by an interaction with kappa receptors, and that EKC may reduce dopamine and serotonin release, as a result PCP induced stereotypy is antagonized. Tam, S. W. *Proc Natl Acad Sci USA* **80:** 6703–6707, 1983. Nabeshima, T. *et al. Eur J Pharmacol* **91:** 455–462, 1983. Watanabe, H. *et al. Pharmacol Biochem Behav* **14:** 494–496, 1981. Lee, A. J. *et al. Neuropharmacology* **18:** 153–158, 1978.

[^{3}H]PCP-3-OH AND [^{3}H] (+)SKF 10047 BIND TO MULTIPLE SIGMA/OPIATE PCP BINDING SITES IN RAT BRAIN. Itzhak, Y. Department of Pharmacology, Hadassah School of Medicine, Jerusalem, Israel.

Previous studies have indicated that specific binding of [^{3}H]PCP and [^{3}H] (+)SKF 10047 in rat brain membranes is associated with a common binding site for both PCP analogs and psychotomimetic opiate benzomorphans. This site was designated as *sigma* opiate/PCP receptor. It has also been reported that the antipsychotic agent, haloperidol, is a potent inhibitor of [^{3}H] (+)SKF 10047 specific binding in mammalian brain membranes. In the present study we have characterized the binding properties of one of the most potent analogs of PCP: [^{3}H]PCP-3-OH, and compared it to the binding of [^{3}H] (+)SKF 10047 in rat brain membranes. Both competition and saturation binding assays revealed that [^{3}H]PCP-3-OH labels two distinct binding sites. High affinity (kd < 1 nM) sites are potently inhibited by both psychotomimetic opiates, such as (+)SKF 10047, and PCP analogs and display pharmacological specificity similar to that for [^{3}H] (+)SKF 10047 binding sites. However, low affinity (kd=20 nM) sites are sensitive only to PCP analogs. These two sites are insensitive to haloperidol. [^{3}H] (+)SKF 10047 labels apparently not only a site which displays pharmacological specificity similar to that for high affinity [^{3}H]PCP-3-OH binding site, but also an additional site which is sensitive to haloperidol. Several lines of evidence suggest that this haloperidol sensitive site may be allosterically coupled to the high and low affinity sites labeled with [^{3}H]PCP-3-OH. The present study provides evidence for the existence of multiple subtypes of binding sites for psychotomimetic agents.

PHARMACOLOGY AND NEUROPROTECTIVE EFFECTS OF THE NMDA ANTAGONIST MK-801. Iversen, L. L., G. N. Woodruff, J. A. Kemp, A. Foster, R. Gill and E. Wong. Merck Sharp & Dohme Research Laboratories, Neuroscience Research Centre, Terlings Park, Eastwick Road, Harlow, Essex, CM20 2QR, England.

MK-801, (+)-5-methyl-10,11-dihydro-5H-dibenzo[a,d] cyclohepten-5,10-imine maleate, was described previously as a potent orally active anticonvulsant of unknown mechanism (Clineschmidt *et al.*, 1982). We found that ^{3}H-MK-801 binds to a specific population of receptor sites in rat brain, and these appear to be associated with glutamate receptors of the N-methyl-D-aspartate (NMDA) type (Wong *et al.*, 1986). Thus MK-801 antagonises the depolarising actions of NMDA in rat cerebral cortex *in vitro* and leaves responses to other selective glutamate-like agonists unchanged (quisqualic acid, kainic acid). The antagonism is non-competitive and agonist-dependent in character. We have studied the protective effect of MK-801 against neuronal degeneration caused by ischaemia in the gerbil and by injection of NMDA or quinolinic acid in the rat brain. MK-801 given 1 hr prior to bilateral carotid artery occlusion (5 min in the gerbil) significantly protected against ischaemia-induced loss of CA1 and CA2 hippocampal cells, with an ED_{50} of 0.3 mg/kg (IP), similar to its anti-convulsant potency. MK-801 remained surprisingly effective even when given after the ischaemic episode, with full neuroprotection at 2 hr, and partial efficacy with a 24 hr delay. Pretreatment of rats with 1–10 mg/kg MK-801 IP 1 hr prior to injection caused almost complete protection of neuronal degeneration caused by NMDA (hippocampus and striatum), or quinolinic acid (striatum) ($p<0.05$). MK-801 was also able to yield significant neuroprotection when administered 1–3 hr after neurotoxin injections. MK-801 also showed neuroprotective effects in other ischaemic models (rat 4-vessel, cat-middle cerebral artery). The results provide strong support for the hypothesis that NMDA receptors are involved in ischaemic neurodegeneration and suggest a therapeutic potential for MK-801 in the treatment of cerebral ischaemia. Clineschmidt, B. V., G. E. Martin and P. R. Bunting. *Drug Dev Res* **2:** 123–134, 1982. Wong, E., J. A. Kemp, T. Priestley, A. R. Knight, G. N. Woodruff and L. L. Iversen. *Proc Natl Acad Sci USA* **83:** 7104–7108, 1986.

