Fluorination Shifts the Profile of an Endogenous Neurotransmitter

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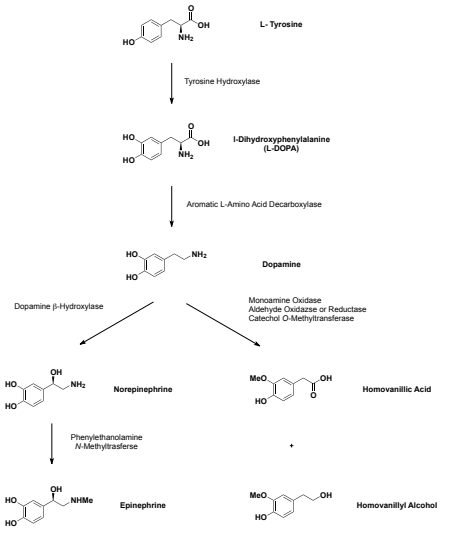
Introduction and Hypothesis

A survey of novel endogenous neurotransmitters revealed an opportunity to explore previously unknown chemical matter. Several fluorination patterns within the catechols were unprecedented in the literature. Furthermore, the inclusion of fluorine might impart interesting biological activity and desirable properties. A summary of the biosynthesis and metabolism of dopaminergic catechols is depicted below.

Catechol Biosynthesis and Metabolism



Initial Target Selection

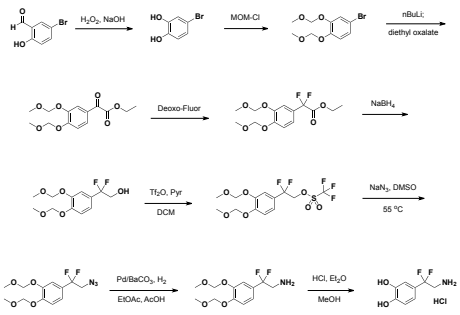
Our original target (SF097) was a dopamine-like catecholamine with geminal difluorination in the benzylic position. This substitution should have an interesting and hopefully useful effect upon the Lewis basicity and acidity of both the amine and catechol functional groups.

Dopamine SF097

The fluorination of the target molecule was accomplished via the treatment of a ketone with Deoxo-Fluor. The transformations utilized to synthesize SF097 were generally straightforward and high yielding; however, the yield of the alpha-ketoester was modest. The final compound is stable both as a solid and in DMSO.

The effect of geminal difluorination was evaluated with a panel of 38 CNS targets. Activities noted by these radioligand displacement assays were further evaluated in functional assays.

Synthetic Chemistry



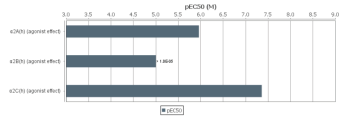
Neurological Targets Screen

Classification	Assay	Radioligand	Result (% control at 10 μM)
Neurotransmitter Related	Adenosine, Non-selective	[3H]NECA	16%
	Adrenergic, Alpha 1, Non-selective	[3H]7-MeOxy-Prazosin	<10%
	Adrenergic, Alpha 2, Non-selective	[3H]RX821002	92%
	Adrenergic, Beta, Non-selective	[3H]DHA	<10%
	Dopamine Transporter	[3H]WIN 35,428	51%
	Dopamine, D1 (h)	[3H]-SCH23390	31%
	Dopamine, D2s (h)	[3H]-Raclopride	42%
	GABA A, Agonist Site	[3H]GABA	<10%
	GABA A, BDZ, alpha 1 site	[3H]Flunitrazepam	19%
	GABA-B	[3H]CGP 54626A	45%
	Glutamate, AMPA Site (Ionotropic)	[3H]AMPA	<10%
	Glutamate, Kainate Site (Ionotropic)	[3H]Kainic acid	<10%
	Glutamate, NMDA Agonist Site (Ionotropic)	[3H]CGP 39653	<10%
	Glutamate,NMDA,Glycine (Stry-insens Site) (Ionot	[3H]-MDL-105,519	<10%
	Glycine, Strychnine-sensitive	[3H]Strychnine	<10%
	Histamine, H1	[3H]Pyrilamine	<10%
	Histamine, H2	[125I]-Aminopotentidine	19%
	Melatonin, Non-selective	[125I]-2-Iodomelatonin	<10%
	Muscarinic, Non-selective, Central	[3H]QNB	<10%
	Nicotinic, Neuronal (a-BnTx insensitive)	[3H]Epibatidine	<10%
	Norepinephrine Transporter	[3H]Nisoxetine	50%
	Opioid, Non-selective	[3H]Naloxone	22%
	Opioid, Orphanin, ORL1 (h)	[3H] Nociceptin	<10%
	Serotonin Transporter	[3H]Citalopram, N-Methyl	13%
	Serotonin, Non-selective	[3H] LSD	<10%
	Sigma, Non-selective	[3H]DTG	<10%
Ion Channels	Potassium Channel, ATP-Sensitive	[3H]Glibenclamide	16%
	Potassium Channel, Ca2+ Act., VI	[125I]Apamin	<10%
	Sodium, Site 2	[3H]Batrachotoxin A 20-a-Benzo	<10%
Second Messengers	Nitric Oxide, NOS (Neuronal-Binding)	[3H]NOARG	<10%
Growth Factors/Hormones	Corticotropin Releasing Factor, Non-selective	[125I]Tyr0-oCRF	<10%
Brain/Gut Peptides	Angiotensin II, AT2	[125I]Tyr4-Angiotensin II	<10%
	Cholecystokinin, CCK2 (CCKB)	[125I]CCK-8	10%
	Endothelin, ET-B (h)	[125I]-Endothelin-1	<10%
Enzymes	Decarboxylase, Glutamic Acid	[14C]Glutamic acid	<10%
	Esterase, Acetylcholine	Acetylthiocholine	<10%
	Oxidase, MAO-A-Peripheral	[14C] - 5HT	23%
	Oxidase, MAO-B, Peripheral	[14C]Phenylethylamine	<10%

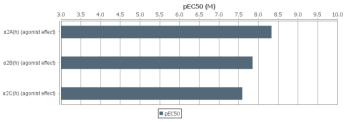
Selective for Adrenergic Alpha 2c

Following up on the CNS panel, our lead was titrated in three functional adrenergic alpha 2 assays head-to-head with norepinephrine. The compound is a selective agonist of alpha2c (2a, 1100nM; 2b, >10000 nM; 2c, 44 nM) and is a full agonist of alpha 2c at the higher doses. The contrasting data for norepinephrine is depicted below.

SF097



Norepinephrine



Counterscreens, Dopamine Functional and COMT

Dopamine itself has varied and often not apparently potent activity in radioligand displacement assays. Titrations in functional assays performed in CHO cells were utilized to confirm a lack of activity in agonist and antagonist modes (reporters adenylyl cyclase for D1, and GTPγS binding for the rest).

Functional Dopamine Assays

	Agonist Mode (10 μM)	Antagonist Mode (10 μM)
Dopamine D_1	< 10%	< 10%
Dopamine D_{2L}	< 10%	< 10%
Dopamine D_{2S}	< 10%	< 10%
Dopamine D_3	< 10%	-
Dopamine $D_{4.2}$	< 10%	< 10%
Dopamine $D_{4.4}$	< 10%	-

Catechol O-Methyl Transferase

	Enzyme Assay
Catechol 0-Methyl Transferase	No inhibition (<5% at 10 μM)

Conclusions

We have developed a compound with a unique and selective profile that could be useful in exploring the pharmacological role of adrenergic alpha2c receptors.

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