observed from $^1$H-coupled HSQC spectrum were all over 169 Hz, indicating the formation of $\alpha$-glycosidic bonds in **17**. Finally, deacylation of **17** with $CH_3ONa$ in $CH_3OH$, followed by Pd-catalyzed hydrogenolytic debenzylation and azide reduction in $t$-BuOH−$CH_2Cl_2$−$H_2O$ co-solvents (v/v/v, 20:10:1) furnished target octasaccharide **1a** in 82% yield, after purification by size-exclusion chromatography on Sephedex G-10 column. Furthermore, global deprotection of **16** and **14** by Zempén condition and/or hydrogenolysis with 10% Pd/C as the catalyst generated the desired hexasaccharide **1b** (91%) and tetrasaccharide **1f** (89%), respectively, which were further purified on Sephedex G-10 column.

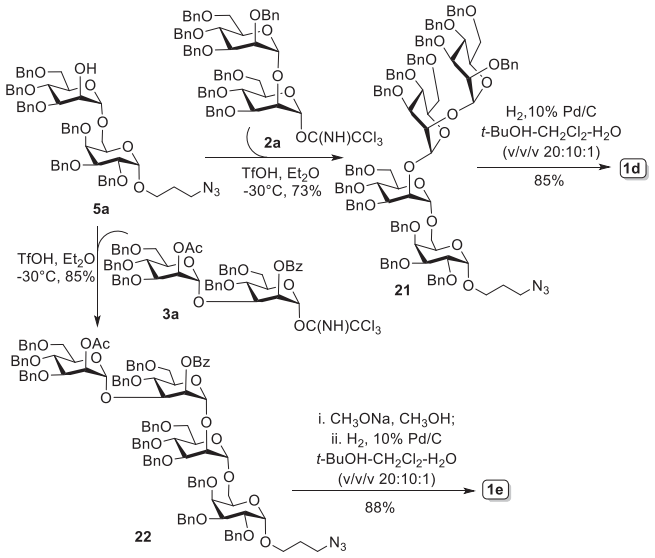
As outlined in Scheme 3, the linear hexasaccharide **1c** was assembled *via* a convergent [2 + 2 + 2] strategy with disaccharide building blocks **3a**, **4b** and **6a**. Glycosylation reaction of acceptor **4b**, generated from Zempén deacetylation of thioglycoside **4**, and imidate **3a** using TfOH (0.1 equiv.) in $Et_2O$ gave tetrasaccharide **18** in high yield (87%). The transformation of tetrasaccharide thioglycoside **18** into trichloroacetimidate form **19** (85%) went smoothly in two steps, as described for preparation of disaccharide imidates **2a–4a**. Then, the active imidate donor **19** reacted with acceptor **6a** under the activation of TfOH to provide the fully protected hexasaccharide **20** in 63% yield. This [4 + 2] reaction was not well stereoselectively controlled by solvent effect of $Et_2O$ with production of inseparable $\beta$-isomer (*ca.* 30%). Likewise, the small coupling constant ($^3J_{1,2} = 3.6$ Hz) of H-$1^{Gal}$ signal at $\delta$ 5.09 ppm in $^1$H NMR confirmed the $\alpha$-configuration of the newly formed galactoside. Eventually, the target hexasaccharide **1c** was obtained by global deprotection in two steps, as described above for **1a**, in overall 89% yield.

The synthesis of tetrasaccharides **1d** and **1e** using disaccharide **2a**, **3a**, and **5a** was shown in Scheme 4. Acceptor **5a** was coupled with imidate **2a** to afford predominantly tetrasaccharide **21** ($\alpha/\beta$ = 10:1) in good yield of 73%, which was subjected to hydrogenolytic debenzylation and purification as described above to give desired tetrasaccharide **1d** in 85% yield. The $^1J_{C\text{-}1,\,H\text{-}1}$ values (>169 Hz) between C-1s and H-1s calculated from $^1$H-coupled HSQC spectrum guaranteed the $\alpha$-glycosidic bonds in **21**. Alternatively, the reaction of **5a** with imidate **3a** promoted by TfOH in dry $Et_2O$ produced tetrasaccharide **22** in 85% yield. Ultimately, global deprotection of **22** using the aforementioned protocols for **1a** and **1c** afforded target tetrasaccharide **1e** in 88% yield. Collectively, the target AC oligosaccharides **1a-1f** together with all synthetic intermediates involved above were completely characterized by 1D- and 2D-NMR and MS spectra.

