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Dementia of the Alzheimer's Type: Changes in Hippocampal L-[^{3}H]Glutamate Binding

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Abstract: Glutamate or a related excitatory amino acid is thought to be the major excitatory neurotransmitter of hippocampal afferents, intrinsic neurons, and efferents. We have used an autoradiographic technique to investigate the status of excitatory amino acid receptors in the hippocampal formation of patients dying with dementia of the Alzheimer type (DAT). We examined L-[^{3}H]glutamate binding to sections from the hippocampal formation of six patients dying of DAT and six patients without DAT and found marked reductions in total [^{3}H]glutamate binding in all regions of hippocampus and adjacent parahippocampal cortex in DAT brains as compared to controls. When subtypes of excitatory amino acid receptors were assayed, it was found that binding to the *N*-methyl-D-aspartate (NMDA)-sensitive receptor was reduced by 75–87%, with the greatest loss found in stratum moleculare and stratum pyramidale of CA1. Binding to quisqualate (QA)-sensitive receptors was reduced by 45–69%. There were smaller reductions (21–46%) in $GABA_A$ receptors in DAT cases. Muscarinic cholinergic receptors assayed in adjacent sections of hippocampal formation were unchanged in DAT. Benzodiazepine receptors were reduced significantly only in parahippocampal cortex by 44%. These results suggest that glutamatergic neurotransmission within the hippocampal formation is likely to be severely impaired in Alzheimer's disease. Such impairment may account for some of the cognitive decline and memory deficits that characterize DAT. **Key Words:** Alzheimer's disease—Glutamate—*N*-Methyl-D-aspartate—Quisqualate—Excitotoxins. **Greenamyre J. T. et al.** Dementia of the Alzheimer's type: Changes in hippocampal L-[^{3}H]glutamate binding. *J. Neurochem.* **48,** 543–551 (1987).

Dementia of the Alzheimer's type (DAT) is a relatively common disease affecting 5–15% of persons over age 65 (McKhann et al., 1984; Khachaturian, 1985). The classic neuropathological changes, neurofibrillary tangles and senile plaques in the cerebral cortex and hippocampus, are well known (Alzheimer, 1907; Terry and Katzman, 1983). Recent neurochemical studies of DAT have revealed decreases in cortical markers for a variety of neurotransmitter systems (for a review, see Wurtman et al., 1984). The relative importance of these changes in the pathogenesis and symptomatology of this disease is unknown.

Glutamate is believed to be a major excitatory neurotransmitter of the mammalian cerebral cortex and hippocampal formation (Storm-Mathisen, 1977; White et al., 1977; Hicks and Geddes, 1981; Baughman and Gilbert, 1981). To date, there have been relatively few studies of the glutamatergic system in DAT. Examination of this excitatory amino acid in DAT is important because of recent suggestions that glutamate and its receptors may have a role in learning and memory (Collingridge, 1985; Morris et al., 1986). Although free glutamate levels, which are a measure of both transmitter and metabolic glutamate pools, have been reported as reduced (Arai et al., 1985; Hyman et al., 1986) or normal (Antuono et al., 1984; Perry et al., 1984) in cortex of DAT brains, Smith et al. (1985) have demonstrated a correlation between CSF glutamate levels and cognitive test scores in DAT patients.

We and others have recently developed autoradiographic techniques to characterize the subtypes of glutamate receptors (Monaghan et al., 1983; Greenamyre et al., 1984, 1985*a*; Halpain et al., 1984). We have reported a relatively selective loss of *N*-methyl-D-aspartate (NMDA) sites in cerebral cortex of patients dying of DAT (Greenamyre et al., 1985*b*). To assess

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Abbreviations used: DAT, dementia of the Alzheimer type; GABA, γ-aminobutyric acid; NMDA, *N*-methyl-D-aspartate; QA, quisqualate; QNB, quinuclidinyl benzilate.

further the role of the glutamatergic system in DAT, we have used the autoradiographic technique to measure NMDA- and quisqualate (QA)-sensitive receptors in the hippocampal formation in DAT.

