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Original article

Design, synthesis and evaluation of potent G-protein coupled receptor 40 agonists

Jing Huang [a,1], Bin Guo [b,1], Wen-Jing Chu [b], Xin Xie [c], Yu-She Yang [b,*], Xian-Li Zhou [a,*]

[a] School of Life Science and Engineering, Southwest Jiao Tong University, Chengdu 610031, China
[b] State Key Laboratory of Drug Research, Shanghai Institute of Materia Medica, Chinese Academy of Sciences, Shanghai 201203, China
[c] CAS Key Laboratory of Receptor Research, The National Center for Drug Screening, Shanghai Institute of Materia Medica, Chinese Academy of Sciences, Shanghai 201203, China

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ABSTRACT

GPR40 has emerged as an attractive drug target for the treatment of type 2 diabetes due to its role in the enhancement of insulin secretion with glucose dependency. With the aim to improve the metabolic and safety profiles, a series of novel phenylpropionic acid derivatives were synthesized. Extensive structural optimization led to identification of compounds **22g** and **23e** as potent GPR40 agonists with moderate liver microsomal stability. All the discovery supported further exploration surrounding this scaffold.

1. Introduction

The prevalence of type 2 diabetes (T2DM) is now a serious global health burden. The total number of people suffering from diabetes is expected to grow from 171 million in 2000 to 366 million by 2030 [1]. Despite some medications are available for treatment of T2DM, current therapy is often associated with weight gain and hypoglycemia (sulfonylureas), also with other adverse effects such as gastrointestinal discomfort or edema [2]. Therefore, there still remains a significant unmet need for new effective, oral anti-diabetic agents that improve glycemic control while maintaining an excellent safety profile.

The G protein-coupled receptor 40 (GPR40, also known as FFA1) primarily expressed in pancreatic β-cells and enteroendocrine cells of the small intestine [3]. When activated by medium to long chain fatty acids, GPR40 elicits enhanced insulin secretion only in the presence of elevated glucose but does not affect insulin secretion at low glucose levels [4,5]. This alluring mechanism to treat type 2 diabetes presents that small molecule agonists of GPR40 may serve as novel insulin secretagogues with little or no risk of hypoglycemia. In recent years, a number of potent GPR40 agonists have been reported and some of them have progressed to clinical trials, exemplified by TAK-875, AMG-837 and LY2881835 (Fig. 1) [6]. Unfortunately, these compounds have been terminated due to safety concerns [6]. By analyzing their structures, we find that there is a common structural moiety of benzyloxy fragment in these compounds. This may cause poor oral pharmacokinetic profiles (PK) and potential safety concern due to benzaldehyde moiety resulted from metabolic oxidation at the benzyl position [7]. Therefore, as an effort to identify novel GPR40 agonists with improved PK and safety profiles, we designed a series of new linkers between the left phenyl (B ring) and phenylpropanoic acid to avoid benzyl oxidation. This paper described the synthesis and biological evaluation of a series of novel phenylpropanoic acid derivatives as potential GPR40 agonists (Fig. 2).

2. Experimental

The synthetic routes of compounds **7** and **13** are outlined in Scheme 1. Condensation of compounds **4a** and **b** with propargyl bromide in the presence of potassium carbonate as a base afforded **5a** and **b**. Compounds **6** and **10** were obtained by Sonogashira cross-coupling reaction of **5a** and **b** and appropriate aromatic bromides [8]. Deprotection of **10** in THF with tetrabutylammonium fluoride and further esterification with triflic anhydride gave **11**. Suzuki-Miyaura cross-coupling of **11** with 3-methoxybenzeneboronic acid provided **12**. Basic hydrolysis of intermediates **6** and **12** afforded the corresponding carboxylic acids **7** and **13**, respectively.

* Corresponding authors.
E-mail addresses: ysyang@simm.ac.cn (Y.-S. Yang), xxbiochem@163.com (X.-L. Zhou).
[1] These authors contributed equally to this work.

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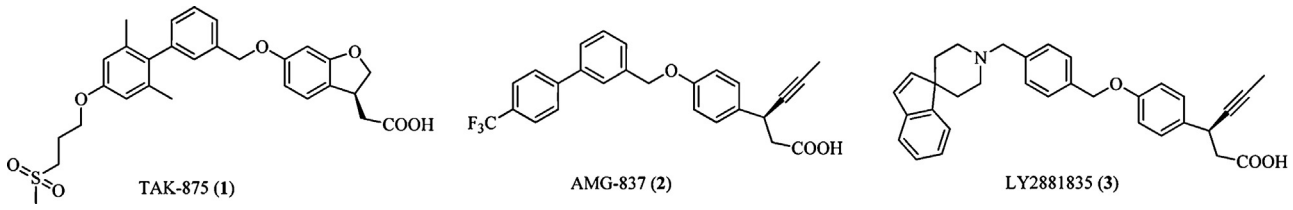


Fig. 1. Structures of representative GPR40 agonists.