With enough amount of the synthesized AC oligosaccharides **1a–1f** in hand, we thereby explored preliminarily their immunostimulatory activity toward Raw264.7 cells. First, the viability of




**Scheme 3.** Synthesis of the linear hexasaccharide **1c**.




**Scheme 4.** Synthesis of the tetrasaccharide **1d** and **1e**.

Raw264.7 cells incubated with different concentration of **1a–1f** was examined using CCK-8 assay [21]. As shown in Fig. 2A, except for tetrasaccharide **1f** that showed none proliferative effect against macrophage cells, all other five oligosaccharides **1a–1e** exhibited strong proliferation effect. The cell viability was significantly promoted with oligosaccharide concentrations under 25 μg/mL, whereas its growths were decreased markedly as oligosaccharide concentration exceeding this dose. Among tested AC oligosaccharides, tetrasaccharide **1d**, hexasaccharide **1b**, and octasaccharide **1a**, which all contained the tetrasaccharide backbone structure, exhibited much stronger immunostimulatory activity towards Raw264.7 cell viability than those having the tetrasaccharide sidechain structure, *i.e.* tetrasaccharide **1e** and hexasaccharide **1c**. Most notably, tetrasaccharide **1d** exerted the best proliferation ability at concentration of 25 μg/mL, indicating its great immunostimulatory capacity for further investigation.

Next, the effect of **1a–1e** on the phagocytic activity of Raw264.7 cells were characterized *via* measuring neutral red uptake assay [22]. These studies were carried out at the optimal concentration of 25 μg/mL of **1a–1e** using lipopolysaccharide (LPS, 1.0 μg/mL), a well characterized macrophage stimulator, as the positive control. As depicted in Fig. 2B, compared with the blank group, the Raw264.7 cells treated with octasaccharide **1a**, hexasaccharide **1b**, or tetrasaccharide **1d** exhibited significantly phagocytic ability to phagocytize neutral red, whereas those cells treated with hexasaccharide **1c** or tetrasaccharide **1e** did not show phagocytosis at all. Likewise, tetrasaccharide **1d** exerted the best ability on influencing macrophage phagocytosis towards intake of neutral red. Additionally, tetrasaccharide **1d** was also more effective than LPS in enhancing the phagocytic activity of macrophages, further revealing the excellent immunomodulatory role of **1d** on Raw264.7 macrophages.

The effect of **1a–1e** (25 μg/mL) on production of pro-inflammatory cytokines, including NO, TNF-$\alpha$, IL-6, and IL-1$\beta$ from Raw264.7 macrophages was further evaluated using LPS (1.0 μg/mL) as a positive control. Clearly, oligosaccharides **1a**, **1b** and **1d** significantly stimulated the secretion of NO, TNF-$\alpha$, and IL-6 in different degree at the tested concentration (Fig. 2C and Figs. S1A-C in Supporting information). Moreover, **1b** and **1d** could promote Raw264.7 macrophages to accumulate IL-1$\beta$ production as compared with the blank control. However, **1c** and **1e** did not exhibit the stimulatory activity on the induction of NO, TNF-$\alpha$, IL-6, and IL-1$\beta$ levels (except for **1e** on TNF-$\alpha$). These findings, together with the above proliferation and phagocytosis results, have again demonstrated that AC oligosaccharides containing tetrasaccharide backbone structure, especially for **1d**, have exerted the excellent immune-enhancing activity on Raw264.7 cells and could be used as a potential immunostimulatory agent.

