

Electronical supplementary information (ESI)

**Design, screening and biological evaluation of novel oxyntomodulin-based hybrid peptides
with prolonged glucose-lowering ability and potent anti-obesity effects**

Lei Zhao ^a, Baohua Wang ^a, Limin Wang^a, Xie Zhao^b, Zhe Chen^c, Lixia Sun^a

^aNorth China University of Science and Technology Affiliated Hospital, Tangshan, Hebei 063000,
P.R. China.

^bPeking University, College of Pharmaceutical Sciences, Beijing, 100000, P.R.

^cNorth China University of Science and Technology, Tangshan, Hebei 063000, P.R. China.

Correspondence: lxsun@ncstmc.cn; Tel.: +86-315-3725120 (L.S.)

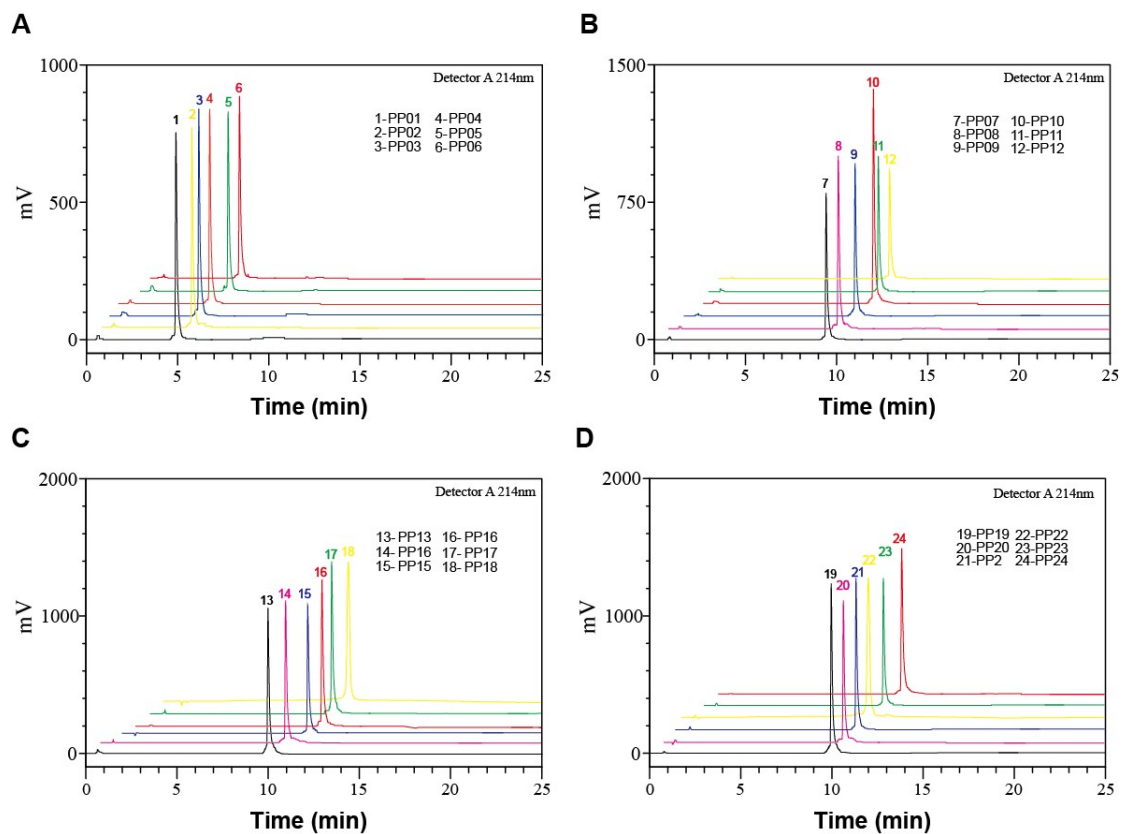


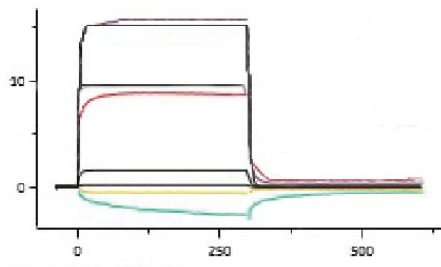
Figure S1. RP-HPLC chromatogram of PP01 to PP24. (A) PP01-PP06; (B) PP07-PP12; (C) PP13-PP18; (D) PP19-PP24.

