Other hippocampal regions and outer layers of cerebral cortex had dense binding. Thalamus, striatum and amygdala had intermediate and brainstem and hypothalamus had low binding densities. Bmax studies demonstrated 4–5 times more NMDA binding sites than TCP binding sites in all regions examined. In human hippocampus, binding for both receptors was high compared to other brain regions. In patients with Alzheimer's disease, both receptor types were decreased 40% in stratum pyramidale of CA1. It may be possible to label NMDA receptor complexes in living humans using positron-labeled dissociative anesthetics. (Supported by USPHS grants AG 06155 and NS 15655, and ADRDA and the A.C. and Ersa Arbogast Foundation.)

SYNTHESIS, RESOLUTION AND DISCRIMINATIVE STIMULUS STUDIES OF THE β-SERIES STEREOISOMERS OF 2,2-DIPHENYL-4-(2-PIPERIDYL)-1,3-DIOXOLANE. Zenk,*[1] P. C., E. L. May,* R. L. Balster* and J. V. Silverton.† *Department of Pharmacology and Toxicology, Medical College of Virginia, Virginia Commonwealth University, Richmond, VA 23298; and †National Heart, Lung, Blood Institute, National Institute of Health, Bethesda, MD 20892.

The synthesis and resolution of dioxadrol (α-series) has been reported but the resolution of the stereoisomers in the corresponding β-series has not been reported. The resolution of the β-series has now been accomplished by using (+) and (−) mandelic acid and the absolute configuration of the β-(−) stereoisomer determined by single crystal X-ray analysis. The stereoisomers in the β-series were also tested for discriminative stimulus properties in phenycyclidine trained rats. (Supported by NIDA grant: DA-01442.) [1]Current address: Department of Chemistry, College of Wooster, Wooster, OH 44691.

HUMAN ENDOGENOUS BRAIN LIGANDS FOR SIGMA AND PHENCYCLIDINE RECEPTORS. Zhang,* A.-Z., K. N. Mitchell, L. Cook and S. W. Tam. *Department of Neurobiology, Shanghai Medical University, China; and Medical Products Department, E. I. du Pont de Nemours & Co., Wilmington, DE 19898.

Sigma and phencyclidine receptors are believed to mediate psychotomimetic behaviors. Sigma and phencyclidine receptors have different regional distributions in the brains of animals. Preliminary reports have suggested the existence of sigma and PCP receptor binding activities in extracts of guinea pig brains and porcine brains, respectively. After extracts of human brains were chromatographed in Fractogel TSK HW-40S columns, peaks of activities which competed with the binding of (+)-[^{3}H]SKF 10,047 and [^{3}H]TCP for sigma and PCP receptors, respectively, were detected. Purification of these endogenous ligands are in progress.

PURIFICATION OF AN ENDOGENOUS LIGAND FOR THE PCP/SIGMA RECEPTOR. Zukin, R. S., W. Vale, J. Rivier and S. R. Zukin. Department of Neuroscience, Albert Einstein College of Medicine, Bronx, NY 10461.

We have identified an endogenous substance in brain which potently inhibits the binding to PCP receptors of the highly specific and potent PCP derivative N-(1-[2-thienyl]-cyclohexyl) piperidine (TCP) and which modulates N-methyl-D-aspartate (NMDA)-induced neurotransmitter release, as does PCP. An extract enriched in peptides was prepared from bovine hippocampus, the brain region of highest *sigma*/PCP receptor density. The resulting extract was purified by ODS cartridge extraction, and then applied to a preparative HPLC column (C-18, 5×30 cm). A single major peak of activity in the radioreceptor assay was observed. That the endogenous material was biological PCP-like agonist activity was indicated by findings from four transmitter release paradigms. The HPLC fractions active in the [^{3}H]TCP binding assay were found to elicit the following actions in rat striatal slices: (1) stimulation of spontaneous acetylcholine efflux; (2) inhibition of NMDA-stimulated dopamine release; (3) inhibition of NMDA-stimulated acetylcholine release; and (4) stimulation of baseline dopamine release. The latter two effects appear most sensitive and selective for the endogenous factor(s) identified by the radioreceptor assay. With respect to NMDA-stimulated transmitter release, the actions of the endogenous factor mimic those of PCP and drugs judged to be PCP-like in the drug discriminative stimulus paradigm. The finding that PCP-like drugs, including the related arylcycloalkylamines, *sigma* opioids, and dioxolanes, modulate the excitatory effects of the excitatory amino acid NMDA in a rank order of potency paralleling that for their PCP-like behavioral effects implies that this system constitutes the first extremely sensitive and relatively specific *in vitro* bioassay to assess PCP agonist activity. The sensitivity of the radioreceptor assay to high ionic strength and to acid pH renders it vulnerable to interference from ionic and pH characteristics of typical HPLC solvent systems. By contrast, the bioassay reported here is relatively insensitive to such effects, in part because of the substantial buffering capacity of the incubation medium. To our knowledge, no neuropeptide has been isolated by a radioreceptor assay alone. The transmitter release assay provides validation of a PCP-like physiological activity exerted by bovine hippocampal extracts partially purified by HPLC. Together these assay methods should prove particu-

