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Effect of Styrene on Dopamine Receptors

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Styrene is widely used in the manufacture of polystyrene, styrene-butadiene rubber, styrene resin, and fiberglass (MARK et al. 1969). The principal human effects of exposure to styrene are irritation of the eyes, nose, and respiratory tract (STEWART et al. 1968; SPENCER et al. 1942). Neurotoxicity of styrene involving central and peripheral nervous systems has been reported in industrial workers (LEIBMAN 1975; LORIMER et al. 1976). Workers handling the monomer have been found to show decreased nerve conduction velocity and electrocephalographic changes (RUTH et al. 1978). Besides the industrial workers, general population can also be exposed to styrene due to the leaching of the unreacted monomer (WIETHY 1976). Thus, exposure to styrene can occur through inhalation as well orally.

Our previous observations of increase in levels of catecholamines (HUSAIN et al. 1980) and decrease in monoamine oxidase in brain of styrene exposed rats have suggested an involvement of neurotransmitter function in the central nervous system (CNS) toxicity of the monomer. To further understand this, we are investigating the effects of styrene on neurotransmitter receptors. The present report describes the alteration in binding of ^{3}H-spiroperidol to dopamine receptors after single and repeated exposure to styrene.

MATERIAL AND METHODS

Treatment of animals. Approximately eight-week-old male albino rats obtained from ITRC animal breeding colony were used. Animals were maintained under standard conditions and raised on commercial pellet diet (Hindustan Lever, Bombay) and water ad libitum. Styrene dissolved in peanut oil was given orally by gavage at a dose of 200 and 400 mg/kg in single administration or up to 90 days in repeated administration. To another set of animals, equal volume of peanut oil was administered in the identical manner to serve as control. Six animals each from treated and control groups were killed by decapitation; brains were removed, and corpus striata were dissected out under cold conditions by the method of GLOWINSKI & IVERSEN (1966) and frozen at -20C until the membrane preparation.

Biochemical procedure. Dopamine receptor binding was studied in crude synaptic membrane (AGRAWAL et al. 1981a). In brief, the frozen tissue was homogenized in 0.32M sucrose and centrifuged at

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40,000 X g for 10 min. The pellets were suspended in 19 volumes of deionized water which helps in removing endogenous dopamine as well as lysing the neuronal cells. After a second centrifugation at 40,000 X g, the pellet was finally suspended in tris-HCl (pH 7.4) and stored at -20C.

High affinity binding of ^{3}H-spiroperidol to striatal membranes was assayed essentially as described previously (SETH et al. 1981a). Striatal membranes (equivalent to 5 mg of original wet weight of tissue) containing 300-400 ug of protein (LOWRY et al. 1951) were incubated in triplicate with 1nM ^{3}H-spiroperidol (sp. activity 22 Ci/mmol, New England Nuclear) in presence of 40 mM tris-HCl (pH 7.4) in a final volume of 1.0 mL for 15 min at 37C. To assess the degree of non-specific binding, parallel assays were run in the presence of 10^{-6} M haloperidol. At the end of the incubation samples were filtered on glass fiber discs (25 mm diameter, 0.3 um pore size Gelman Inc., Ann Arbor MI) and rapidly washed three times with 5 mL cold tris buffer. Filters were then dried and counted in 5 mL scintillation mixture (Aquasol) in Packard Tricab scintillation counter with 38-43% efficiency for tritium. Specific binding was determined by substracting the non-specific binding obtained in the presence of competitor haloperidol from the total binding and was corrected for protein concentration. The results are expressed in terms of pmole of ^{3}H-spiroperidol bound/g of membrane protein. The method used was thus essentially similar to other filtration binding methods (YAMAMURA et al. 1978) and satisfies the requirements for saturability, specificity, reversibility, and regional distribution.