Table S1. RP-HPLC analysis of the synthesized peptides

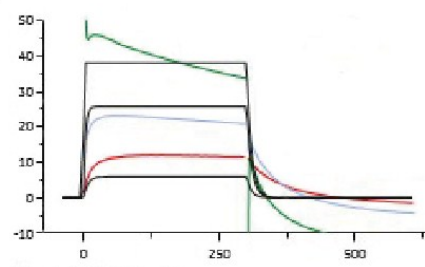
Peptide	HPLC analysis		Peptide	HPLC analysis	
	Purity (%)	Retention time (min)		Purity (%)	Retention time (min)
PP01	98.5	4.95	PP13	97.4	10.14
PP02	98.1	5.11	PP14	98.3	10.16
PP03	98.2	5.05	PP15	97.8	10.22
PP04	97.2	4.96	PP16	98.1	10.16
PP05	98.9	5.04	PP17	98.3	10.32
PP06	98.1	5.12	PP18	97.2	10.01
PP07	98.5	9.33	PP19	97.8	10.17
PP08	98.1	9.51	PP20	97.7	10.25
PP09	98.5	9.42	PP21	97.8	10.01
PP10	97.9	9.37	PP22	98.4	10.05
PP11	98.4	9.45	PP23	97.8	10.11
PP12	97.9	9.61	PP24	97.9	10.29

Table S2. MS analysis of the synthesized peptides

Peptide	Molecular mass		Peptide	Molecular mass	
	Calculated	Found		Calculated	Found
PP01	246.28[M+5H]5+	246.28[M+5H]5+	PP13	879.76[M+7H]7+	879.69[M+7H]7+
PP02	230.29[M+5H]5+	230.14[M+5H]5+	PP14	887.91[M+7H]7+	887.79[M+7H]7+
PP03	249.49[M+5H]5+	249.54[M+5H]5+	PP15	892.71[M+7H]7+	892.81[M+7H]7+
PP04	215.77[M+5H]5+	215.69[M+5H]5+	PP16	896.06[M+7H]7+	896.06[M+7H]7+
PP05	237.56[M+5H]5+	237.45[M+5H]5+	PP17	904.19[M+7H]7+	904.28[M+7H]7+
PP06	254.47[M+5H]5+	254.38[M+5H]5+	PP18	908.96[M+7H]7+	908.77[M+7H]7+
PP07	793.36[M+7H]7+	793.31[M+7H]7+	PP19	896.06[M+7H]7+	896.12[M+7H]7+
PP08	781.91[M+7H]7+	781.85[M+7H]7+	PP20	904.19[M+7H]7+	904.19[M+7H]7+
PP09	795.61[M+7H]7+	795.51[M+7H]7+	PP21	908.96[M+7H]7+	908.81[M+7H]7+
PP10	771.66[M+7H]7+	771.56[M+7H]7+	PP22	896.06[M+7H]7+	896.18[M+7H]7+
PP11	787.16[M+7H]7+	787.08[M+7H]7+	PP23	904.19[M+7H]7+	904.11[M+7H]7+
PP12	799.19 [M+7H]7+	799.26 [M+7H]7+	PP24	908.96[M+7H]7+	908.81[M+7H]7+

A

Model 1:1 binding
ka(1/Ms) 1.97e+05 **kd(1/s)** 1.12e-01
Rmax(RU) 8908.2 **KD (M)** 5.66e-07

B

Model 1:1 binding
ka(1/Ms) 8.68e+05 **kd(1/s)** 3.15e-01
Rmax(RU) 40.2 **KD (M)** 3.63e-07

Figure S2. SPR measurements. SPR chromatogram of PP18 interaction with GLP-1R ECD (A) and GcgR ECD (B).