As well known, macrophage activation is usually associated with mitogen-activated protein kinase (MAPK, including JNK, p38, and ERK1/2), nuclear factor-kappa B (NF-$\kappa$B), and/or phosphatidylinositide 3-kinase (PI3K)/AKT signaling pathways [23]. Based on the observed significant immunostimulatory activity of tetrasaccharide **1d** on macrophages, the potential signaling pathways activated by **1d** were further probed. To this end, Raw264.7 cells were pretreated with SP6000125 (JNK inhibitor), SB203580 (p38 inhibitor), PD98059 (ERK1/2 inhibitor), BAY11–7082 (IKK inhibitor) or LY294002 (PI3K inhibitor) for 2 h and then co-treated with **1d** (25 μg/mL) for 24 h. The secretion level of NO in Raw264.7 cells was determined by Griess assay [24]. As shown in Fig. 2D, the NO production induced by **1d** was significantly decreased in macrophages that pretreated with SP600125 inhibitor. However, SB203580 (p38 inhibitor) and BAY11–7082 (IKK inhibitor) atten-

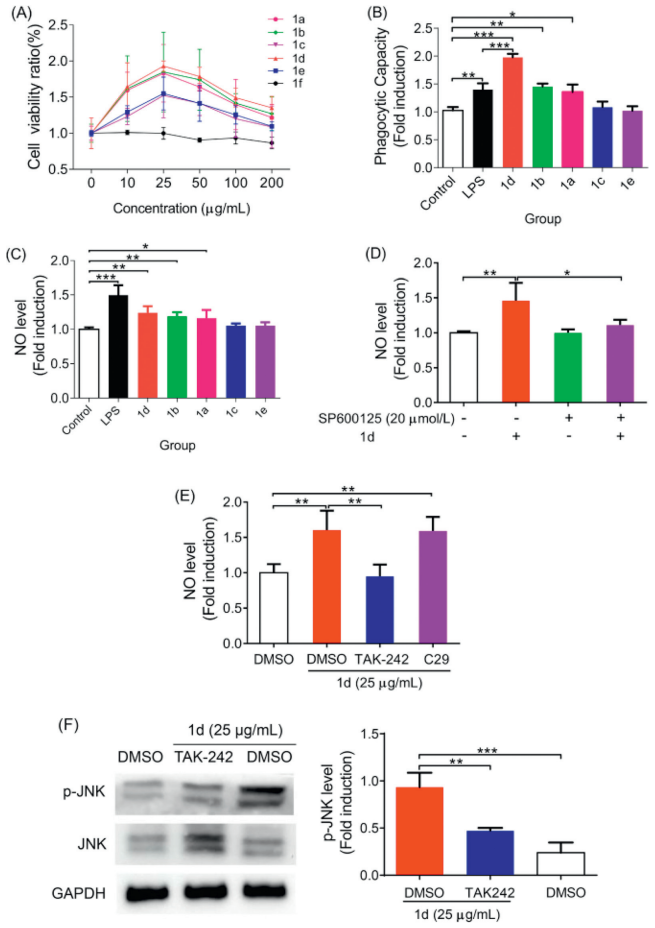



**Fig. 2.** The effect of AC oligosaccharides **1a–1f** on proliferation (A), phagocytosis (B) and NO secretion (C) of Raw264.7 macrophages. (D) The effect of JNK signaling pathway on **1d**-mediated NO production in Raw264.7 cell lines. The effect of TLR4 on NO production (E) and JNK signaling pathway activation (F) in Raw264.7 cell line. Results are expressed as the means ± SD obtained from triplicate experiments. $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$.