PSYCHOPHARMACOLOGICAL PROFILE OF THE NMDA RECEPTOR ANTAGONIST MK-801. Iversen, S. D., L. Singh, R. J. Oles and M. D. Tricklebank. Merck Sharp & Dohme Research Centre, Harlow, U.K.

The non-competitive NMDA receptor antagonist, MK-801 induces a complex behavioural syndrome in the rat involving lateral head weaving, body rolling, hyperlocomotion and ataxia. Similar behaviours are seen after the ICV administration of 2-DL-amino-7-phosphonoheptanoic acid (APH), a competitive NMDA receptor antagonist, or following systemic injection of phencyclidine (PCP), ketamine and (+)-SKF 10,047, compounds having high affinity for *sigma* recognition sites, in addition to an antagonist action at NMDA receptors. In drug discrimination studies, PCP, ketamine and SKF 10,047 generalised to the introceptive cue induced by MK-801 while MK-801, ketamine, (+)-SKF 10,047 and APH (given ICV) generalised to that induced by PCP. These findings are inconsistent with the involvement of the *sigma* recognition site in the expression of the motor and discriminative stimulus properties of MK-801, since both MK-801 and APH possess negligible affinity for this site. The ability of MK-801, APH, ketamine, PCP and (+)-SKF 10,047 to block the neurophysiological actions of N-methyl-D-aspartate suggests that their overt behavioural effects are mechanisms based in the NMDA receptor, although various neurotransmitters may be involved in the full expression of these behaviors.

COMPOUNDS BASED ON 2-MDP AND DEXOXADROL WITH POTENTIAL PCP-LIKE PHARMACOLOGICAL ACTIVITY: SYNTHESIS AND RECEPTOR BINDING. Jacobson, A. E., A. Thurkauf, M. V. Mattson, K. C. Rice and J. H. Woods.* National Institute of Diabetes, Digestive and Kidney Diseases, National Institutes of Health, Bethesda, MD 20892; and *Department of Pharmacology, University of Michigan, Ann Arbor, MI 48109.

(−)-2-Methyl-3,3-diphenyl-3-propanolamine (2-MDP) has

recently been shown by Tang *et al.* (1984) to have PCP-like pharmacological activity. We have found that its affinity for the PCP receptor (as defined by [^{3}]TCP) is about the same as PCP itself. We found it intriguing that the open-chain compound could mimic, in its receptor affinity and pharmacological activity, PCP's actions. Based on our determination of the conformation of dexoxadrol which overlaps pertinent moieties in PCP (from our study of the absolute configuration of dexoxadrol [Jacobson *et al.*, 1987]), the presumed significant atoms of 2-MDP will overlap more readily with those of PCP and dexoxadrol if the molecule is modified. Thus, we synthesized a number of analogs of 2-MDP (4-hydroxy-2-methyl-4-phenylbutylamine, 4,4-diphenyl-4-hydroxy-2-methylbutylamine, 4,4-diphenyl-4-hydroxybutylamine, 4,4-diphenyl-4-hydroxy-2,N-dimethylbutylamine, 3,3-diphenyl-3-hydroxypropylamine, and 4,4-diphenyl-4-hydroxy-3-methylbutylamine. The receptor binding affinities of these compounds have been determined, and their discriminative stimulus properties are being examined. It was noted by Hardie *et al.* (1966), that N-derivatives of dexoxadrol retained some of the biological activities of dexoxadrol itself. Insofar as we were aware, these compounds had never been examined for their affinity for the PCP receptor. In order to find the biochemical effect of the molecular change from a secondary to a tertiary or quaternary nitrogen atom in dexoxadrol, we prepared the N-methyl, N-benzyl, and N-allyl dexoxadrol, as well as the N-dimethyl quaternary salt of dexoxadrol, and determined their affinity for the PCP receptor. These results, as well as those with the 2-MDP analogs, will be discussed. (A.T. supported by the National Institute on Drug Abuse through National Research Service Award No. 5F32 DAO5287-02.)

