A

A SINGLE INJECTION OF IBOGAINE PRODUCES SELECTIVE NEUROCHEMICAL CHANGES IN MOUSE BRAIN

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The alkaloid ibogaine and other indole alkaloids have been known to have hallucinogenic as well as stimulant properties. Recently, the U.S. Food and Drug Administration (FDA) and National Institute on Drug Abuse (NIDA) has approved the use of ibogaine on a limited basis to treat cocaine addiction. Several laboratories have reported that single or multiple injections of ibogaine produced neurochemical changes, however, its mechanism of action and neurotoxic potential are still unknown. The present study was designed to determine whether a single injection of ibogaine would produce significant neurochemical alterations in the mouse brain. Adult male C57/B6N mice were dosed with 50 mg/kg ibogaine, ip, and sacrificed 0.5, 1, 2, 4 and 24 hour later. Brains were dissected for neurochemical analyses. In vitro exposure to ibogaine (0.5-250 uM) produced dose-dependent decreases in nitric oxide synthase (NOS) activity in cerebellum as measured by a radioactive method in which the rate of conversion of ^{3}H-arginine to ^{3}H-citrulline was monitored. In vivo exposure to ibogaine also produced a significant decrease in NOS activity in the striatum, hippocampus and cerebellum. Monoamine concentrations measured by HPLC/EC showed a significant depletion of dopamine (DA) at 0.5, 1, 2 and 4 hr after drug administration, however, levels returned to control values 24 hr after dosing. Serotonin (5-HT) concentration in the striatum decreased 4 and 24 hours after dosing. These data indicate that a single injection of ibogaine can produce significant alterations in NOS activity, and DA and 5-HT concentrations which suggest that ibogaine has a complex neurotoxicity profile and could interfere with several major neural functions.

B

PHYSICOCHEMICAL AND METABOLIC BASIS FOR THE NEUROTOXICITY OF THE PYRROLIZIDINE ALKALOID, TRICHODESMINE

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The pyrrolizidine alkaloids (PAs) monocrotaline (MONO) and trichodesmine (TRIC) differ structurally only in the substitution of an isopropyl group in TRIC for a methyl in MONO. However, TRIC is neurotoxic whereas MONO is primarily hepato- and pneumotoxic. Toxicity is due to hepatic metabolism to the corresponding dehydroalkaloids (DHAs), which are alkylating agents. We have established that: (i) TRIC (15 mg/kg i.p.) leads to neuronal cell death in the cerebral cortex in rats after 10 days whereas an equitoxic dose of MONO (60 mg/kg) does not; (ii) Fourfold greater amounts of DHA are released from isolated rat livers perfused with TRIC than are seen with the same concentration of MONO; (iii) Dehydrotrichodesmine is more stable than dehydromonocrotaline in aqueous solution, the former having a half-life of 5.4 $\pm$ 0.9 sec and the latter a half-life of 3.4 $\pm$ 1.0 sec ($p < .01$); (iv) that TRIC is more liposoluble than MONO, the partition coefficient with phosphate buffer (100 mM; pH 8.5) in chloroform being 107 for TRIC and 6.7 for MONO, and in heptane being 0.047 for TRIC and 0.005 for MONO; and (v) injection i.p. of the parent PAs lead to a tenfold greater accumulation of protein-bound DHAs in the brain for TRIC compared with MONO. We conclude that the neurotoxicity of TRIC is largely a phenomenon of greater delivery of DHA from the liver to the brain combined with a greater ability of dehydrotrichodesmine to penetrate the brain.

C

MONITORING NEURONAL DAMAGE-RELATED CHANGES IN THE LEVELS OF TRANSMITTERAMINES, SOME MAJOR METABOLITES AND SALICYLATE - DERIVED DIHYDROXYBENZOATES IN RODENT BRAIN REGIONS

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A high - performance reversed - phase ion - pair chromatographic method coupled with electrochemical detection was developed for the simultaneous separation of 2,5- and 2,3-dihydroxybenzoates (DHB-s), indicators of in vivo free radical formation, some transmitteramines (DA, 5-HT) and the major metabolites (DOPAC, HVA and 5-HIAA) in rodent brain regions. The method was used to monitor peak pattern and peak size variations in 5 regions of adult SPRD rats and 3 regions of C57BL mice, after neuronal damage caused by ischemia (bilateral carotis occlusion for 3 h) and reperfusion injury (with rats) or by treatment with the neurotoxins MPTP (5×30 mg/kg, i.p.) or (+)-methamphetamine (4× 10 mg/kg, i.p.) (with mice). The brain regions examined were: corpus striatum (STR), substantia nigra (SN), sensory cortex (CX), hypothalamus (HYT) and hippocampus (HIP) (with rats), and STR, SN and HIP (with mice). At time points 15 min—65 h post-ischemia and post-treatment with the neurotoxin, respectively, and 15 min—2.5 h before decapitation, uninjured (control) and post-injury animals were injected i.p. with either a 100 mg/kg or 300 mg/kg dose of salicylate. Statistical analysis of the results of the various tissue samples suggests that, due to an increased metabolic hydroxylation of salicylate to 2,5-DHB in mice, mouse models are less reliable to measure oxidative-stress-induced (OSI) changes in brain. Conversely, in rat HIP, CX and HYT significant changes ($p \leq 0.01$) in the OSI to control concentration ratios were found for DHB-s. In rat HIP, 2,3-DHB and 2,5-DHB OSI levels increased ten-fold and six-fold, respectively, as compared with control levels, and there was also a 2-fold increase in the 5-HIAA/5-HT ratio, offering a good chance for the development of a method for testing agents with a potential lipidperoxidation inhibitory effect.

D

NEUROCHEMICAL PECULIARITIES OF THE BRAIN OF RATS OF DIFFERENT STRAINS.

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The study of some brain formations of Wistar rats differing in the activity in the "open field" test (high "HMA} and low "LMA} motor activity) and of rats with high and low capability for acquiring of conditional reflexes of active escape in shuttle chamber ("well" and "badly learnable" rats) has shown certain morphochemical peculiarities that are manifested on functional (interferometric protein content in nucleus and cytoplasm), regulatory (protein concentration in nucleus and cytoplasm) and tructural nuclear and cytoplasmic sizes) levels. For HMA and LMA rats the differences in the parameters studied were revealed in cortical (layers III and V) and subcortical formations of motor system, but not in the neurones of hippocampus (field CAs). For "well" and "badly learnable" rats differences have been found both in the structures of motor system and in hippocampus. Comparative analysis has shown that in sensomotor cortex the parameters studied are higher in "well learnable" and in LMA animals, in caudate nucleus and in hippocampus - in "badly learnable" and in LMA rats. Neurochemical correlates of behavior are discussed.