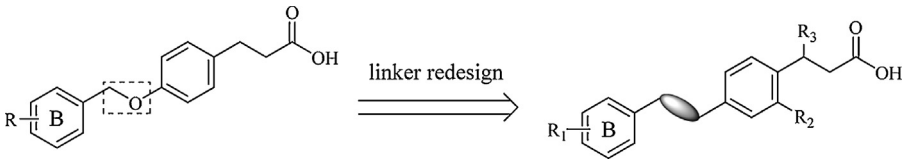
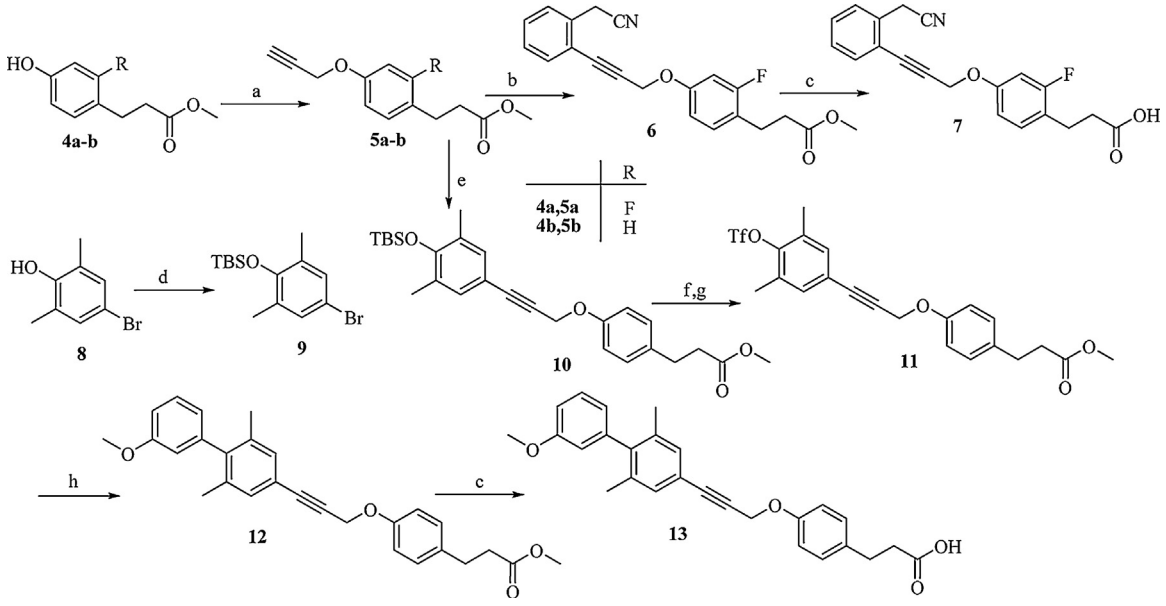


Fig. 2. Design of phenylpropanoic acid derivatives with new linker.

