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Appetite Suppressants, Cardiac Valve Disease and Combination Pharmacotherapy

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Abstract

The prevalence of obesity in the United States is a major health problem associated with significant morbidity, mortality and economic burden. Although obesity and drug addiction are typically considered distinct clinical entities, both diseases involve dysregulation of biogenic amine neuron systems in the brain. Thus, research efforts to develop medications for treating drug addiction can contribute insights into the pharmacotherapy for obesity. Here we review the neurochemical mechanisms of selected stimulant medications used in the treatment of obesity, as well as issues related to fenfluramine-associated cardiac valvulopathy. In particular, we discuss the evidence that cardiac valve disease involves activation of mitogenic 5-HT_{2B} receptors by norfenfluramine, the major metabolite of fenfluramine. Advances in medication discovery suggest that novel molecular entities that target two different neurochemical mechanisms, i.e. “combination pharmacotherapy”, will yield efficacious anti-obesity medications with reduced adverse side-effects.

Introduction

Epidemiological data reveal that the prevalence of obesity is increasing in the United States¹, and obesity is associated with considerable morbidity, mortality and costs to the health care system². The causes of excessive weight gain are complex and involve the interplay between biological, genetic, and psychosocial factors. As aptly stated by Bray and Champagne³, “obesity is a chronic, relapsing, stigmatized, neurochemical disease.” Diet and exercise are primary treatment approaches to reduce body weight, but for many patients the results are disappointing and patients often regain lost weight⁴. Indeed, obese individuals often cycle (“yo-yo”) through periods of dieting and weight loss, followed by weight regain^{5, 6}. Pharmacotherapy can be a crucial component of a weight loss program. A recent study reported that pharmacotherapy combined with lifestyle modification is more effective than lifestyle modification alone⁷.

Numerous publications provide excellent reviews on the medications currently in use or under investigation for the treatment of obesity^{8, 9}. Because obesity is a chronic relapsing disorder, many clinicians believe that long-term treatment with anorectic medications is appropriate to help patients maintain lower body weight^{10, 11}. Within the central nervous system (CNS), multiple neuronal mechanisms are involved in the regulation of appetite, food intake and maintenance of body weight¹². Perhaps not surprisingly, medications that target one neurochemical mechanism produce relatively small reductions in body weight (decrease of 5%-10% body weight). Weight loss of this magnitude can diminish the risk factors associated

with cardiovascular morbidity and mortality¹³⁻¹⁵, though many patients expect more substantial, cosmetically meaningful, reductions in weight (decrease of 20-25% body weight)¹⁶. The administration of two medications that work via different neurochemical mechanisms, i.e. “combination pharmacotherapy”, is one approach to obtaining cosmetically relevant reductions in weight. The most effective example of this approach was the combination of phentermine and fenfluramine¹⁷⁻²¹. Phentermine targets norepinephrine (NE) and dopamine (DA) neurons in the brain, whereas fenfluramine targets serotonin (5-HT) neurons. Analogous combination pharmacotherapies have proven beneficial in the treatment of various diseases including hypertension²², diabetes²³, and possibly drug addiction. Regrettably, fenfluramine was associated with serious side-effects, including cardiac valve disease (CVD), which prompted removal of this medication from the market.

Obesity and drug addiction are distinct clinical disorders that share underlying neurocircuitry and neurochemical mechanisms²⁴⁻²⁶. Thus, it is not surprising that efforts to develop medications for treating cocaine and methamphetamine addiction (i.e., stimulant addiction) can provide insights for similar efforts underway to treat obesity. In particular, our work has led us to advocate the development of single molecular entities that target two different neurochemical mechanisms for treating stimulant addiction²⁷⁻²⁹. Since anorectic medications and illicit stimulants interact with biogenic amine transporters in the CNS, our efforts have focused on determining the mechanism of action of these drugs. Additionally, determining the mechanisms underlying adverse effects of anorectic medications is necessary to aid in the development of new medications devoid of serious side-effects. In this paper, the pharmacology of selected anorectic agents is reviewed. We then discuss evidence supporting the hypothesis that fenfluramine-associated CVD is caused by the agonist actions of norfenfluramine, the major metabolite of fenfluramine, at 5-HT_{2B} receptors³⁰. Finally, suggestions are made with regard to the feasibility of employing combination therapies as adjuncts in the management of obesity.

Biogenic amine transporters

Many appetite suppressants are amphetamine-related stimulants, and these agents interact with biogenic amine neurons in the CNS. Neurons that synthesize, store, and release amine transmitters - NE, DA, and 5-HT - are widely distributed in the mammalian CNS. These neurons express specialized plasma membrane proteins that transport previously released transmitter molecules from the extracellular space back into the cytoplasm^{31, 32}. Substantial evidence has shown that there are distinct transporter proteins expressed by NE neurons (NET), DA neurons (DAT), and 5-HT neurons (SERT). These proteins belong to a superfamily of Na⁺/Cl⁻ dependent transporters that share genetic, structural, and functional homologies^{33, 34}. Under normal circumstances, the transporter-mediated uptake of amine transmitters is the principal mechanism for inactivation of amine signaling in the brain. The biogenic amine neurotransmitters and their receptors play a critical role in the pathogenesis and treatment of a wide range of psychiatric disorders³⁵.