MATERIALS AND METHODS

Tissue

The brains from five neurologically normal patients and one Huntington's disease patient (controls) and six patients who died with DAT were removed and one hemisphere from each brain was immediately frozen and stored at −70°C; the other hemisphere from each brain was preserved in formalin and examined for neurofibrillary tangles and senile plaques. The age of the control patients was 62 ± 3 (SEM) years and that of the DAT patients was 67 ± 3 years. Postmortem delays were 16 ± 3 h for the control brains and 19 ± 4 h for the DAT brains. Brains with more than five tangles or plaques per high-powered (400×) field in cerebral cortex were considered to have DAT.

The frozen hemispheres were cut into 2-cm thick coronal slabs. The hippocampus and adjacent cortex were removed en bloc from the slab containing the lateral geniculate body. Starting at the lateral geniculate and sectioning caudally toward the septal pole, serial 20-μm frozen sections of the hippocampal formation and adjacent cortex were cut from this block, stored for less than 24 h at −20°C, and then assayed for glutamate binding and other receptors as described below. For both control and DAT brains, there was some variability in the rostro-caudal level examined using a given assay. However, similar rostro-caudal extents for each assay were represented in both DAT and control brains.

Receptor assays

A detailed description of the autoradiographic glutamate binding assay has been published (Greenamyre et al., 1984, 1985*a*,*b*). In brief, in glutamate saturation studies, tissue sections were incubated with 30 n*M* L-[^{3}H]glutamate and nine concentrations of unlabeled glutamate ranging from 25 n*M* to 1 μ*M* in 50 m*M* Tris-HCl containing 2.5 m*M* $CaCl_2$ for 45 min at 2°C. Nonspecific binding was determined in the presence of 1 m*M* unlabeled glutamate and represented <10% of total binding. Similar "blanks" were obtained with 1 m*M* QA or 2.5 μ*M* QA plus 100 μ*M* NMDA. Detailed competition studies were carried out in the presence of 200 n*M* L-[^{3}H]glutamate (sp act, 4.5 Ci/mmol) using 16–23 QA concentrations ranging from 1 n*M* to 1 m*M*.

Previous studies have shown that glutamate binds to at least two receptor classes with very similar affinities and that saturation isotherms of glutamate binding yield linear Scatchard plots. Hill plots of the binding data reveal a Hill number of one. In contrast, QA displaces all specific glutamate binding biphasically with K_i values for the high-affinity and low-affinity sites that differ by 1,000-fold (Greenamyre et al., 1984, 1985*a*). The sites with a low affinity for QA are equivalent to the NMDA sites (Greenamyre et al., 1985*a*). Since the K_i values for the two QA-sensitive sites are separated by three orders of magnitude, binding determined in the presence of 2.5 μ*M* QA can be used to estimate quite accurately the number of each binding site at a given [^{3}H]glutamate concentration. Thus at 2.5 μ*M* QA, 96% of binding to the high-affinity QA-sensitive site is blocked, but 98% of binding to the low-affinity QA-sensitive site remains. QA receptors were defined, therefore, by their high affinity for QA in detailed competition curves using the LIGAND program to resolve the sites as described previously by Munson and Rodbard (1980) and Greenamyre et al. (1984, 1985*a*). Similar results were obtained by defining QA sites as the specific glutamate binding sites remaining in the presence of 100 μ*M* NMDA. In analogous fashion, NMDA receptors were defined as either those glutamate binding sites having a low affinity for QA in detailed competition curves or those sites remaining in the presence of 2.5 μ*M* QA. There was no specific glutamate binding in the presence of 2.5 μ*M* QA plus 100 μ*M* NMDA. Under assay conditions, in the presence of 2.5 m*M* $CaCl_2$, there was no binding of [^{3}H]glutamate to kainate-sensitive sites (Monaghan et al., 1983; Greenamyre et al., 1985). After the incubation, sections were quickly rinsed three times with cold buffer, rinsed once with cold 2.5% glutaraldehyde in acetone, then blown dry with warm air. The total rinse time was <10 s.

