## ChemMedChem

**Supporting Information** 

## Evaluation of 4-(4-Fluorobenzyl)piperazin-1-yl]-Based Compounds as Competitive Tyrosinase Inhibitors Endowed with Antimelanogenic Effects

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<sup>1</sup>H-NMR and selected representative <sup>13</sup>C-NMR spectra



Figure S2: <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-phenylethan-1-one (7)



Figure S3: <sup>13</sup>C-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-phenylethan-1-one (7)



Figure S4: <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-3-phenylpropan-1-one (8)



**Figure S5:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-3-phenylpropan-1-one (8)



**Figure S6:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2,2-diphenylethan-1-one (9)



**Figure S7:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2,2-diphenylethan-1-one (9)



**Figure S8:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(2-chlorophenyl)ethan-1-one (**10**)



**Figure S9:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(2-chlorophenyl)ethan-1-one (**10**)



**Figure S10:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(3-chlorophenyl)ethan-1-one (**11**)



**Figure S11:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(3-chlorophenyl)ethan-1-one (**11**)



**Figure S12**: <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(4-chlorophenyl)ethan-1-one (**12**)



**Figure S13:** <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(2-bromophenyl)ethan-1-one (**13**)



**Figure S14:** <sup>13</sup>C-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(2-bromophenyl)ethan-1-one (**13**)



**Figure S15:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(3-bromophenyl)ethan-1-one (**14**)



**Figure S16:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(3-bromophenyl)ethan-1-one (**14**)



**Figure S17**: <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(4-bromophenyl)ethan-1-one (**15**)



**Figure S18:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(2-nitrophenyl)ethan-1-one (**16**)



**Figure S19:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(2-nitrophenyl)ethan-1-one (**16**)



**Figure S20**: <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(3-nitrophenyl)ethan-1-one (**17**)



**Figure S21:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(4-nitrophenyl)ethan-1-one (**18**)



**Figure S22:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(4-nitrophenyl)ethan-1-one (**18**)



**Figure S23**: <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(4-methoxyphenyl)ethan-1-one (**19**)



**Figure S24:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(2-aminophenyl)ethan-1-one (**20**)



**Figure S25:**<sup>13</sup>C-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(2-aminophenyl)ethan-1-one (**20**)



**Figure S26**: <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(3-aminophenyl)ethan-1-one (**21**)



**Figure S27:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(4-aminophenyl)ethan-1-one (**22**)



**Figure S28:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-2-(4-aminophenyl)ethan-1-one (**22**)



**Figure S29:** <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of [4-(4-Fluorobenzyl)piperazin-1-yl]-[3-fluoro-2-(trifluoromethyl)phenyl]methanone (**23**)



**Figure S30:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of [4-(4-Fluorobenzyl)piperazin-1-yl]-[3-fluoro-2-(trifluoromethyl)phenyl]methanone (**23**)



**Figure S31**: <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-[4-fluoro-2-(trifluoromethyl)phenyl]methanone (**24**)



**Figure S32**: <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-[5-fluoro-2-(trifluoromethyl)phenyl]methanone (**25**)



**Figure S33**: <sup>1</sup>H-NMR (DMSO-d6) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-(3-chloro-2-nitro-phenyl)methanone (**26**)



**Figure S34:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of [4-(4-Fluorobenzyl)piperazin-1-yl]-(4-chloro-2-nitro-phenyl)methanone (**27**)



**Figure S35:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of [4-(4-Fluorobenzyl)piperazin-1-yl]-(4-chloro-2-nitro-phenyl)methanone (**27**)



**Figure S36**: <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-(5-chloro-2-nitro-phenyl)methanone (**28**)



**Figure S37:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of [4-(4-Fluorobenzyl)piperazin-1-yl]-(2-chloro-6-nitro-phenyl)methanone (**29**)



**Figure S38:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of [4-(4-Fluorobenzyl)piperazin-1-yl]-(2-chloro-6-nitro-phenyl)methanone (**29**)



**Figure S39**: <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-(3-methoxy-2-nitrophenyl)methanone (**30**)



**Figure S40:** <sup>1</sup>H-NMR (DMSO-d6) spectrum of [4-(4-Fluorobenzyl)piperazin-1-yl]-(4-methoxy-2-nitrophenyl)methanone (**31**)



**Figure S41:** <sup>13</sup>C-NMR (DMSO-d6) spectrum of [4-(4-Fluorobenzyl)piperazin-1-yl]-(4-methoxy-2-nitrophenyl)methanone (**31**)



**Figure S42**: <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of 1-[4-(4-Fluorobenzyl)piperazin-1-yl]-(5-methoxy-2-nitrophenyl)methanone (**32**)



**Figure S43.** Representation of AbTYR binding site. A) AbTYR surface. The binding site defined for the docking calculation is lined by a black square. B) Close view of AbTYR binding site. The residues of the pocket are represented as wheat sticks.