In general, drugs that target transporter proteins can be divided into two classes based on their precise mechanism of action: reuptake inhibitors and substrate-type releasers. Reuptake inhibitors bind to transporter proteins, but are not themselves transported. As depicted in Figure 1, these drugs elevate extracellular transmitter concentrations by blocking transporter-mediated recapture of transmitter molecules from the synapse. Substrate-type releasers bind to transporter proteins, and these drugs are subsequently transported into the cytoplasm of nerve terminals. Figure 2 illustrates that releasers, often referred to as substrates, elevate extracellular transmitter concentrations by a two-pronged mechanism: (1) they promote efflux of transmitter by reversing the normal direction of transporter activity, and (2) they increase cytoplasmic levels of transmitter by disrupting storage of transmitters in vesicles via interactions with

vesicular monoamine transporters (VMAT) ^{36, 37}. The exact mechanism underlying transporter-mediated release is complex and a topic of intensive investigation ³⁸⁻⁴⁰. Because substrate-type releasing agents must be transported into nerve terminals to promote transmitter release, reuptake inhibitors can block the effects of releasers.

Basic pharmacology of anorectics

Most appetite suppressants that are currently available, or were prescribed in the past, interact with biogenic amine transporters as reuptake inhibitors or substrates. Figure 3 shows the chemical structures of commonly prescribed anorectics. With the exception of sibutramine, these medications are substituted phenylethylamine analogs (i.e. related to amphetamine). Table 1 summarizes the interaction of these anorectics with NET, DAT and SERT. The prototypical anorectic agent (+)-amphetamine ⁴¹ potently releases NE ($EC_{50} = 7$ nM) and DA ($EC_{50} = 24.8$ nM) from rat brain synaptosomes. In contrast, (+)-amphetamine has much weaker effects on 5-HT release ($EC_{50} = 1765$ nM). Consistent with the *in vitro* data, administration of anorectic doses of (+)-amphetamine markedly increases extracellular DA levels in rat brain while having minimal effects on extracellular 5-HT ⁴². Phentermine is one of the more widely prescribed appetite suppressant medications. The data in Table 1 reveal that phentermine is a substrate at NET, DAT and SERT, with its most potent action being NE release ($EC_{50} = 39$ nM). Similar to (+)-amphetamine, administration of anorectic doses of phentermine evokes DA release in the brain with lesser effects on 5-HT release ⁴³. Ephedrine has a profile of transporter activity that is similar to phentermine.

Studies conducted in baboons indicate that phentermine and ephedrine may not release DA at clinically-relevant doses ⁴⁴. In one study, high (3 mg/kg) intravenous doses of phentermine and ephedrine increased plasma NE levels, but did not suppress plasma prolactin or release central DA, as detected by displacement of [¹¹C]raclopride binding. By contrast, intravenous (+)-amphetamine suppressed plasma prolactin and released central DA, in addition to increasing plasma NE concentrations. The possibility that phentermine and ephedrine do not release DA in humans could explain why these medications have lower abuse liabilities than (+)-amphetamine. It will be of interest to verify these results in controlled clinical investigations.

Diethylpropion and phendimetrazine display minimal interactions with monoamine transporters *in vitro*, even though these drugs produce psychomotor stimulation when administered *in vivo*. For example, phendimetrazine and diethylpropion are self-administered by animals ^{45, 46}, and both drugs exhibit discriminative stimulus properties that generalize to cocaine ^{47, 48}. The available data suggests that diethylpropion and phendimetrazine are “prodrugs” which are converted to bioactive metabolites upon systemic administration. In the case of diethylpropion, the *N*-deethylated metabolite *N*-ethylaminopropiophenone appears to be the bioactive metabolite since this compound potently releases NE ($EC_{50} = 99.3$ nM) with less potent effects on 5-HT release ($EC_{50} = 2118$ nM). Interestingly, *N*-ethylaminopropiophenone is not a DAT substrate, but instead blocks DA reuptake ($EC_{50} = 1014$ nM) ⁴⁹. In the case of phendimetrazine, the *N*-demethylated metabolite phenmetrazine potently releases NE and DA but not 5-HT.

(±)-Fenfluramine (PondiminTM) and its more potent stereoisomer, (+)-fenfluramine (dexfenfluramine, ReduxTM), are anorectic agents that were widely prescribed until their removal from the market due to the occurrence of CVD ⁵⁰. Historical evidence established that fenfluramines stimulate 5-HT transmission in the CNS by increasing synaptic levels of 5-HT ⁵¹. More recent data indicate that fenfluramines and their major metabolites, norfenfluramines, increase synaptic 5-HT by acting as substrates for SERT (for review see ⁵²). *In vivo* microdialysis studies confirm that 5-HT release evoked by (±)-fenfluramine or (+)-

fenfluramine is antagonized by pretreatment with the SERT inhibitor fluoxetine (Prozac™) 53, 54.

Pharmacokinetic investigations show that stereoisomers of (±)-fenfluramine are *N*-de-ethylated by liver enzymes to yield the metabolites, (+)- and (-)-norfenfluramine (see Figure 4) 55. In humans and animals treated with systemically administered (±)-fenfluramine, circulating concentrations of (+)- and (-)-norfenfluramine are similar to or greater than concentrations of fenfluramine itself 55, 56. Moreover, the stereoisomers of (±)-fenfluramine and (±) norfenfluramine cross the blood-brain barrier to accumulate in the brain. Thus, peripheral administration of (±)-fenfluramine gives rise to four pharmacological agents with potential neurobiological activity. With few exceptions 57-59, investigations examining the neuropharmacology of fenfluramines and norfenfluramines have focused on the 5-HT effects of these drugs (for review see 52).