Adjacent sections of the same brains were assayed for $GABA_A$, benzodiazepine, and muscarinic cholinergic receptors using [^{3}H]muscimol (5–150 n*M*), [^{3}H]flunitrazepam (5–50 n*M*), and [^{3}H]quinuclidinyl benzilate ([^{3}H]-QNB) (1 n*M*), respectively. These assays were carried out as described previously by Penney and Young (1982, 1984) and by Walker et al. (1984).

Quantitative autoradiography

After the assays, slides were placed in x-ray cassettes along with appropriate standards and exposed to Ultrofilm-^{3}H (LKB) for 10 days ([^{3}H]flunitrazepam and [^{3}H]QNB), 16 days ([^{3}H]glutamate), or 21 days ([^{3}H]muscimol). Following exposure, the films were developed and a spot densitometer (Dauth et al., 1984) was used as described previously by Pan et al. (1983) to measure radioactivity in stratum moleculare and stratum pyramidale of the CA1 region of hippocampus, the stratum moleculare of the dentate gyrus, stratum moleculare and stratum lucidum of CA3, and the isocortex of the parahippocampal gyrus. At least 16 densitometer readings were taken from each of multiple areas of interest on the autoradiographic image. Twelve sections were analyzed from each glutamate saturation curve and 16–23 sections were analyzed from QA competition curves.

The numbers and affinities of glutamate, $GABA_A$, and benzodiazepine receptors were calculated using Scatchard analysis since previous studies have shown that glutamate, muscimol, and flunitrazepam all produce monophasic, linear Scatchard plots with Hill coefficients near unity (Pan et al., 1983, 1984; Greenamyre et al., 1984, 1985*a*). Subtypes of glutamate receptors were determined as described above. The number of muscarinic cholinergic receptors equalled the amount of [^{3}H]QNB specifically bound, because a saturating concentration of [^{3}H]QNB was used. Binding to each region of control and DAT brain was compared using parametric (Student's *t* test) and nonparametric (Mann-Whitney) tests.

RESULTS

Glutamate binding in the hippocampal formation of control brains had a laminar distribution (Fig. 1). The pattern of lamination is somewhat different in humans than it is in rodents, corresponding to differences in the anatomy of the CA1 region. In rodents the CA1 pyramidal cells form a single layer of cells