N-METHYL-D-ASPARTATE ENHANCED ^{3}H-TCP BINDING TO RAT CORTICAL MEMBRANES: EFFECTS OF DIVALENT CATIONS AND GLYCINE.

Johnson, K. M., L. D. Snell and R. S. Morter. Department of Pharmacology and Toxicology, University of Texas Medical Branch, Galveston, TX 77550.

PCP and related substances can potently and specifically antagonize excitatory amino acid depolarizations mediated by the N-methyl-D-aspartate (NMDA) receptor. Pharmacological evidence strongly suggests that the inhibition of NMDA-induced excitation by PCP is not competitive in nature and that PCP and related drugs act instead to block the open state of the NMDA-activated ion channel. In accord with this model is the report by Fagg and Baud (1986) that the binding of ^{3}H-TCP to membrane preparations rich in postsynaptic densities can be greatly enhanced by addition of exogenous L-glutamate (Glu) and that this effect is mediated by action on the NMDA receptor. We report here our own investigations of excitatory amino acid induced ^{3}H-TCP binding in rat cortical homogenates that have been lysed twice in distilled H_2O (30 min at 37°C) and washed repeatedly in 10 mM HEPES (pH 7.5). This final membrane preparation is referred to as the twice lysed P_2 (LLP). Initial studies revealed that in the crude P_2, specifically-bound TCP was 90% of total binding (2.5 nM ^{3}H-TCP, non-specific binding measured in the presence of 300 μM PCP) which was reduced up to 80% in the LLP. The addition of NMDA increased TCP binding two- to three-fold (K_d=2.5 μM), although the maximal enhancement (at 100 μM NMDA) was still below that seen in the P_2. Agonists at the other excitatory amino acid receptors, kainate or α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA), were without effect. Glu and aspartate (both at 1 μM) produced a three-fold increase in ^{3}H-TCP binding. Our most recent experiments have established that low concentrations of both $MgCl_2$ and $CaCl_2$ can enhance TCP binding in the LLP although both these cations could inhibit binding at 1 mM. The enhancement of binding by these ions may reflect the presence of low concentrations of endogenous ligand for the NMDA receptor as these effects were blocked by DL-2-amino-5-phosphonovalerate (APV). We have also found that glycine (0.1 and 1 μM) can enhance TCP binding via a mechanism that is reversible by APV but not by strychnine. These studies suggest that divalent cations and glycine enhance binding by stabilizing the NMDA channel in the open state. (Supported by DA-02073.)

REORGANIZING GLUTAMATE PATHWAYS IN THE DEVELOPING BRAIN MAY PROVIDE A SUBSTRATE FOR HYPOXIC-ISCHEMIC NEURONAL INJURY.

Johnston, M. V., F. S. Silverstein, J. Barks, R. MacDonald, A. B. Young, J. Penney and T. Greenamyre. Department of Pediatrics and Neurology, The University of Michigan, Ann Arbor, MI 48104.

Hypoxia-ischemia damages selected regions of the fetal and neonatal brain. The basal ganglia and hippocampus are especially vulnerable and significant injury is usually accompanied by damage to the hippocampus manifested by seizures. We studied the distribution of glutamate receptors in human fetal and infant brain using *in vitro* receptor autoradiography. The globus pallidus (GP), which lacks a glutamate innervation in the adult brain, is heavily endowed with glutamate receptors in the newborn. Studies in rats suggest that the caudate-putamen and the GP at 7 days of age both contain adult densities of glutamate receptors which disappear over the next 2 weeks in the GP. Autoradiography of fetal human brain shows heavy concentrations of glutamate receptors in the caudate, GP, sub-thalamic nucleus and the reticular nucleus of the thalamus by 18–24 weeks gestation. In a model of unilateral hypoxic-ischemic injury in 7 day old rats, glutamate receptors in the caudate and GP and hippocampus are markedly reduced and histologic injury correlates well with the distribution of receptors in the vulnerable structures. Pharmacologic characteristics of the glutamate receptors in the immature brain appear to be unique and dissimilar from those in adulthood. Microinjection of the glutamate analogue quisqualic acid destroys glutamate receptor bearing areas in the immature rat and replicates key features of hypoxic-ischemic brain injury. The characteristics of the immature glutamate receptors, their co-localization with TCP receptors and the neuroprotective effects of glutamate blocking compounds are subject of cur-