uated slightly the NO production on **1d**-mediated macrophages (Figs. S2A and B in Supporting information). Furthermore, neither inhibition of ERK1/2 signaling pathway by PD98059 nor blockade of PI3K/AKT signaling pathway by LY294002 affected **1d**-induced NO production (Figs. S2C and D in Supporting information). Our findings suggested that tetrasaccharide **1d** might stimulate the NO secretion mainly through the activation of JNK signaling pathway. This speculation was further demonstrated by the results that the inhibition of JNK signaling pathway with SP600125 could dramatically suppressed the expression of other immune cytokines (IL-1$\beta$, IL-6, and TNF-$\alpha$) promoted by **1d** (Figs. S3A-C in Supporting information). To further confirm the effect of **1d** on the activation of JNK signaling pathway, the phosphorylation level of JNK protein was measured by western blot analysis [25] after treating Raw264.7 cells with 10 or 25 μg/mL of **1d**. As shown in Fig. S3D (Supporting information), the phosphorylation level of JNK protein in **1d**-treated Raw264.7 cells were significantly enhanced as compared with the blank control. These results indicated that tetrasaccharide **1d** might activate the JNK signaling pathway.

It has been well documented that functional glycan ligands could bind to Toll-like receptors (TLRs), such as TLR2 and TLR4, to further activate MAPK signaling pathway, leading to the secretion of various immune factors [26]. Accordingly, we explored the potential role of TLR2 and TLR4 in the activation of immune factors by tetrasaccharide **1d** in Raw264.7 cells. As compared to the cells treated with **1d** alone, the inhibition of TLR2 with C29 inhibitor (100 μmol/L) had no influence on the NO production triggered by **1d** in cells, whereas the inhibition of TLR4 by TAK242 inhibitor (5 μmol/L) significantly reduced **1d**-stimulated NO production in cells (Fig. 2E). Furthermore, the significant down-regulated expression of TNF-$\alpha$, IL-6 and IL-1$\beta$ by **1d** has been observed in Raw264.7 cells that pretreated with TAK242 (Figs. S4A-C in Supporting information). In addition, the inhibition of TLR4 with TAK242 could suppress the phosphorylation of JNK protein induced by **1d** as comparison to that treated with **1d** alone (Fig. 2F). These results indicated that TLR4 might be the main receptor for tetrasaccharide **1d** binding to activate macrophages *via* the JNK signaling pathway.

In summary, we described here the efficient synthesis of homogenous and structurally well-defined AC galactomannan tetra-, hexa- and octa-saccharides **1a–1f** *via* highly convergent [2 + 2], [2 + 2 + 2] and [2 + 2 + 2 + 2] glycosylation strategy. In these syntheses, disaccharide trichloroacetimidates **2a–4a** were proved to be more active glycosyl donors, as compared with their corresponding thioglycosides **2–4**, to efficiently complete the synthesis of target molecules. Also, all $\alpha$-glycosidic bonds in **1a–1f** were stereoselectively controlled *via* the neighboring group participation effect (Ac, ClAc, or Bz group) and the solvent effect ($Et_2O$ solvent) and successfully achieved in satisfying yields. The preliminary immunostimulant effect of the synthesized oligosaccharides **1a–1f** on Raw264.7 macrophages have disclosed that oligosaccharides carried with tetrasaccharide backbone structure of AC galactomannan polysaccharide, such as octasaccharide **1a**, hexasaccharide **1b**, and tetrasaccharide **1d**, could significantly promote the proliferation, phagocytosis, and cytokine production of NO, TNF-$\alpha$, IL-6 and IL-1$\beta$ in macrophages. Most particularly, tetrasaccharide **1d** exerted the best immunomodulatory activity towards Raw264.7 cells. Furthermore, the inhibition of TLR4 and JNK signaling pathway with TAK-242 and SP6000125 inhibitors, respectively, extremely reduced the **1d**-induced secretion of pro-inflammation cytokines and, as well, the JNK phosphorylation. All these findings indicated that TLR4 receptor on the surface of macrophages might be the putative receptor for AC oligosaccharide binding first, which in turn activates intracellular JNK signaling pathway and thus induces cytokine production. Collectively, based on the fine structure and excellent immunomodulatory activity observed in this study, the synthetic tetrasaccharide **1d** has been identified as a potential immunomodulator candidate to enhance immunity. Its immunostimulatory potential on nonspecific immune responses is undergoing and will be communicated in due course.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Acknowledgments

This work was supported by grants from the National Key Research and Development Program of China (No. 2018YFA0902000) and the National Natural Science Foundation of China (No. 21877074).

## Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.cclet.2023.109089.

## References

[1] A. Bhambri, M. Srivastava, V.G. Mahale, et al., Front. Microbiol. 13 (2022) 837266.

[2] V. Bell, C.R.P.G. Silva, J. Guina, T.H. Fernandes, Front. Nutr. 9 (2022) 1050099.
[3] J. Xu, R. Shen, Z. Jiao, et al., Nutrients 14 (2022) 2622.
[4] S.H. Tu, C.H. Wu, L.C. Chen, et al., Agric. Food Chem. 60 (2012) 3612–3618.
[5] N. Perera, F.L. Yang, C.M. Chang, et al., Org. Lett. 19 (2017) 3486–3489.
[6] N. Perera, F.L. Yang, Y.T. Lu, et al., Int. J. Biol. Sci. 14 (2018) 1378–1388.
[7] C.S. Chao, C.Y. Lin, S. Mulani, W.C. Hung, K.K.T. Mong, Chem. Eur. J. 17 (2011) 12193–12202.
[8] A. Kafle, J. Liu, L. Cui, Can. J. Chem. 94 (2016) 894–901.
[9] F. Yamazaki, S. Sato, T. Nukada, Y. Ito, T. Ogawa, Carbohydr. Res. 201 (1990) 31–50.
[10] K. Chayajarus, D.J. Chambers, M.J. Chughtai, A.J. Fairbanks, Org. Lett. 6 (2004) 3797–3800.
[11] D. Wang, D.C. Xiong, X.S. Ye, Chin. Chem. Lett. 29 (2018) 1340–1342.
[12] K.K.T. Mong, K.S. Shiau, Y.H. Lin, K.C. Cheng, C.H. Lin, Org. Biomol. Chem. 13 (2015) 11550–11560.
[13] C. Li, Y. Sun, J. Zhang, et al., Carbohydr. Res. 376 (2013) 15–23.
[14] C.H. Wang, S.T. Li, T.L. Lin, et al., Angew. Chem. Int. Ed. 52 (2013) 9157–9161.
[15] D. Wang, W. Zhuge, Z. Guo, G. Gu, Carbohydr. Res. 442 (2017) 41–51.
[16] R.R. Schmidt, J. Michel, Angew. Chem. Int. Ed. 19 (1980) 731–732.
[17] K. Ruda, J. Lindberg, P.J. Garegg, S. Oscarson, P. Konradsson, J. Am. Chem. Soc. 122 (2000) 11067–11072.
[18] M. Bertolini, C.P.J. Glaudemans, Carbohydr. Res. 15 (1970) 263–270.
[19] K. Bock, C. Pedersen, J. Chem. Soc., Perkin Trans. 2 (1974) 293–297.
[20] J. Duus, C.H. Gotfredsen, K. Bock, Chem. Rev. 100 (2000) 4589–4614.
[21] Y. Li, M. Liu, K. Yang, J. Tian, Chin. Herb. Med. 14 (2022) 254–262.
[22] Q.M. Liu, S.S. Xu, L. Li, et al., Carbohydr. Polym. 165 (2017) 189–196.
[23] Z. Liu, Z. Liu, L. Li, et al., Food Sci. Nutr. 10 (2022) 1093–1102.
[24] H. Sun, J. Zhang, F. Chen, et al., Carbohydr. Polym. 121 (2015) 388–402.
[25] Y. Chen, P. Li, Y. Peng, et al., Free Radic. Biol. Med. 172 (2021) 590–603.
[26] N.G. Geum, H.J. Eo, H.J. Kim, et al., J. Funct. Foods 73 (2020) 104139.