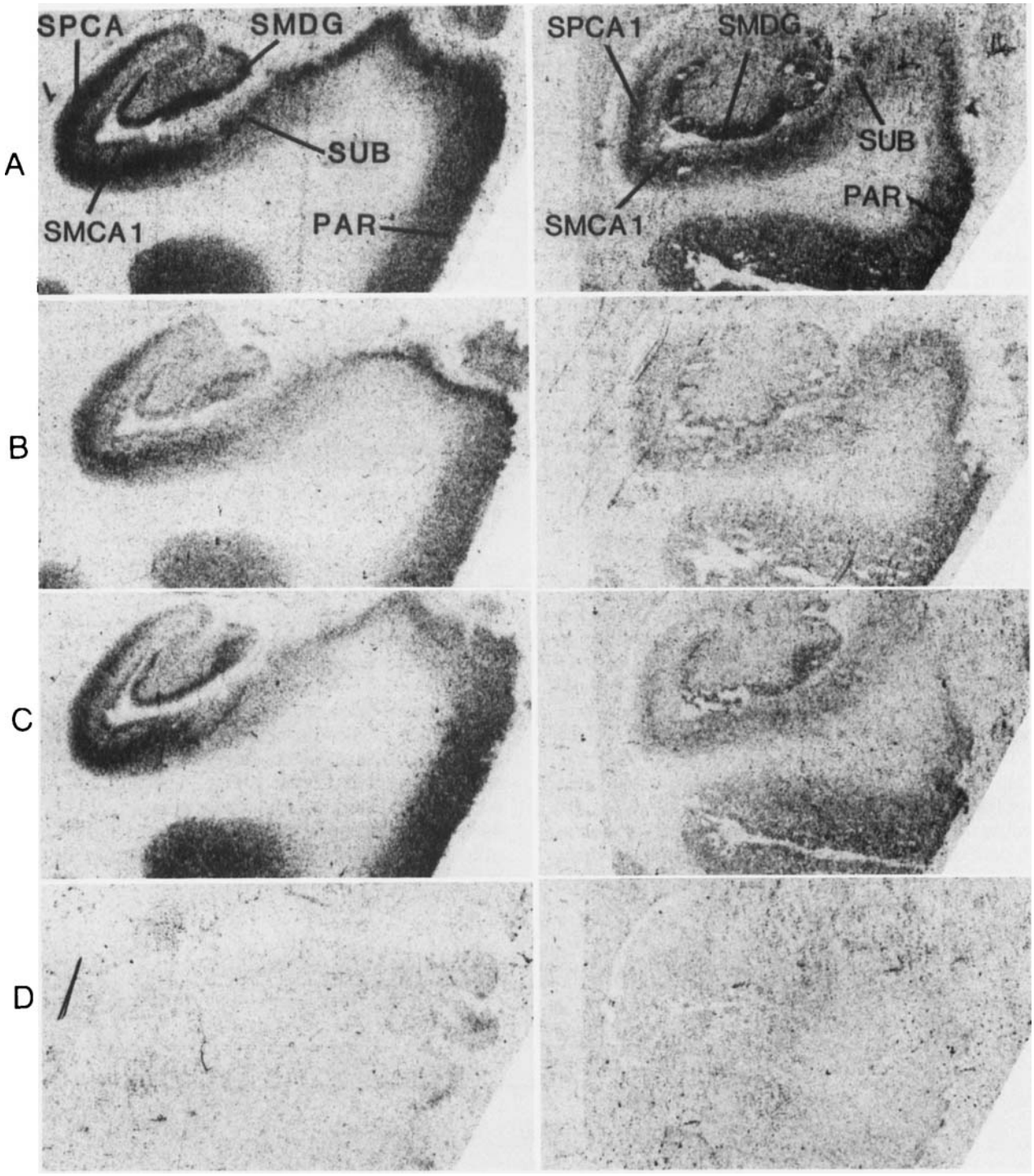

FIG. 1. Comparison of glutamate binding in hippocampal formation and adjacent cortex of control (**left**) and DAT (**right**) brains. The concentration of [^{3}H]glutamate was 200 n*M* (4.5 Ci/mmol). **A:** Total glutamate binding. **B:** NMDA receptors (glutamate binding in the presence of 2.5 μM QA). **C:** QA receptors (glutamate binding in the presence of 100 μM NMDA). **D:** Nonspecific binding (glutamate binding in the presence of 2.5 μM QA plus 100 μM NMDA). Sections from control and DAT brains were cut and incubated under the same conditions as described under Materials and Methods. Each piece of Ultrofilm ^{3}H was exposed to tissue from a control and a DAT brain. Photographic processing of the resulting autoradiographs was identical. PAR, parahippocampal cortex; SMCA1, stratum moleculare of CA1; SMDG, stratum moleculare of dentate gyrus; SPCA1, stratum pyramidale of CA1; SUB, subiculum. The region of low binding between the strata molecularia is the hippocampal fissure which has opened artifactually in these frozen, postmortem human samples.

with distinct layers of basal dendrites (stratum oriens) and apical dendrites (strata radiatum, lacunosum, and moleculare; Lorente de No, 1934). In primates, the CA1 pyramidal cells are a more diffuse band so that basal dendrites of one neuron may be next to apical dendrites or somas of other neurons (Lorente de No, 1934). In the CA1 region of human brain there was a broad band of dense binding corresponding to strata oriens, pyramidale, and proximal stratum radiatum. The densest binding in this band was to stratum pyramidale. In CA1, there was also a thin layer of dense binding immediately adjacent to the hippocampal fissure. This layer appeared to correspond to the stratum moleculare as defined in Golgi and pathway tracing experiments (Lorente de No, 1934; Van Hoesen and Pandya, 1975). Binding was also dense in the outer two-thirds of the stratum moleculare of the dentate gyrus (Table 1). Binding in stratum lucidum and stratum pyramidale of CA3 was 20% and 25%, respectively, of that in stratum pyramidale of CA1. There was similar low binding in the hilum of the dentate gyrus. In subiculum, there was heavy glutamate binding in a thin band that corresponded to the pyramidal cell layer.