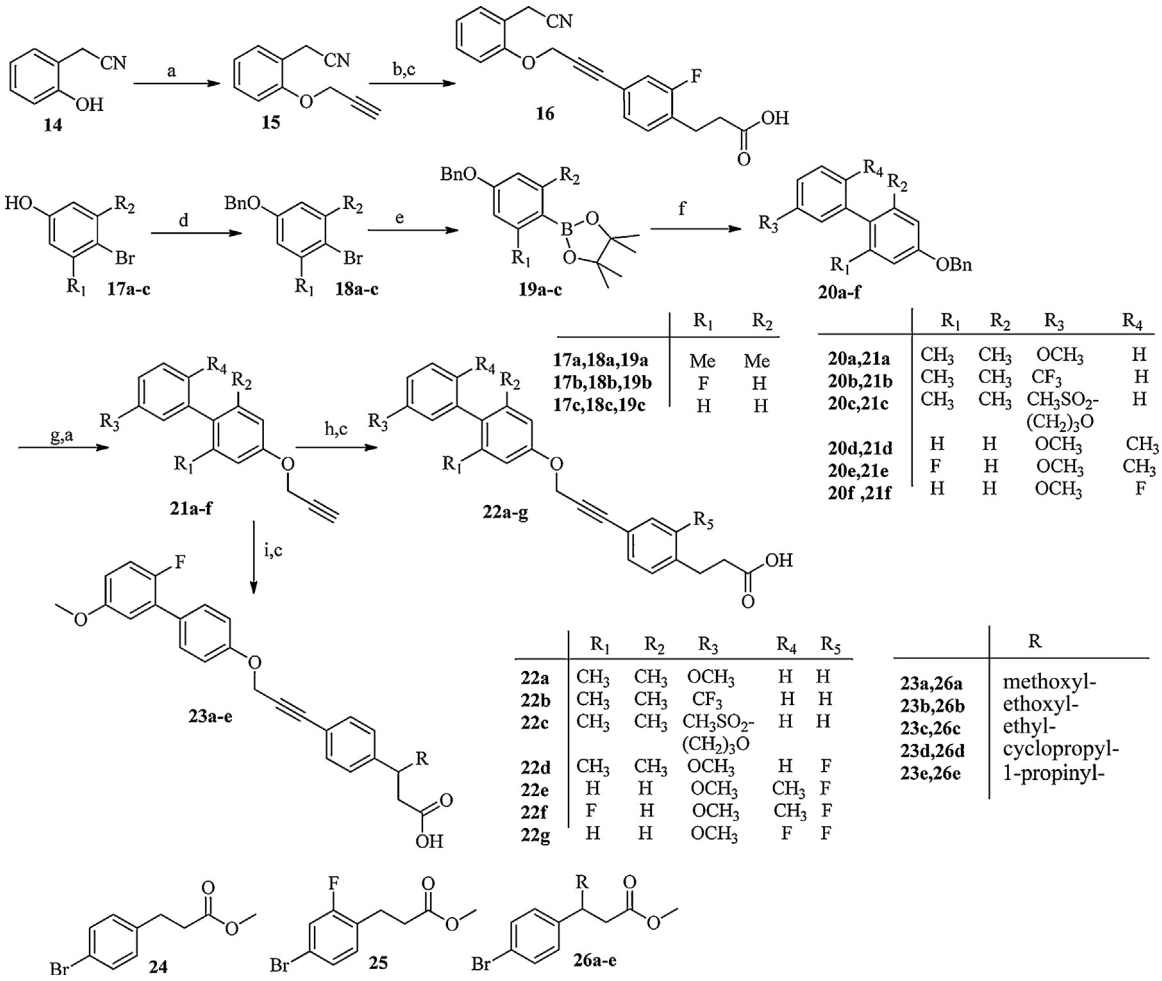


Scheme 1. Synthesis of compounds **7** and **13**. Reagents and conditions: (a) K_2CO_3, DMF, propargyl bromide, 80 °C, 88%–91%; (b) **5a** and 2-bromophenylacetonitrile, Na_2PdCl_4, CuI, H_2O, TMEDA, 2-(di-*tert*-butylphosphino)-1-phenylindole, 80 °C, 62%; (c) 1 mol/L LiOH aq., MeOH, r.t., 82%–87%; (d) TBSCl, TEA, THF, r.t., 92%; (e) **5b** and **9**, Na_2PdCl_4, CuI, H_2O, TMEDA, 2-(Di-*tert*-butylphosphino)-1-phenylindole, 80 °C, 76%; (f) TBAF, THF, r.t., 94%; (g) triflic anhydride; pyridine, DCM, 92%; (h) 3-methoxybenzeneboronic acid, 2 mol/L Na_2CO_3, LiCl, $Pd(PPh_3)_4$, 85 °C, 75%.

Compounds **16**, **22a–g** and **23a–e** were synthesized according to Scheme 2. The preparation of compound **16** began with the conversion of compound **14** to **15**, which was followed by Sonogashira coupling with **25** and basic hydrolysis. Compounds **17a–c** were protected by benzyl bromide and then coupled with pinacolborane to give boronic esters **19a–c**. Suzuki coupling of **19a–c** with appropriate aromatic bromides provided **20a–f**. Compounds **21a–f** were yielded by deprotection of intermediates **20a–f** and then alkylation of the phenols. Sonogashira coupling of **21a–f** with group **24** or **25**, and followed by final basic hydrolysis resulted in **22a–g**. Compounds **23a–e** were synthesized from the intermediate **21f** and **26a–e** [9] according to the synthesis procedures of **22a–g**.