larly useful in the further purification of the endogenous ligand of the PCP receptor. Such a ligand may serve as the endogenous modulator of NMDA-receptor mediated mechanisms in the limbic system.

NON-COMPETITIVE REGULATION OF PHENCYCLIDINE/σ RECEPTORS BY THE N-METHYL-D-ASPARTATE RECEPTOR ANTAGONIST D(−)2-AMINO-5-PHOSPHONOVALERIC ACID. Zukin, S. R., D. Javitt and R. Sircar. Departments of Psychiatry and Neuroscience and Bronx Psychiatric Center, Albert Einstein College of Medicine/Montefiore Medical Center, Bronx, NY 10461.

Electrophysiological and neurochemical studies have established a noncompetitive inhibition of N-methyl D-aspartate-type excitatory amino acid (EAA) receptor mediated CNS phenomena by PCP-like drugs. Anatomical studies have demonstrated similar distribution patterns for limbic PCP/σ receptors and NMDA receptors. These findings suggest a model in which the NMDA and PCP/σ receptors may be separate protein entities within a supramolecular complex. In order to determine the biochemical basis of the interaction between NMDA and PCP/σ receptors, specific binding of [^{3}H]TCP ([2-thienyl]-cyclohexylpiperidine) was determined in the presence of varying concentrations of EAA agonists and of the specific direct NMDA antagonist D(−)2-amino-5-phosphono-valeric acid (D(−)AP5). D(−)AP5 dose-dependently decreased the apparent B_{max} of [^{3}H]TCP binding without affecting apparent K_D. In D(−)AP5-treated membranes, pre-incubation with glutamate at doses of up to 250 μM partially reversed the D(−)AP5-induced decrement in apparent B_{max} of [^{3}H]TCP binding without affecting apparent K_D. D(−)AP5 maximally inhibited [^{3}H]TCP binding >95%. A Hill coefficient of -1.04 ± 0.23 calculated from this binding data suggested that D(−)AP5 mediates its inhibitory effects on [^{3}H]TCP binding via interaction at a single (NMDA) receptor. The IC_{50} of D(−)AP5 for inhibiting [^{3}H]TCP binding is comparable to its IC_{50} in electrophysiological paradigms. Following osmotic lysis, freezing for 48 hr and thawing of the crude membrane pellet the IC_{50} of inhibition of [^{3}H]TCP binding by D(−)AP5 decreased significantly. The rank order of a series of amino acid receptor antagonists for displacement of 10 nM [^{3}H]TCP were D(−)AP5 > D-α-aminoadipate > 1-glutamyl diethylester, corresponding to their known rank order of potency as direct NMDA receptor antagonists. These data support the hypothesis that D(−)AP5 exerts its effects on [^{3}H]TCP binding via a specific interaction with NMDA receptors. This study provides the first evidence from a receptor-binding paradigm for a non-competitive interaction between PCP/σ and NMDA receptors. The ability of D(−)AP5, a selective NMDA receptor antagonist, to abolish [^{3}H]TCP binding is consistent with the hypothesis that PCP/σ receptors may be solely localized to an NMDA receptor complex. The ability of [^{3}H]TCP to bind to an activated NMDA receptor complex suggests that TCP might be a useful probe of NMDA receptor functioning *in vitro*. The PCP receptor-endogenous ligand-NMDA receptor-ion channel complex appears to constitute an important functional CNS unit.