As reported in Table 1, the most potent action of (±)-fenfluramine and its stereoisomers is to evoke [³H]5-HT release, whereas these compounds are essentially inactive in the DA release assay. (+)-Fenfluramine releases [³H]5-HT with an EC₅₀ value of 51.7 nM whereas (-)-fenfluramine releases [³H]5-HT with an EC₅₀ value of 147 nM, a 2.8-fold difference in potency. In the [³H]NE release assay, (+)-fenfluramine displays appreciable activity (EC₅₀ = 302 nM) that is about 6-fold weaker than its activity in the [³H]5-HT release assay. (-)-Fenfluramine is very weak in NE release assay. Thus, (-)-fenfluramine appears more selective than (+)-fenfluramine as a [³H]5-HT releaser, but the (-)-isomer is generally less potent.

(±)-Norfenfluramine and its stereoisomers are much more potent at releasing [³H]5-HT when compared to [³H]DA. (+)-Norfenfluramine releases [³H]5-HT with an EC₅₀ value of 59.3 nM, whereas (-)-norfenfluramine releases [³H]5-HT with an EC₅₀ value of 287 nM. (±)-Norfenfluramine and its stereoisomers are much more potent at releasing [³H]NE and [³H]DA than fenfluramines. For example, (+)-norfenfluramine releases [³H]NE with an EC₅₀ value of 72.7 nM, compared to (+)-fenfluramine, which releases [³H]NE with an EC₅₀ value of 302 nM (see above). It is important to note that norfenfluramines release [³H]NE and [³H]5-HT with roughly equivalent potency. Additionally, the potency of (+)-norfenfluramine to evoke [³H]NE release is similar to the potency of phentermine, a known NE-releasing agent. Consistent with these in vitro data, in vivo microdialysis experiments showed that (+)-fenfluramine produced dose-related increases in extracellular NE in the frontal cortex of rats 59. These data indicate that noradrenergic, as well as serotonergic, mechanisms contribute to the anorectic effect of fenfluramine.

Unlike amphetamine and related compounds described above, sibutramine and its metabolites are uptake inhibitors that vary in their potency and selectivity for NET, DAT and SERT (Table 1) 60. It is notable that sibutramine itself is relatively less potent as an uptake inhibitor than its metabolites, indicating that sibutramine acts as a pro-drug. Several of the metabolites are potent DA uptake inhibitors, perhaps providing an explanation for the greater weight loss observed at higher than approved doses (30 mg per day) 15.

Fenfluramine and cardiac valvulopathy

As mentioned already, (±)-fenfluramine and (+)-fenfluramine were removed from clinical use due to the occurrence of CVD in some patients 50. A meta-analysis of available data indicates the odds ratio for fenfluramine-associated CVD is 2.0, suggesting a moderate albeit significant risk for the disease in patients taking these medications 61. Although a serotonergic mechanism was suspected as a possible cause of CVD 62, little was known about the pathogenesis of this adverse effect in the late 1990s. In light of the established role of 5-HT as a mitogen 63, we carried out a study to determine if stereoisomers of fenfluramine or norfenfluramine might activate mitogenic 5-HT receptors 30. Several other drugs were examined to provide both

positive and negative controls. Additional positive control drugs included methysergide, its active metabolite methylergonovine⁶⁴ (see Figure 4), and ergotamine. Methysergide and ergotamine are well known to produce primarily left-sided CVD affecting the mitral valve^{65, 66}. Negative controls included phentermine, fluoxetine and its metabolite, norfluoxetine, which are not associated with CVD. We also included the antidepressant trazodone and its metabolite *m*-chlorophenylpiperazine (mCPP) as additional negative control drugs. mCPP displays agonist activity at a wide range of 5-HT receptors⁶⁷ and also is capable of releasing 5-HT via a SERT-mediated mechanism⁶⁸. The clinical use of trazodone is not associated with VHD. Our working hypothesis was that positive control drugs would share in common the ability to activate a mitogenic 5-HT receptor subtype expressed in heart valves, whereas negative control drugs would not have this effect. An initial receptorome screen⁶⁹ led to a detailed evaluation of the binding of these drugs to the 5-HT₂ family of 5-HT receptors.

Table 2 reports binding data, while Table 3 reports functional effects of these compounds at cloned human 5-HT_{2A}, 5-HT_{2B} and 5-HT_{2C} receptor subtypes. Fenfluramines had low affinity for all 5-HT₂ receptor subtypes. In contrast, norfenfluramines had high affinity ($K_1 = 10\text{-}50$ nM) for 5-HT_{2B} receptors but not for 5-HT_{2A} or 5-HT_{2C} sites, in confirmation of other studies^{70, 71}. Functional studies demonstrated that norfenfluramines were full agonists at the 5-HT_{2B} site. Ergotamine was a potent partial agonist at the 5-HT_{2B} receptor, whereas methysergide was a low efficacy partial agonist at this site. Methylergonovine, the active metabolite of methysergide, was a high affinity partial agonist at 5-HT_{2B} sites. Among the negative control drugs, mCPP was a partial agonist at 5-HT_{2B} receptors with the same efficacy as methylergonovine. With the exception of the findings with mCPP, our data suggested that the receptor responsible for producing CVD was the 5-HT_{2B} receptor. Trazodone binds with moderate affinity to the 5-HT_{2B} receptors but functions as an antagonist. The 5-HT_{2B} antagonist activity of trazodone probably explains why trazodone administration in humans is not associated with VHD.