3. Results and discussion

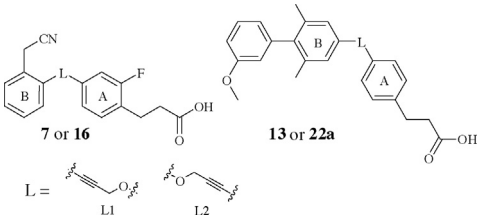
Agonist activities of the synthesized compounds were measured with Calcium flux assay in GPR40-transfected HEK293 cells [10]. As a starting modification effort, we exchanged benzyloxy moiety with propinyloxy group to avoid benzyl oxidation. First we investigated the effect of different connection position of propinyloxy with two phenyl rings (A ring and B ring) on the GRP40 agonistic activity (Table 1). Docosa-hexaenoic acid (DHA), the endogenous ligand for GPR40, was selected as positive control. The results indicated that compounds with linker L2 showed more potent GPR40 agonistic activity than those with linker L1 (compound **7** *vs.* **16**; **13** *vs.* **22a**). Accordingly, we chose compound **22a** as a new lead compound for further chemical optimization and focused our investigation to the substituents on the phenyl ring and β-position to the carboxylic function (Table 2). The CF_3 (**22b**) substituent provided a significant decrease in agonistic activity. When introduced the same tail as seen in compound TAK-875, the derivative (**22c**) showed slightly weaker potency than compound **22a**. So we kept the methoxy group as the favorable substituent at the 5″-position of C ring. Then a fluoro group was introduced into the 2-position of the A ring, which increased the activity significantly (**22d**). We next turned our attention to optimize the biphenyl group. About 2-fold increase in potency was observed



Scheme 2. Synthesis of compounds **16**, **22a–g** and **23a–e**. Reagents and conditions: (a) K_2CO_3, DMF, propargyl bromide, 80 °C, 81%–86%; (b) **25**, Na_2PdCl_4, CuI, 2-(di-tert-butylphosphino)-1-phenylindole, H_2O, TMEDA, 80 °C, 86%; (c) 1 mol/L LiOH aq., MeOH, r.t., 62%–91% (2steps); (d) BnBr, K_2CO_3, DMF, 50 °C, 95%–97%; (e) Potassium acetate, $Pd(dppf)Cl_2$, 1,4-dioxane, bis(pinacolato)diboron, 85 °C, 90%–96%; (f) $Pd(PPh_3)_4$, Cs_2CO_3, appropriate aromatic bromides, 85 °C, 84%–92%; (g) Pd/C, H_2, Etethyl acetate, r.t., 96%–98%; (h) **24** or **25**, Na_2PdCl_4, CuI, 2-(Di-*tert*-butylphosphino)-1-phenylindole, H_2O, TMEDA, 80 °C, 56%–82%; (i) **21f** and **26a–e**, Na_2PdCl_4, CuI, 2-(di-*tert*-butylphosphino)-1-phenylindole, H_2O, TMEDA, 80 °C, 51%–67%.

when the methyl group was moved from 3′-positon of B ring to 2″-position of C ring (**22e**). Unfortunately, the agonistic activity drastically decreased when incorporation another fluoro group in the B ring (**22f**). Replacement of the methyl of **22f** with a fluoro group in the C ring led to 2-fold improvement on potency (**22g**).

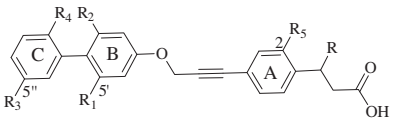
Table 1
GPR40 agonistic activities of compounds **7**, **13**, **16** and **22a**.



Compound	L	EC_{50}[a] (μmol/L)
7	L1	>10
13	L1	>10
16	L2	1.22
22a	L2	2.60
DHA		14.86

[a] Calcium flux assay in GPR40-transfected HEK293 cells. Means of two experiments.

Table 2
GPR40 agonistic activities of compounds **22a–g** and **23a–e**.



Compound	R_1	R_2	R_3	R_4	R_5	R	EC_{50}[a] (μmol/L)
22a	CH_3	CH_3	CH_3O	H	H	H	2.60
22b	CH_3	CH_3	CF_3	H	H	H	10.72
22c	CH_3	CH_3	$CH_3SO_2(CH_2)_3O$	H	H	H	2.64
22d	CH_3	CH_3	CH_3O	H	F	H	0.951
22e	H	H	CH_3O	CH_3	F	H	0.526
22f	F	H	CH_3O	CH_3	F	H	0.913
22g	H	H	CH_3O	F	F	H	0.266
23a	H	H	CH_3O	F	H	methoxy-	>10
23b	H	H	CH_3O	F	H	ethoxy-	0.692
23c	H	H	CH_3O	F	H	ethyl-	>10
23d	H	H	CH_3O	F	H	cyclopropyl-	>10
23e	H	H	CH_3O	F	H	1-propinyl-	0.268
DHA							14.86

[a] Calcium flux assay in GPR40-transfected HEK293 cells. Means of two experiments.

