

Supporting Information

The Melanocortin Tetrapeptide Ac-His-DPhe-Arg-Trp-
NH₂ Modified at the Para Position of the Benzyl Side
Chain (DPhe): Importance for Mouse Melanocortin-3
Receptor Agonist Versus Antagonist Activity

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Table Supplemental Data. Analytical Data for Peptides Synthesized in this Study.

Peptide	Structure	HPLC k' ¹ (system 1)	HPLC k' ² (system 2)	M+1 (calculated)	Mass spectral analysis (M+1)	Purity (%) ³
5	Ac-His-DPhe ⁷ -Arg-Trp-NH ₂	4.1	6.7	686.8	686.5	>97
6	Ac-His-DTyr-Arg-Trp-NH ₂ Tyr	3.6	5.4	702.8	702.3	>97
7	Ac-His-DPhe(pNH ₂)-Arg-Trp-NH ₂	2.1	5.0	701.8	701.5	>97
8	Ac-His-DPhe(pCH ₃)-Arg-Trp-NH ₂	4.6	8.0	700.8	700.5	>97
9	Ac-His-DPhe(pt-Bu)-Arg-Trp-NH ₂	6.1	10.4	742.9	742.5	>96
10	Ac-His-DPhe(pBz)-Arg-Trp-NH ₂	4.5	9.5	776.9	777.1	>97
11	Ac-His-DPhe(pCN)-Arg-Trp-NH ₂	4.2	6.9	711.8	711.3	>97
12	Ac-His-DPhe(pNO ₂)-Arg-Trp-NH ₂	4.8	7.3	731.8	731.5	>98
13	Ac-His-DPhe(pF)-Arg-Trp-NH ₂	5.5	7.3	704.8	704.6	>98
14	Ac-His-DPhe(pCl)-Arg-Trp-NH ₂	4.4	8.2	721.2	720.4	>97
15	Ac-His-DPhe(pBr)-Arg-Trp-NH ₂	4.9	8.5	765.7	766.4	>99
16	Ac-His-DPhe(pI)-Arg-Trp-NH ₂	5.1	8.9	812.7	812.5	>98
17	Ac-His-DPhe(pCF ₃)-Arg-Trp-NH ₂	5.0	9.1	754.3	754.7	>98
19	Ac-His-DPhe(3,4-diCl)-Arg-Trp-NH ₂	5.2	9.2	755.6	754.3	>99
20	Ac-His- DPhe(m-Cl)-Arg-Trp-NH ₂	4.5	8.3	721.2	720.3	>99

The HPLC k' value equals [(peptide retention time – solvent retention time)/ solvent retention time]. Two different solvent systems were used. ¹Solvent system 1 equals (10% acetonitrile in 0.1% trifluoroacetic acid/H₂O and a gradient to 90% acetonitrile over 35 min). ²Solvent system 2 equals (10% methanol in 0.1% trifluoroacetic acid/ H₂O and a gradient to 90% methanol over 35 min). The analytical Vydac C18 column (Vydac 218TP104) in combination with a flow rate of 1.5 ml/min was used for analytical characterization. ³The peptide purity was determined by HPLC at a wavelength of 214 nm.