Among the receptors assayed, several lines of evidence indicated that the 5-HT_{2B} receptor mediates the valvulopathic effects of fenfluramine: (1) 5-HT_{2B} receptors are located on both mitral and aortic valves⁷⁰; (2) these receptors mediate mitogenesis⁷²; (3) norfenfluramines have high affinity and efficacy at the 5-HT_{2B} receptor; (4) methylergonovine, the active metabolite of methysergide, is a high affinity partial agonists for the 5-HT_{2B} receptor; (5) most of the negative control drugs (fluoxetine, norfluoxetine, phentermine) have very low affinity and lack agonist effects at this receptor. The finding that mCPP has agonist activity at 5-HT_{2B} receptors must be reconciled with observations that trazodone is not associated with CVD. Therapeutic oral doses of trazodone generate plasma levels of mCPP from 150-550 nM⁷³, and these concentrations are within the range needed to activate 5-HT_{2B} receptors. However, trazodone is a potent 5-HT_{2B} receptor antagonist, and its plasma levels are about five-fold higher than that of mCPP⁷³. Thus, when trazodone is administered in vivo, antagonist actions of the drug would serve to block activation of 5-HT_{2B} receptors by mCPP.

Further support for the 5-HT_{2B} receptor hypothesis of drug-induced CVD comes from studies of 3,4-methylenedioxymethamphetamine (MDMA). Setola et al. reported that MDMA and its *N*-demethylated metabolite, 3,4-methylenedioxyamphetamine (MDA) (see Figure 4), are 5-HT_{2B} receptor agonists that elicit prolonged mitogenic responses in human valvular interstitial cells⁷⁴. As predicted by the in vitro findings, chronic heavy users of MDMA are reported to exhibit an increased prevalence of valvular regurgitation indicative of CVD⁷⁵. The critical role of 5-HT_{2B} receptors in mediating CVD has been strengthened by recent data demonstrating that two ergot medications known to cause valvulopathy- carbergoline and pergolide- are potent agonists at 5-HT_{2B} receptor sites (for review see: ⁷⁶). Collectively, these findings indicate the importance of screening potential medications for agonist activity at 5-HT_{2B} receptors⁶⁹. The recent description of lorcaserin as a potential anti-obesity medication

illustrates this strategy⁷⁷. Lorcaserin displays potent 5-HT_{2C} agonist activity that is much greater than its effects on 5-HT_{2B} receptors. This medication is currently in phase 3 clinical trials (<http://www.arenapharm.com/wt/page/lho.html>)⁷⁸.

Combination therapy for obesity

As noted in the Introduction, the regulation of appetite, food intake and body weight are controlled by multiple neuronal mechanisms working in a parallel manner¹². Medications that targets one neurochemical mechanism produce a relatively small degree of weight loss, such as that observed with the cannabinoid receptor antagonist rimonabant⁷⁹. While weight loss of this magnitude can produce significant reductions in risk factors associated with cardiovascular morbidity and mortality¹³⁻¹⁵, patients often expect more significant weight loss¹⁶. Combination pharmacotherapy is one potential strategy for obtaining greater, cosmetically-relevant, reductions in weight; this approach is illustrated by phentermine/fenfluramine combination pharmacotherapy¹⁷⁻²¹. As shown in Table 1, phentermine releases NE and DA from neurons whereas fenfluramines release 5-HT. By recruiting both catecholaminergic and serotonergic mechanisms, the phentermine/fenfluramine combination engenders greater effects on weight loss than either treatment administered alone.

The occurrence of CVD in patients receiving phentermine/fenfluramine treatment raised serious concerns about the use of anti-obesity medications⁸⁰, and more broadly about using combinations of medications⁸¹. Based on the research reviewed in this article, it is now apparent that CVD associated with the use of phentermine/fenfluramine resulted from activation of 5-HT_{2B} receptors localized to heart valves. The major culprits in stimulating these receptors are the fenfluramine metabolites, (+)- and (-)-norfenfluramine. Importantly, there is no evidence to suggest that phentermine alone increases the risk for CVD, or that co-administration of phentermine with fenfluramine somehow augments the risk posed by fenfluramine.

Having identified the likely cause of fenfluramine-associated CVD, the principle of combining phentermine with a serotonergic agent to achieve improved efficacy in the treatment of obesity remains a valid approach worthy of further exploration. The one important caveat is that serotonergic medications should not activate 5-HT_{2B} receptors. For example, the combination of phentermine with the 5-HT precursor, L-5-hydroxytryptophan, is a feasible means of increasing synaptic DA and 5-HT. This combination would be predicted to display superior efficacy as an appetite suppressant. Another approach that we have suggested²⁹ is the development of single molecular entities that release DA and 5-HT⁸². As long as such treatments do not activate 5-HT_{2B} receptors they should be safe and efficacious mediations. Finally, reinforcing the fact that obesity and drug addiction are driven in part by common neurochemical mechanisms²⁶, it is predicted that research conducted to discover medications for treating stimulant addiction will provide clues for the development of medications to treat obesity.