In DAT brains, glutamate binding was significantly reduced in all areas of the hippocampal formation measured (Figs. 1 and 2). The reductions in binding in stratum pyramidale and stratum moleculare of CA1 were more profound than those in dentate gyrus or in adjacent parahippocampal cortex. The dense layer of binding found in normal subiculum was never seen in DAT cases. In most cases binding was so low that the pyramidal layer could not reliably be distinguished from background. The large reductions in glutamate binding made it impossible to obtain accurate saturation data on all but three DAT brains. In those brains where saturation data could be quantitated, the reduction in glutamate binding appeared to be due to a decrease in the number of binding sites and not caused by a reduced affinity of binding (Table 2).

Examination of excitatory amino acid receptor subtypes showed differential changes in NMDA and QA receptors in various hippocampal and cortical regions (Fig. 2). In control brains, QA competition studies indicated that QA-sensitive sites were highest in stratum moleculare of dentate gyrus (Table 3). In the other regions, QA-sensitive and NMDA-sensitive sites were present in nearly equal concentrations. The affinities of the two sites for QA differed by approximately three orders of magnitude. In DAT hippocampi, the reductions in binding were so profound that accurate affinity constants and B_{max} values for the receptor subtypes could not be estimated using the LIGAND program. Only the numbers of QA-sensitive and NMDA-sensitive sites present at 200 n*M* [^{3}H]glutamate could be accurately obtained (Fig. 2). QA-sensitive binding was reduced by 45–69% in various regions of the hippocampal formation. Stratum moleculare of dentate gyrus had a 45% reduction in QA-sensitive sites whereas stratum pyramidale and stratum moleculare of CA1 had 68 and 69% reductions, respectively ($p < 0.05$). NMDA receptors were reduced by at least 75% in all areas examined. In stratum pyramidale and stratum moleculare of CA1, NMDA receptors were reduced by 84 and 87%, respectively ($p < 0.05$ and $p < 0.01$).

Binding of other ligands to adjacent sections of hippocampus revealed no significant changes in benzodiazepine or muscarinic cholinergic receptors in hippocampal formation (Table 2; Fig. 3). In parahippocampal cortex, benzodiazepine binding was reduced by 44%. Benzodiazepine receptors were not decreased in other areas of cerebral cortex, as reported previously by Greenamyre et al. (1985*b*).

TABLE 1. *[^{3}H]Glutamate binding to various areas of control and DAT hippocampus*

Area	[^{3}H]Glutamate bound[a] (pmol/mg protein)		Percent decrease	p value[b]	
	Control	DAT		Parametric	Nonparametric
Stratum moleculare (dentate gyrus)	2.75 ± 0.70	0.91 ± 0.20	60	0.10	0.10
Stratum lucidum (CA3)	0.61 ± 0.20	0.23 ± 0.04	62	0.10	NS
Stratum pyramidale (CA3)	0.76 ± 0.24	0.27 ± 0.05	65	0.07	0.1
Statum moleculare (CA1)	2.67 ± 0.68	0.55 ± 0.09	79	0.01	0.01
Stratum pyramidale (CA1)	2.83 ± 0.75	0.68 ± 0.16	76	0.02	0.01
Parahippocampal cortex	2.39 ± 0.49	0.63 ± 0.15	74	0.006	0.01

Values represent means ± SEM.
[a] Glutamate binding was carried out at 200 n*M*.
[b] Comparisons were made with parametric (Student's *t* test) and nonparametric (Mann-Whitney) tests.

