G58 MDMA Neurotoxicity

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After attending this presentation, attendees will gain knowledge on how to characterize MDMA neurotoxicity in rat brain.

This presentation will impact the forensic community by demonstrating how the results of even one single administration of MDMA can significantly alter the brains cellular antioxidant defense system and produce oxidative stress in both the striatum, and frontal cortex. Thus one possible mechanism of MDMA neurotoxicity appears to be a direct toxic effect of MDMA or its red-ox active metabolites

3, 4-Methylenedioxymethamphetamine (MDMA or "Ecstasy") is an increasingly popular psychoactive and hallucinogenic drug of abuse. It acts on the CNS by increasing the release of serotonin and other catecholamines in addition to preventing their reuptake. MDMA has been shown, in both man and animal, to damage serotonergic and dopaminergic nerve terminals and to cause neurodegeneration in multiple areas of the brain, including the cortex, hippocampus, striatum and thalamus. The closely related drug, methamphetamine (METH) and its derivates have been shown to produce long-lasting depletion in dopamine and its metabolites, as well as dopamine reuptake sites in the rat and primate striatum, but not in other dopamine rich areas such as the nucleus accumbens and the prefrontal cortex, in contrast to the neurotoxic effect of dopamine to striatal DA terminals. Two other important aspects of MDMA neurotoxicity have been identified: hyperthermia and neurodegeneration. The former appears to be a direct action of MDMA, while the later is due to the production of reactive oxygen (ROS). Mounting evidence suggests that MDMA-induced 5HT neurotoxicity is due to the increased production of free radical induced oxidative stress. Attempts were made to clarify the mechanisms of MDMA in rats' brain by administering a single dose of the drug and studying the effects using combined toxicological, biochemical and immunohistochemical analysis.

Fifty rats were used for the study, each weighing 200-250 grams. Twenty-five rats were used for the histopathological and toxicological examination. They were divided into three experimental groups of seven animals each and administered one 20mg/Kg dose of MDMA intraperitoneally. The four controls were injected with saline. The first group of animals was sacrificed six hours after injection, the second at 16 hours, and the third at 24 hours. Plasma samples obtained immediately after sacrificed, stored at – 80°C and then analyzed for MDMA/MDA with gas chromatography/mass spectroscopy (GC-MS). Histological sections of the brains were also obtained and immunohistochemical stains were used to localized MDMA and its metabolites, MDA and MDEA, within the various areas of the brain. Other immunohistochemical stains were used to localized growth associated protein 43 (GAP43}, tryptophan hydroxylase (TrypH), markers of synaptic plasticity of the serotonergic innervation and the vesicular monoamine transporter -2 (VMAT2) as a stable marker of striatal dopaminergic terminal integrity. Microglial activation and damage were measured using a different immunohistochemical stain for glial fibrillary acid protein (GFAP), Heat shock proteins (Hsp 27, Hsp 70 and Hsp 90) and beta-amyloid precursor protein (βAPP). Apoptosis was measured using the tunnel assay, which identified apoptosis via DNA fragmentation. For the evaluation of oxidative stress, the others twenty- five rats, divided into three groups of seven male albino rats weighing 200-250 g were used to analyze the effect of MDMA administration (20 mg/Kg, i.p) on rats' brain. Hippocampus, striatum, and frontal cortex were removed 3 and 6 hours after treatment and analyzed for the activity of antioxidant enzymes (super oxide dismutase, SOD; glutathione reductase, GR; glutathione peroxidase, GPx). Reduced and oxidized glutathione (GSH and GSSG) were measured using a spectrophotometric assay. Ascorbic acid (AA) levels were determined reverse-phase HPLC method. Finally, lipid peroxidation was measured by quantitating the release of malonaldehyde (MDA) using UV-HPLC.

The acute administration of MDMA produces a decrease of GSH/GSSG ratio and oxidative stress in all of the brain areas examined. SOD activity was significantly reduced after 3 hours in hippocampus (- 60.7%) and after 6 hours in striatum, hippocampus, and frontal cortex (- 43.3%, -86.1% and -23.4% respectively). GR and GPx activities were reduced after 3 hours (-22%) and after 6 hours (-33.3%) in frontal cortex. AA levels strongly increased in striatum, hippocampus and frontal cortex after 3 (+159%, +84% and 17.6%) and 6 (+162%, +154% and +23.4%) hours respectively. High levels of MDA respect to control were measured in striatum after 3 hours (+276%) and 6 hours (162%); in hippocampus (71.8%) and in frontal cortex (+18.22%) after 6 hours.

The results of even one single administration of MDMA can significantly alter the brains cellular antioxidant defence system and produce oxidative stress in both the striatum and frontal cortex. Thus one possible mechanism of MDMA neurotoxicity appears to be a direct toxic effect of MDMA or its redox active metabolites.

MDMA Neurotoxicity, Immunohistochemical, Oxidative Stress

 ** Presenting Author*