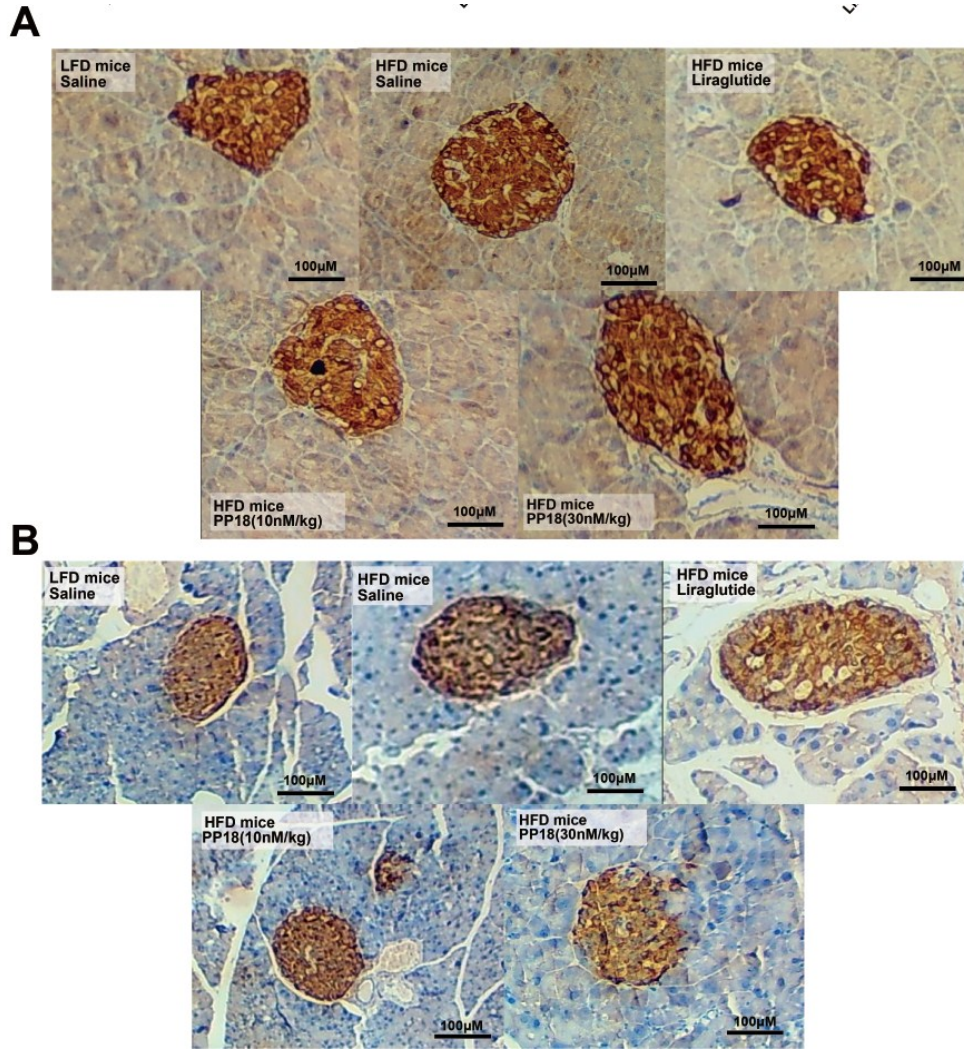


Figure S3. Effects of 4 weeks of treatment with PP18 on the beta cell proliferation (A) and beta cell apoptosis (B).