Table 3
Liver microsomal stability of compounds **22g** and **23e**[a].

Compound	Species	Microsomal $t_{1/2}$ [b] (min)	Cl_{int} [c] (μL/min/mg protein)
22g	Human	53.4	39
	Mouse	56.2	38
23e	Human	40.7	52
	Mouse	46.6	45

[a] 0.33 mg/mL microsomal protein, $NADP^+$-regenerating system,[inhibitor], 0.1 μmol/L, incubation at 37 °C, samples taken at 0, 7, 17, 30, and 60 min, determination of parent compound by MS.
[b] $t_{1/2}$: elimination half-life in rat liver microsomes.
[c] Cl_{int}: intrinsic body clearance.

In order to reduce the potential for β-oxidation of the propionic acid head, a series of small residues were introduced into the β-position [10]. As shown in Table 2 (compound **23a**–**e**), the activity on the GPR40 varied significantly with different groups at the β-position. The activity almost disappeared when the methoxy, ethyl, or cyclopropyl groups were introduced (compound **23a, 23c** and **23d**), while the introduction of an ethoxy or alkyne groups were tolerant. The most potent alkyne derivative **23e** displayed comparable agonistic activity with compound **22g**.

To evaluate the metabolic stability, compounds **22g** and **23e** were subjected to the human and mouse liver microsomal stability assays. As shown in Table 3, both compounds displayed moderate metabolic stability with an acceptable clearance and half-life.

4. Conclusion

In conclusion, to improve the pharmacokinetic and safety profiles of the reported benzyloxy-like GPR40 agonists, a series of novel phenylpropionic acid analogs were designed and synthesized with redesign linker between the B ring and phenylpropionic acid moiety. Their GPR40 agonistic activities were then evaluated in HEK293 cells stably expressing human GPR40. The results showed that oxypropinyl (L2) was a preferred linker. Around this new structure, comprehensive chemical modification was carried on. By focusing investigation to the substituents on the phenyl ring and β-position of propionic acid, compounds **22g** and **23e** were identified as the most potent GPR40 agonists with EC_{50} 0.266 μmol/L and 0.268 μmol/L, respectively. Additionally, both compounds showed moderate metabolic stability in liver microsomal stability assay. All these finding support further exploration based on this scaffold and the optimization results will be reported in due course.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.cclet.2015.09.002.

References

[1] S. Wild, G. Roglic, A. Green, R. Sicree, H. King, Global prevalence of diabetes: estimates for the year 2000 and projections for 2030, Diabetes Care 27 (2004) 1047–1053.
[2] B. Ahrén, Islet G protein-coupled receptors as potential targets for treatment of type 2 diabetes, Nat. Rev. Drug Discov. 8 (2009) 369–385.
[3] S. Edfalk, P. Steneberg, H. Edlund, *Gpr40* is expressed in enteroendocrine cells and mediates free fatty acid stimulation of incretin secretion, Diabetes 57 (2008) 2280–2287.
[4] A.D. Mancini, V. Poitout, The fatty acid receptor FFA1/GPR40 a decade later: how much do we know? Trends Endocrinol. Metab. 24 (2013) 398–407.
[5] N.G. Morgan, S. Dhayal, G-protein coupled receptors mediating long chain fatty acid signalling in the pancreatic beta-cell, Biochem. Pharmacol. 78 (2009) 1419–1427.
[6] E. Defossa, M. Wagner, Recent developments in the discovery of FFA1 receptor agonists as novel oral treatment for type 2 diabetes mellitus, Bioorg. Med. Chem. Lett. 24 (2014) 2991–3000.
[7] R. Takano, M. Yoshida, M. Inoue, et al., Discovery of DS-1558: a potent and orally bioavailable GPR40 agonist, ACS Med. Chem. Lett. 6 (2015) 266–270.
[8] E. Christiansen, M.E. Due-Hansen, C. Urban, et al., Discovery of a potent and selective free fatty acid receptor 1 agonist with low lipophilicity and high oral bioavailability, J. Med. Chem. 56 (2013) 982–992.
[9] J.B. Houze, L.S. Zhu, Y. Sun, et al., AMG 837: a potent, orally bioavailable GPR40 agonist, Bioorg. Med. Chem. Lett. 22 (2012) 1267–1270.
[10] N. Negoro, S. Sasaki, M. Ito, et al., Identification of fused-ring alkanoic acids with improved pharmacokinetic profiles that act as G protein-coupled receptor 40/free fatty acid receptor 1 agonists, J. Med. Chem. 55 (2012) 1538–1552.