University of Utah
UNDERGRADUATE RESEARCH JOURNAL

METHCATHINONE DECREASES DOPAMINE TRANSPORTER FUNCTION

Yasmeen Siripathane (Annette Fleckenstein, Charlotte Magee, Matthew Ream)
School of Dentistry

Abstract

Methcathinone (MCAT) is a synthetic cathinone chemically related to the psychostimulant, methamphetamine (METH). MCAT was popularized in Michigan in the 1990's and gained popularity throughout the country, resulting in designation as a schedule I compound. The purpose of this study is to investigate the effects of MCAT on the dopamine (DA) transporter (DAT) because the DAT affects extracellular and intracellular DA concentrations. Using untreated rat striatal tissue, *in vitro* application of MCAT or METH decreased [^{3}H]DA uptake. To ensure that there was no residual drug directly impacting [^{3}H]DA uptake, preincubation of the striatal synaptosomes with MCAT was tested at 37°C and 4°C. At 37°C, MCAT reduced [^{3}H]DA uptake. There was no drug effect at 4°C, suggesting that the drug washes were effective in removing MCAT. Using this *in vitro* model, a concentration response of MCAT revealed that a decrease in [^{3}H]DA uptake occurred at 100 µM, 10 µM, and 1 µM. These data suggest that dopaminergic (DAergic) pathways are affected by MCAT. The implications of these findings will be discussed.

Introduction

Methcathinone (MCAT) is a severely addictive substance that can cause physical and psychological dependence in humans. The rapid neurochemical impact of MCAT on the dopamine (DA) transporter (DAT) is largely unknown. Investigating this interaction is important because the mechanism by which MCAT affects the DAT determines the levels of DA found intracellularly and extracellularly. Large amounts of extracellular DA promote the formation of reactive oxygen species (Fleckenstein et al, 1997). Pre-clinical studies involving "binge" MCAT treatment of rodents reveal that this compound has a steep dose-response profile. Moreover, preclinical studies indicated that multiple administrations of MCAT cause deficits in the nigrostriatal dopaminergic (DAergic) system for up to 30 days (Gygi et al, 1997). The goal of this current study was to extend this work by determining the impact of MCAT on the DAT.

Methods

Striatal tissue was obtained from untreated male rats. The tissue was homogenized and centrifuged (800 x g, 12 min). The supernatant fractions were then removed and centrifuged (22,000 x g, 15 min) to obtain the pellets containing synaptosomes. The pellet was resuspended in assay buffer (in nM): 126 NaCl, 4.8 KCl, 1.3 $CaCl_2$, 1.4 $MgSO_4$, 11 glucose, 3.7 $NaPO_4$ monobasic, 1 ascorbic acid, 12.7 $NaPO_4$ dibasic. In MCAT preincubation experiments, samples were preincubated at 37°C unless stated otherwise in the figure legends (i.e., in the

legend to Fig. 2). Uptake of [3H]DA via DAT was performed as described in Sandoval et al (2001). Bradford protein assays were conducted to normalize data.

Results

In Vitro Application of MCAT and METH, but not MDPV or Nicotine, Decreases Striatal [3H]DA Uptake

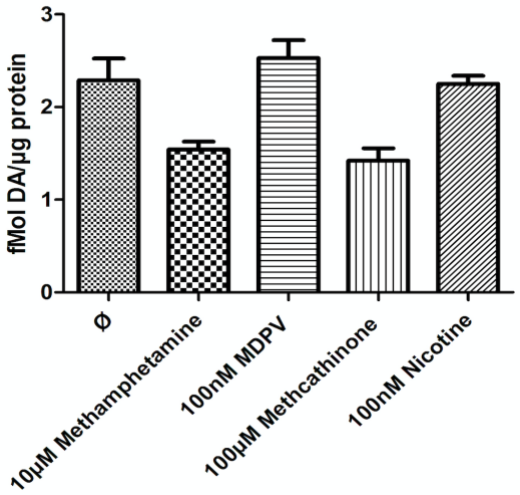

Figure 1. Striatal synaptosomes were incubated with the various agents for 30 min at 37˚C. [3H]DA uptake assays were conducted as described in Methods.

In Vitro MCAT Application at 4˚C Does Not Alter Striatal [3H]DA Uptake

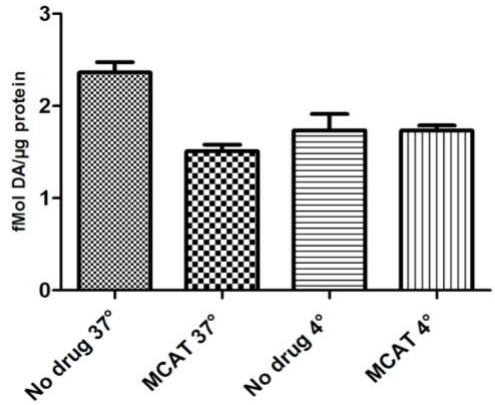

Figure 2. Striatal synaptosomes were preincubated with MCAT or vehicle for 30 min at 37°C or 4°C. [3H]DA uptake assays were conducted as described in Methods.

In Vitro MCAT Application Decreases Striatal [3H]DA Uptake

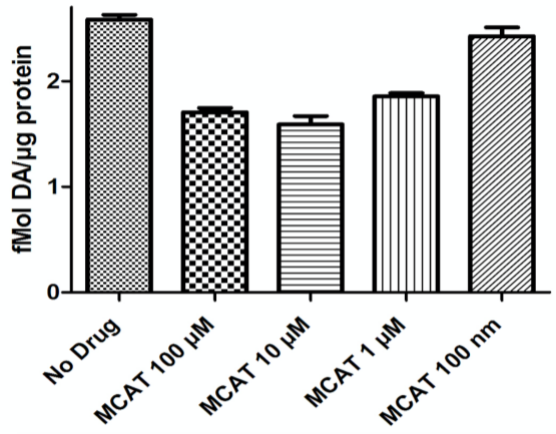

Figure 3. Striatal synaptosomes were preincubated with MCAT or vehicle for 30 min at 37°C. [3H]DA uptake assays were conducted as described in Methods.

Conclusions

In Vitro MCAT application decreases [3H]DA uptake; an effect that appears independent of residual drug. This permits speculation that MCAT promotes accumulation of DA in the synaptic cleft and the formation of reactive species. The reactive species may lead to persistent DAergic deficits (Gygi et al, 1997).

References

Fleckenstein AE; Metzger RR; Beyeler ML; Gibb JW; Hanson GR. Oxygen radicals diminish dopamine transporter function in rat striatum. European Journal of Pharmacology. 1997; ISSN: 0014-2999, Vol: 334, Issue: 1, Page: 111-114

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