**Figure S44.** Representation of hTYR binding site. A) hTYR surface. The binding site defined for the docking calculation is lined by a black square. B) Close view of hTYR binding site. The residues of the pocket are represented as lightblue sticks.

Target MLLAVLYCLLWSFQTSAGHEPRACVSSKNLMEKECCPPWSGDRSPCGQLSGRGSCQNILLSNAPLGPQFPFTGVDD	76
5m8q.1.A	62
Target RESWPSVFYNRTCQCSGNFMGFNCGNCKFGFWGPNCTERRLLVRRNIFDLSAPEKDKFFAYLTLAKHTISSDYVIPIGTY	156
5m8q.1.A REVWPLRFFNRTCHOMENCGOOLGGIGGGACTOVIVERKIDLSKEKNHFVRALMAKBTHELEVI	142
Target GOM KNGSTPMFNDINIYDLFVWMHYYVSMDALLGGS - EIWRDIDFAHEAPAFLPWHRLFLLRWEQEIQKLTGDENFTI	233
5m8q.1.A DILGF GNTP FIL YN FVWHYYSVKF LGVG ES SEVDFSHEFAFLWHR HLLR EKD OF LGFSFSL	222
Target PYWDWRDA-EKCDICTDEYMGGQHPTNPNLLSPASFFSSWQIVCSRLEEYNSHQSLCNGTPEGPLRRNPGNH-DKSRTPR	311
5m8q.1.APWN AT GKNVCDICDD COG DC C C C C C C C C C C C C C C C C C C	302
Target LPSSADVEFCLSLTQYESGSMDKAANFSFRNTLEGFASPLTGIADASQSSMHNALHIYMNGTMSQVQGSANDPIFLLHHA	391
5m8q.1.ALP CDVACLEVCLED TFFYSSISTSFRNTVEGF PTC-MCDPAVSS HN AH DINGTGQVHISPNDPIFVLHT	381
Target FVDSIFEQWLRRHRPLQEVMPEANAPIGHNRESYMVPFIPLYRNGDFFISS-KDLGYDYSYLQDSDPDSFQDYIKSYLEQ	470
5m8q.1.AFTDAVEDEWLRBYNALISTEPLENAPIGHNRGYMVPFWPENDEVQNTEDFV GPDNDGYTY	444
Target ASRIWSWLLGAAMVGAVLTALLAGLVSLLCRHKRKQLPEEKQPLLMEKEDYHSLYQSHL 5m8q.1.A	529

Figure S45: Sequence alignment of hTYR (target) and (TYRP-1) mutant (T391V-R374S-Y362F) 5m8q.



**Figure S46:** B16F10 melanoma cell viability after treatment with compounds **23**, **25** and **26**. The mean value and standard deviation were calculated from triplicate experiments.

COMPOUND	CAS Number	SMILES				
7.	423743-29-7	Fc1ccc(CN2CCN(C(Cc3ccccc3)=O)CC2)cc1				
8.	439848-20-1	Fc1ccc(CN2CCN(CC2)C(=O)CCc3ccccc3)cc1				
9.	423739-67-7	Fc1ccc(CN2CCN(CC2)C(=O)C(c3ccccc3)c4ccccc4)cc1				
10.	1387739-82-3	Fc1ccc(CN2CCN(CC2)C(=O)Cc3ccccc3Cl)cc1				
11.	-	Fc1ccc(CN2CCN(CC2)C(=O)Cc3cccc(Cl)c3)cc1				
12.	1329294-83-8	Fc1ccc(CN2CCN(CC2)C(=O)Cc3ccc(Cl)cc3)cc1				
13.	1988172-38-8	Fc1ccc(CN2CCN(CC2)C(=O)Cc3ccccc3Br)cc1				
14.	1985981-75-6	Fc1ccc(CN2CCN(CC2)C(=O)Cc3cccc(Br)c3)cc1				
15.	1146917-07-8	Fc1ccc(CN2CCN(CC2)C(=O)Cc3ccc(Br)cc3)cc1				
16.	-	[O-][N+](=O)c1ccccc1CC(=O)N2CCN(Cc3ccc(F)cc3)CC2				
17.	-	[O-][N+](=O)c1cccc(CC(=O)N2CCN(Cc3ccc(F)cc3)CC2)c1				
18.	-	[O-][N+](=O)c1ccc(CC(=O)N2CCN(Cc3ccc(F)cc3)CC2)cc1				
19.	1796840-99-7	COc1ccc(CC(=O)N2CCN(Cc3ccc(F)cc3)CC2)cc1				
20.	-	Nc1ccccc1CC(=O)N2CCN(Cc3ccc(F)cc3)CC2				
21.	-	Nc1cccc(CC(=O)N2CCN(Cc3ccc(F)cc3)CC2)c1				
22.	-	Nc1ccc(CC(=O)N2CCN(Cc3ccc(F)cc3)CC2)cc1				
23.	-	Fc1ccc(CN2CCN(CC2)C(=O)c3cccc(F)c3C(F)(F)F)cc1				
24.	-	Fc1ccc(CN2CCN(CC2)C(=O)c3ccc(F)cc3C(F)(F)F)cc1				
25.	-	Fc1ccc(CN2CCN(CC2)C(=O)c3cc(F)ccc3C(F)(F)F)cc1				
26.	-	[O-][N+](=O)c1c(Cl)cccc1C(=O)N2CCN(Cc3ccc(F)cc3)CC2				
27.	2344352-87-8	[O-][N+](=O)c1cc(Cl)ccc1C(=O)N2CCN(Cc3ccc(F)cc3)CC2				
28.	-	[O-][N+](=O)c1ccc(Cl)cc1C(=O)N2CCN(Cc3ccc(F)cc3)CC2				
29.	-	[O-][N+](=O)c1cccc(Cl)c1C(=O)N2CCN(Cc3ccc(F)cc3)CC2				
30.	-	COc1cccc(C(=O)N2CCN(Cc3ccc(F)cc3)CC2)c1[N+](=O)[O-]				
31.	-	COc1ccc(C(=O)N2CCN(Cc3ccc(F)cc3)CC2)c(c1)[N+](=O)[O-]				
32.	2345032-69-9	COc1ccc(c(c1)C(=O)N2CCN(Cc3ccc(F)cc3)CC2)[N+](=O)[O-]				