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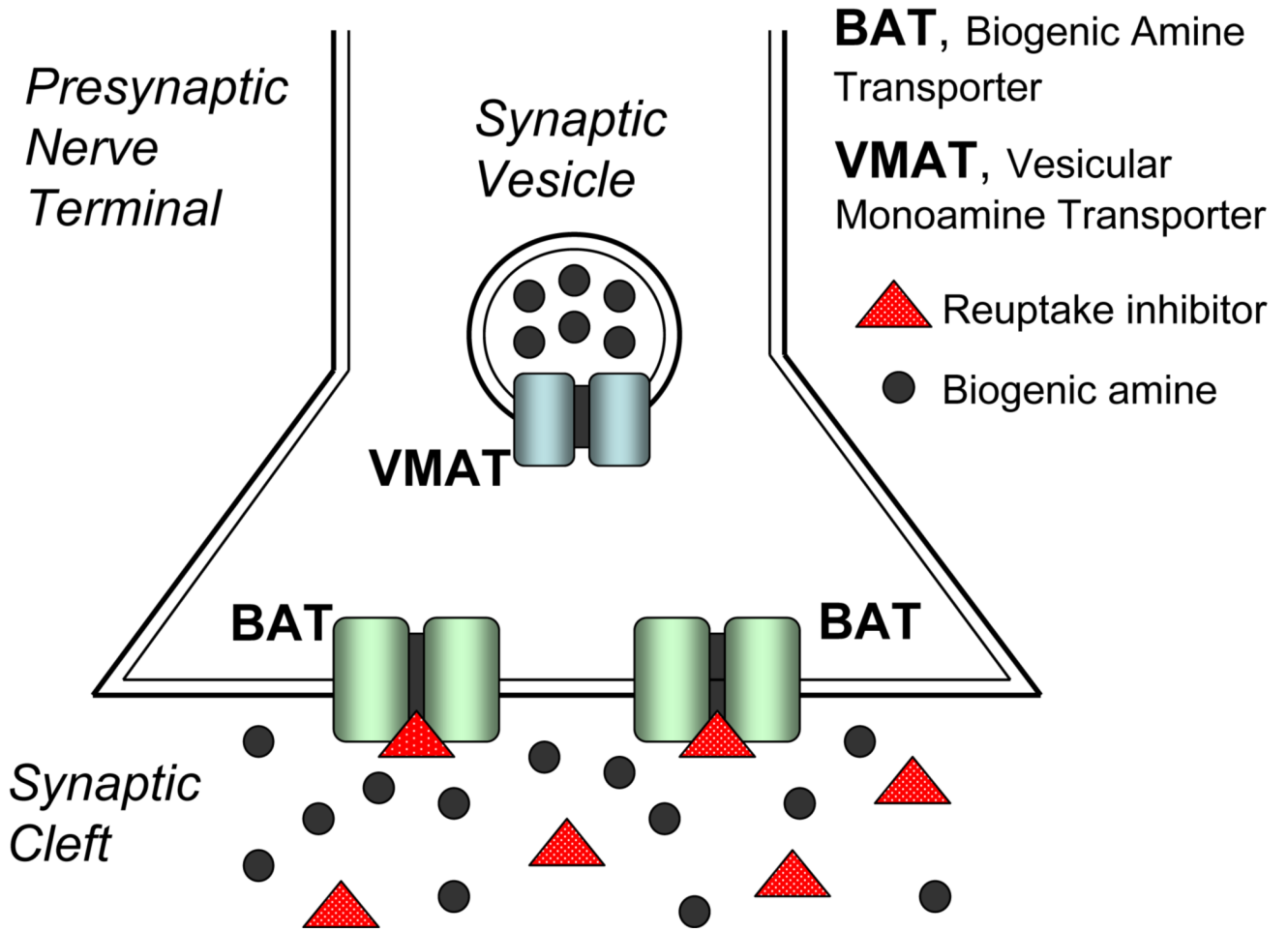


Figure 1. Mechanism of biogenic amine transporter (BAT) reuptake inhibitors. Reuptake inhibitors increase synaptic transmitter concentrations by binding to BAT proteins and blocking the reuptake of previously released biogenic amine transmitters.