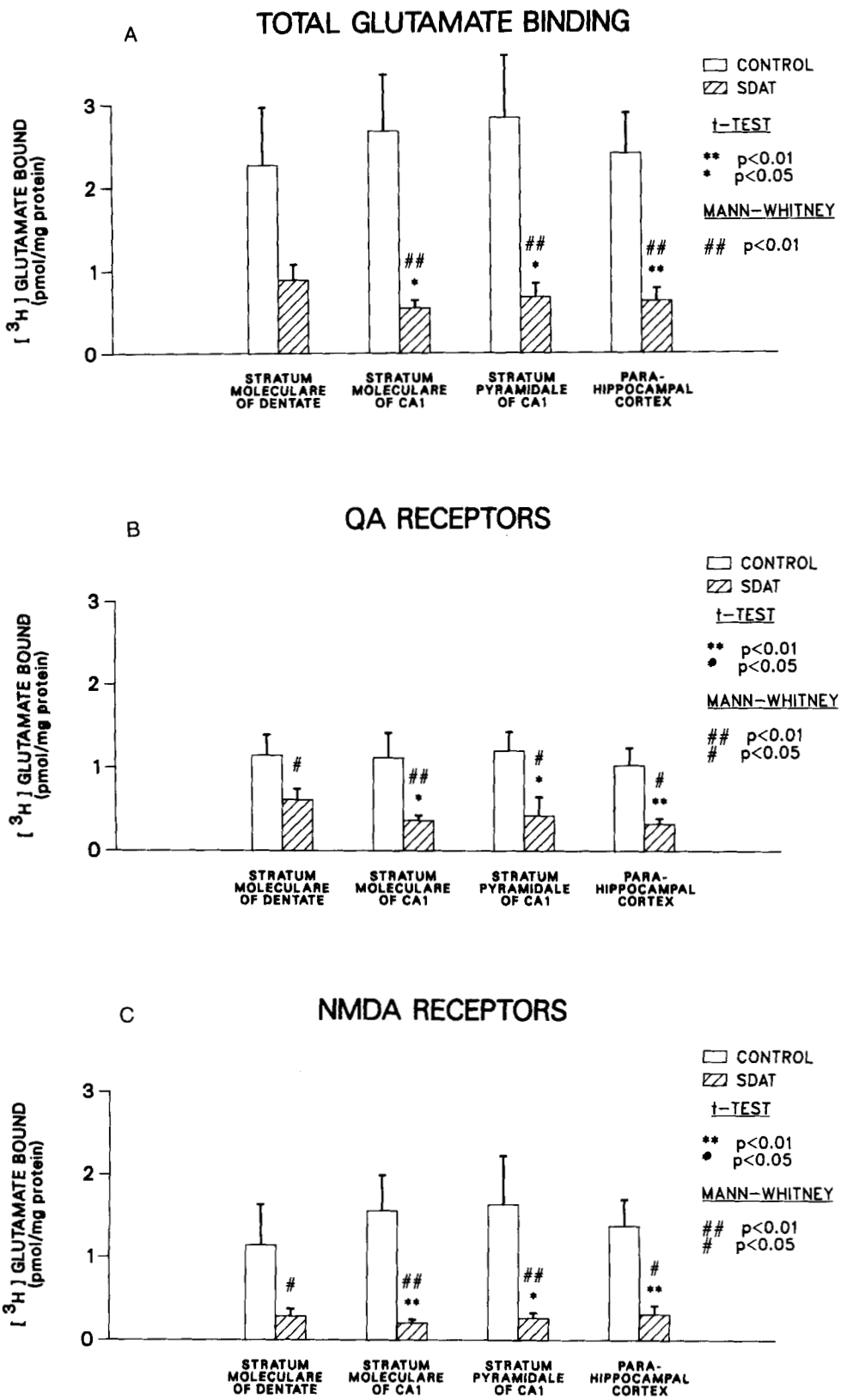

FIG. 2. Specific glutamate binding (at 200 n*M* [^{3}H]glutamate) in various regions of the hippocampal formation of control and DAT brains (means ± SEM; n = 6 for each group). **A:** Total glutamate binding. **B:** NMDA-sensitive receptors. **C:** QA-sensitive receptors. Receptor types were defined as described under Materials and Methods. Level of significance was determined by unpaired, two-tailed *t* test and by a nonparametric test, the Mann-Whitney.