Table S1: CAS numbers and smiles for compounds 7-32

TPSA\* TPSA\*\* iLogP\* PAINS\* MW (g/mol)\* miLogP\*\* Water solubility\* Lipinski\* Ghose\* entry 7 312.38 23.55 23.55 3.21 2.92 Soluble Yes Yes 0 alert 0 violation 326.41 23.55 23.55 3.54 3.44 Soluble Yes 8 Yes 0 alert 0 violation 9 388.48 23.55 23.55 3.79 4.31 Moderately soluble Yes Yes 0 alert 0 violation 23.55 23.55 3.44 Soluble Yes 0 alert 10 346.83 3.55 Yes 0 violation 11 346.83 23.55 23.55 3.55 3.58 Soluble Yes Yes 0 alert 0 violation 12 3.52 346.83 23.55 23.55 3.60 Soluble Yes Yes 0 alert 0 violation 13 23.55 23.55 Soluble 0 alert 391.28 3.51 3.68 Yes Yes 0 violation 391.28 23.55 23.55 3.64 3.71 Soluble Yes 14 Yes 0 alert 0 violation 15 23.55 391.28 23.55 3.63 3.73 Soluble Yes Yes 0 alert 0 violation 69.37 69.37 2.83 16 357.38 2.83 Soluble Yes 0 alert Yes 0 violation 17 357.38 69.37 2.95 Soluble Yes 69.37 2.86 Yes 0 alert 0 violation 18 357.38 69.37 69.37 2.92 2.88 Soluble Yes Yes 0 alert 0 violation 19 342.41 32.78 32.78 3.46 0 alert 2.98 Soluble Yes Yes 0 violation 327.40 49.57 49.57 2.92 Soluble Yes 0 alert 20 2.36 Yes 0 violation 49.57 1.97 21 327.40 49.57 2.88 Soluble Yes Yes 0 alert 0 violation 22 327.40 49.57 49.57 2.78 2.00 Soluble Yes Yes 1 alert: 0 violation anil\_no\_alk 23 384.34 23.55 23.55 3.27 3.32 Soluble Yes Yes 0 alert 1 violation: MLogP>4.15 24 384.34 23.55 23.55 3.36 3.35 Soluble Yes 0 alert Yes 1 violation: MLogP>4.15 25 384.34 23.55 23.55 3.34 3.35 Soluble Yes Yes 0 alert 1 violation: MLogP>4.15

**Table S2.** Selected physicochemical parameters, lipophilicity, solubility and drug-likeness for compound **7-32** predicted by SwissADME(http://swissadme.ch/), and Molinspiration tools(https://molinspiration.com).

26	377.80	69.37	69.37	2.84	2.90	Moderately soluble	Yes	Yes	0 alert
							0 violation		
27	377.80	69.37	69.37	2.89	2.92	Moderately soluble	Yes	Yes	0 alert
							0 violation		
28	377.80	69.37	69.37	2.89	2.92	Moderately soluble	Yes	Yes	0 alert
							0 violation		
29	377.80	69.37	69.37	2.76	2.90	Moderately soluble	Yes	Yes	0 alert
							0 violation		
30	373.38	78.60	78.61	2.66	2.28	Soluble	Yes	Yes	0 alert
							0 violation		
31	373.38	78.60	78.61	2.65	2.30	Soluble	Yes	Yes	0 alert
							0 violation		
32	373.38	78.60	78.61	2.72	2.30	Soluble	Yes	Yes	0 alert
							0 violation		

\*SwissADME Water solubility: Soluble= Log S (Ali) values between -4 and -2; Moderately soluble= Log S (Ali) values between -6 and -4.

\*\*Molinspiration