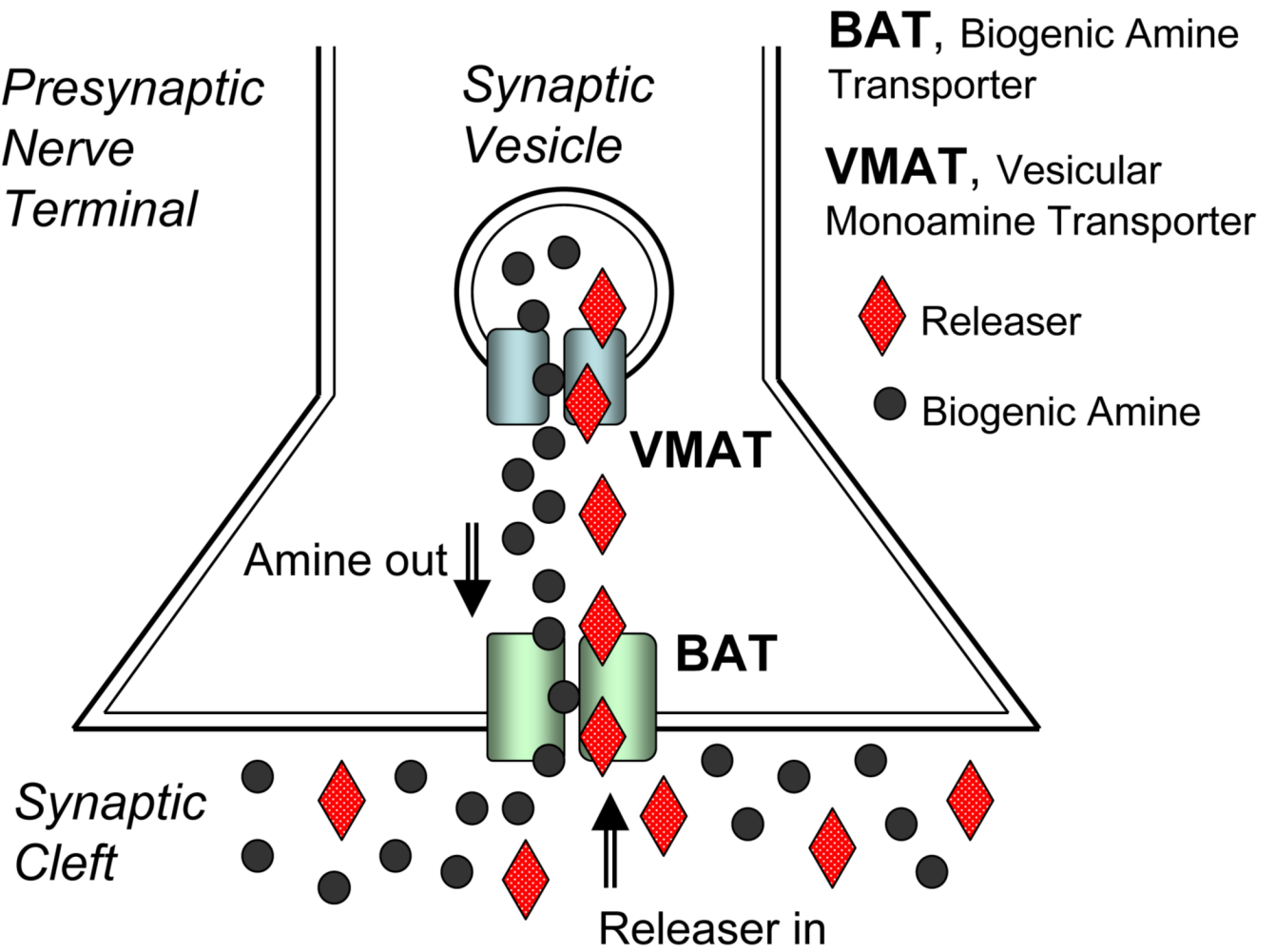
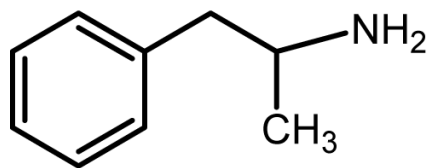
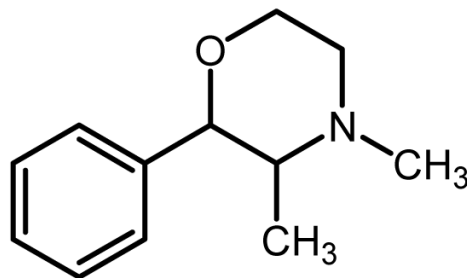


Figure 2.

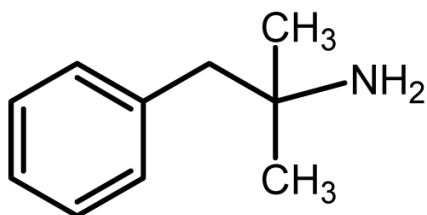
Mechanism of biogenic amine transporter (BAT) substrate-type releasers. Releasers increase synaptic transmitter concentrations by a two pronged mechanism: 1) they promote biogenic amine efflux out of the cell by reversing the normal direction of transporter activity, and 2) they disrupt the storage of biogenic amines in vesicles by interacting with vesicular monoamine transporters (VMAT). The disruption of biogenic amine storage increases the cytoplasmic concentrations of amines available for release.



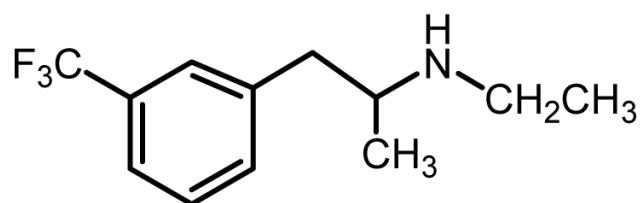
Amphetamine



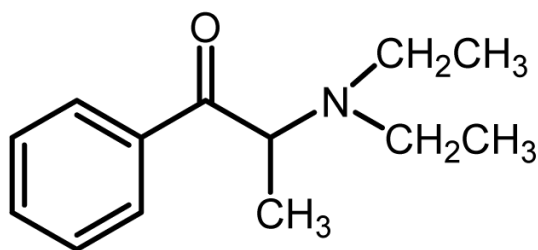
Phendimetrazine



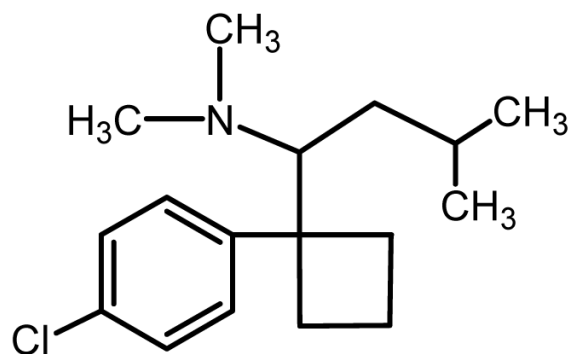
Phentermine



Fenfluramine



Diethylpropion



Sibutramine

Figure 3. Chemical structures of commonly prescribed anorectic medications. With the exception of sibutramine, these medications are substituted phenylethylamine analogs, related to amphetamine. Fenfluramines are no longer available for clinical use.