RESULTS AND DISCUSSION

The results of single and repeated exposures of styrene on ^{3}H-spiroperidol binding are summarized in figures 1 and 2, respectively. Single exposure of styrene caused a significant increase in the specific binding of ^{3}H-spiroperidol (1nM) at the dose of 200 and 400 mg/kg ($P<0.05$ and $P<0.01$, respectively). The repeated administration of styrene for 90 days caused a significant increase ($P<0.01$) at 200 and 400 mg/kg in the specific binding of ^{3}H-spiroperidol to striatal membranes 24 h after the last dose (fig. 2). No significant effect of styrene exposure was seen on body weight and striatal weight of rats (data are not shown). The present result shows an increase in the binding of ^{3}H-spiroperidol to dopamine receptors in corpus striatum of rats after single or repeated exposure to styrene. The increase in binding may be due to alterations in the affinity of receptor sites or increase in the number of receptors. The increased sensitivity of dopamine receptors may be due to the destruction of dopamine neurons of the nigrostriatal pathway by styrene exposure as has been observed with other pharmacological agents (UNGERSTED 1971). Our previous results have shown that neurotoxic chemicals like acrylamide (AGRAWAL et al. 1981b), chlordecone (SETH et al. 1981a) and manganese (SETH et al. 1981b) also alter the dopamine receptors sensitivity. Pharmacological agents like apomorphine and amphetamine, a known dopamine

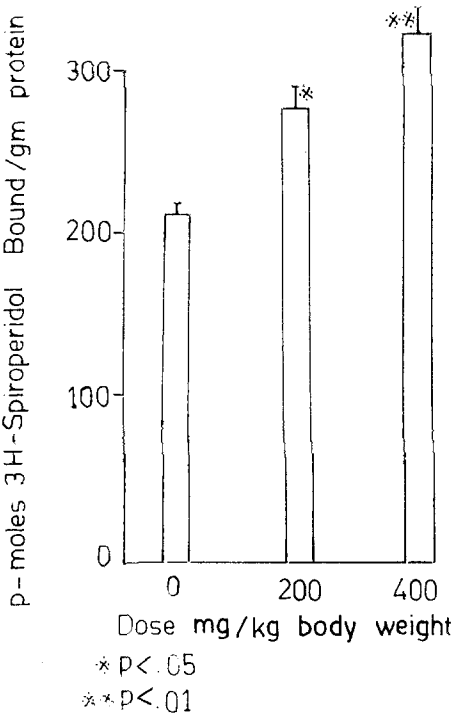

Fig. 1. effect of single exposure of styrene on ^{3}H-spiroperidol binding in rat corpus striatal membrane. Data are mean ± S.E. from 6 animals.

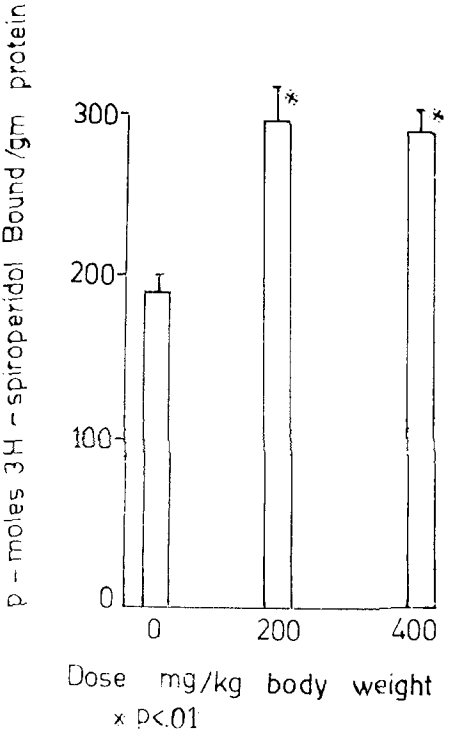

Fig. 2. Effect of repeated exposure of styrene on ^{3}H-spiroperidol binding in corpus striatal membrane of adult rats. Data are mean ± S.E. from 6 animals.

antagonist, have also been shown to alter the sensitivity of dopamine receptors (ROSENGARTEN & FRIEDHOFF 1979; MULLER & SEEMAN 1977; SCHWARTZ et al. 1978). The increase in the sensitivity of dopamine receptors in styrene exposed animals suggests the involvement of dopaminergic system in its neurotoxicity.

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