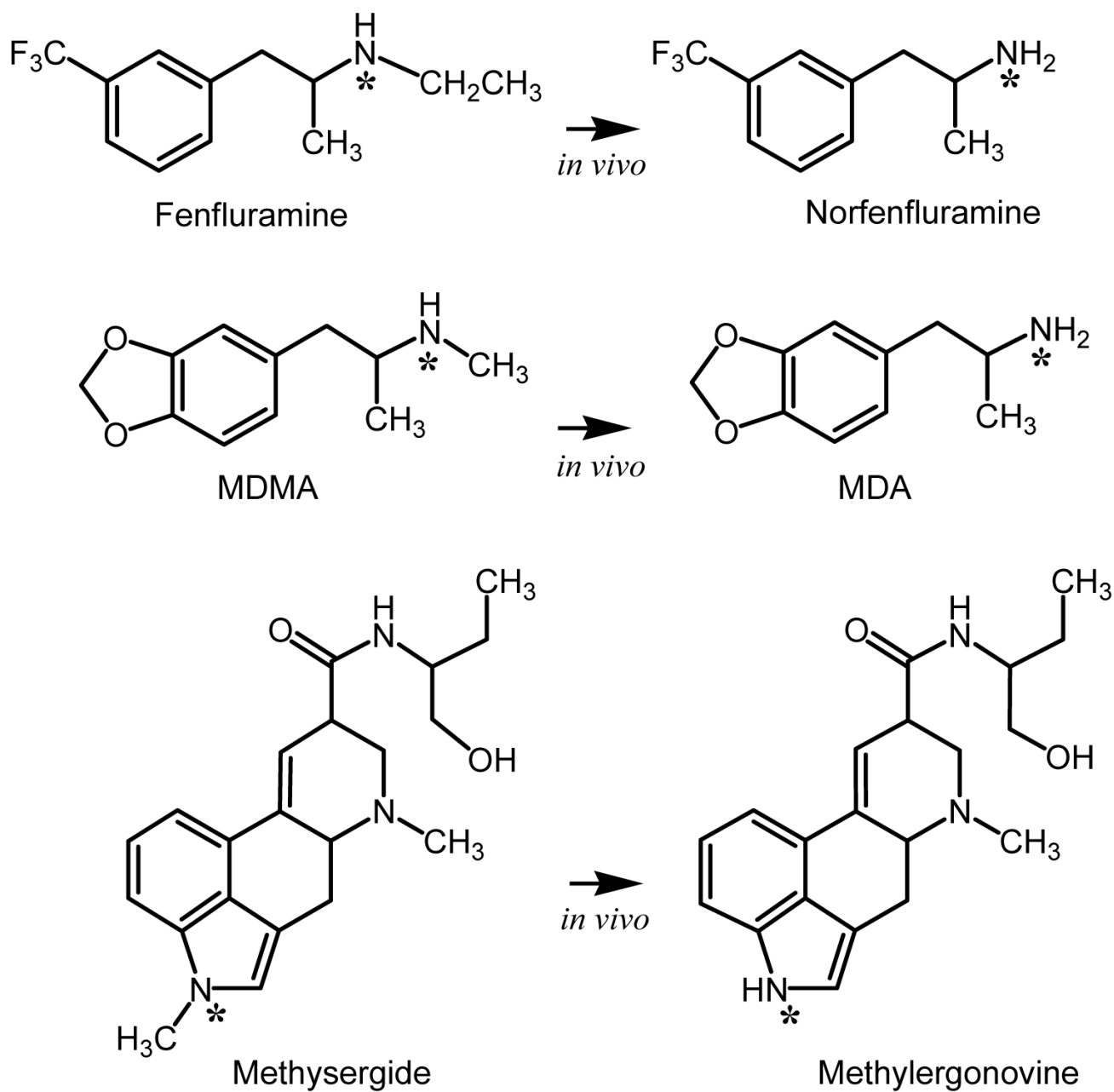


Figure 4. In vivo metabolism of selected drugs to form potent 5-HT_{2B} receptor agonists. In each case illustrated, *N*-demethylation or *N*-deethylation produces bioactive metabolites that display much greater agonist activity at 5-HT_{2B} receptors when compared to their corresponding parent compounds.

Table 1
Interaction of Selected Anorectic Agents with the Biogenic Amine Transporters

Test Drug	Release NET ^a EC ₅₀ (nM ± SD)	NE Uptake K _i (nM ± SD)	Release DAT ^a EC ₅₀ (nM ± SD)	DA Uptake K _i (nM ± SD)	Release SERT EC ₅₀ (nM ± SD)	5-HT Uptake K _i (nM ± SD)
(+)-Amphetamine	7.1 ± 0.9		24.8 ± 3.5		1765 ± 94	
Phentermine	39.4 ± 6.6		262 ± 21		3511 ± 253	
(-)-Ephedrine	43.1 ± 4.0		236 ± 9		>10,000	>50,000
(+)-Ephedrine	218 ± 14		2104 ± 68		inactive	
Diethylpropion	> 10,000	> 10,000	> 10,000	> 10,000	> 10,000	> 10,000
N-Ethylaminopropiophenone	99.3 ± 6.6			1014 ± 80	2118 ± 98	
Phendimetrazine	8300 ± 445	>10,000	19,000 ± 537	>10,000	>100,000	>100,000
(±)-Phenmetrazine	50.4 ± 5.4		131 ± 11		7765 ± 610	
(+)-Fenfluramine	302 ± 20			22000 ± 1100	51.7 ± 6.1	
(-)-Fenfluramine		7187 ± 559	>10,000	>20,000	147 ± 19	
(±)-Fenfluramine	739 ± 57			23700 ± 1300	79.3 ± 11.5	
(±)-Norfenfluramine	168 ± 17		1925 ± 295		104 ± 5	
(+)-Norfenfluramine	72.7 ± 5.4		924 ± 112		59.3 ± 2.4	
(-)-Norfenfluramine	474 ± 40			19194 ± 1048	287 ± 14	
(R,S)-Sibutramine ^b		350		1200		2800
(R)-DesmethyIsibutramine ^b		4		12		44
(S)-DesmethyIsibutramine ^b		870		180		9200
(R)-DidesmethyIsibutramine ^b		13		8.9		140
(S)-DidesmethyIsibutramine ^b		62		12		4300

The data in the Table are taken from: 28.

^a Some studies used [³H]MPP⁺, with appropriate blocking agents, to measure release via DAT and NET.

^b The sibutramine data are taken from: 61.

Table 2Ki Values of Test Drugs at 5-HT₂ Receptors

Drug	Human 5-HT _{2a}	Human 5-HT _{2B}	Human 5-HT _{2C}
(±)-Fenfluramine	5216 ± 423	4134±1281	3183 ± 637
(+)-Fenfluramine	11107 ± 2303	5099 ± 1173	6245 ± 874
(-)-Fenfluramine	5463 ±600	5713 ± 2285	3415 ± 922
(±)-Norfenfluramine	2316 ± 278	52.1±21	557 ± 61
(+)-Norfenfluramine	1516 ± 150	11.2 ± 7.3	324 ± 12
(-)-Norfenfluramine	3841 ± 614	47.8 ± 30.6	814 ± 98
Ergotamine	9.0 ± 1.0	3.0 ± 0.4	12 ± 1.5
Methysergide	15.0 ± 4.0	9.1 ± 4.9	1.8 ± 0.2
Methylergonovine	12.6 ± 1.0	0.49 ± 0.16	12.4 ± 1.0
Fluoxetine	299 ± 53	5030±1960	50 ± 10
Norfluoxetine	638 ± 108	5063±1974	286 ± 60
Trazodone	19.8±2.4	73.6 ± 36	402±44
m-CPP	391±47	3.2 ± 1.0	59±11
5-HT	614±74	4.0 ± 1.9	12.2±1.3
Phentermine	>10,000	>10,000	>10,000

Values are mean ± SD for n=3 experiments. Data taken from Data taken from ³⁰.

Table 3
Functional Activity of Test Drugs at 5-HT₂ Receptors

Drug	Human 5-HT _{2a} Kact (nM±SD) Vmax (Percent of 5-HT±SD)	Human 5-HT _{2B} Kact (nM±SD) Vmax (Percent of 5-HT±SD)	Human 5-HT _{2C} Kact (nM±SD) Vmax (Percent of 5-HT±SD)
(±)-Fenfluramine	4131±2448 15±4	ND	ND
(+)-Fenfluramine	>10,000 Not Done	379 ± 120 38 ± 14	362 ± 109 80 ± 10
(-)-Fenfluramine	5279± 998 43 ± 7.2	1248 ± 430 47 ± 5	360 ± 155 84 ± 15%
(±)-Norfenfluramine	ND	ND	ND
(+)-Norfenfluramine	630 ± 240 88± 9	18.4 ± 9 73 ± 6	13 ± 4 100 ± 11
(-)-Norfenfluramine	1565 ± 323 93 ± 9	357± 180 71 ± 15	18 ± 9 80 ± 17
Ergotamine	16 ± 4 75 ± 3	9.8 ± 3 56 +/- 3	5 ± 3 75 ± 15
Methysergide	3.5 ± 1.7 24 ± 3	150 ± 43 18 ± 4	2.9 ± 1.5 33 ± 3.5
Methylergonovine	1.3 ± 0.4 70 ± 7	0.8 ± 0.5 40 ± 3	2.5 ± 1.2 103 ± 7
Fluoxetine	ND	ND	Antagonist K _i = 616±172
Norfluoxetine	ND	ND	Antagonist K _i = 43±17
Trazodone	Antagonist	Antagonist	Antagonist
m-CPP	65 ± 17 55 ± 11	64 ± 27 43 ± 14	0.64 ± 0.3 79 ± 15
5-HT	66 ± 26 100%	2.4 ± 1.5 100	0.6 ± 0.18 100
Phentermine	ND	ND	1394 ± 450 66 ± 10

Values are mean ± SD for n=3 experiments. Data taken from Data taken from ³⁰.