TABLE 2. *$GABA_A$, benzodiazepine, and muscarinic cholinergic receptor binding in hippocampus of DAT and control brains*

Region and ligand	Control		DAT	
	B_{max} (pmol/mg protein)	K_D (nM)	B_{max} (pmol/mg protein)	K_D (nM)
[^{3}H]Muscimol				
Stratum moleculare (dentate gyrus)	4.1 ± 0.9	73 ± 8	3.0 ± 0.4	44 ± 10
Stratum moleculare (CA1)	3.4 ± 1.0	76 ± 28	2.1 ± 0.3	59 ± 10
Stratum pyramidale (CA1)	3.9 ± 1.0	66 ± 10	2.5 ± 0.3	61 ± 10
Parahippocampal cortex	4.4 ± 0.8	72 ± 8	2.7 ± 0.1	50 ± 5
[^{3}H]Flunitrazepam				
Stratum moleculare (dentate gyrus)	1.6 ± 0.2	7.0 ± 0.8	1.8 ± 0.3	3.4 ± 0.1
Stratum moleculare (CA1)	1.2 ± 0.2	7.5 ± 1.3	1.3 ± 0.1	4.1 ± 1.7
Stratum pyramidale (CA1)	1.3 ± 0.2	5.7 ± 1.1	1.2 ± 0.1	4.1 ± 0.5
Parahippocampal cortex	1.6 ± 0.1	10.0 ± 2.5	0.9 ± 0.2[a]	5.3 ± 2.1
[^{3}H]QNB				
Stratum moleculare (dentate gyrus)	1.3 ± 0.2	ND	1.4 ± 0.1	ND
Stratum moleculare (CA1)	0.8 ± 0.2	ND	0.9 ± 0.2	ND
Stratum pyramidale (CA1)	1.7 ± 0.3	ND	1.3 ± 0.2	ND
Parahippocampal cortex	1.6 ± 0.4	ND	2.0 ± 0.3	ND
[^{3}H]Glutamate				
Stratum moleculare (dentate gyrus)	18 ± 6	990 ± 100	4.1 ± 0.7	830 ± 130
Stratum moleculare (CA1)	20 ± 11	1,150 ± 470	3.8 ± 1.1	1,480 ± 190
Stratum pyramidale (CA1)	12 ± 2	810 ± 140	4.9 ± 1.4	830 ± 160
Parahippocampal cortex	14 ± 2	1,150 ± 210	5.3 ± 1.0	1,190 ± 240

Values are means ± SEM from six controls and six DAT brains except for [^{3}H]glutamate values which are from five control and three DAT brains.

[a] $p < 0.05$.

TABLE 3. *Binding values for QA- and NMDA-sensitive sites in human control hippocampus*

Region	K_H (nM)	K_L (μM)	R_H (pmol/mg protein)	R_L (pmol/mg protein)	n
Stratum moleculare (dentate gyrus)	32 ± 22	48 ± 17	14 ± 2	8 ± 1	4
Stratum moleculare (CA1)	98 ± 32	68 ± 11	9 ± 2	8 ± 2	5
Stratum pyramidale (CA1)	171 ± 73	59 ± 10	9 ± 1	10 ± 3	5
Parahippocampal cortex	55 ± 31	84 ± 28	9 ± 1	8 ± 1	5

Data were obtained from detailed QA displacement studies using the LIGAND program and saturation data. QA-sensitive sites were defined as the high-affinity QA sites; NMDA-sensitive sites represented the low-affinity QA sites.

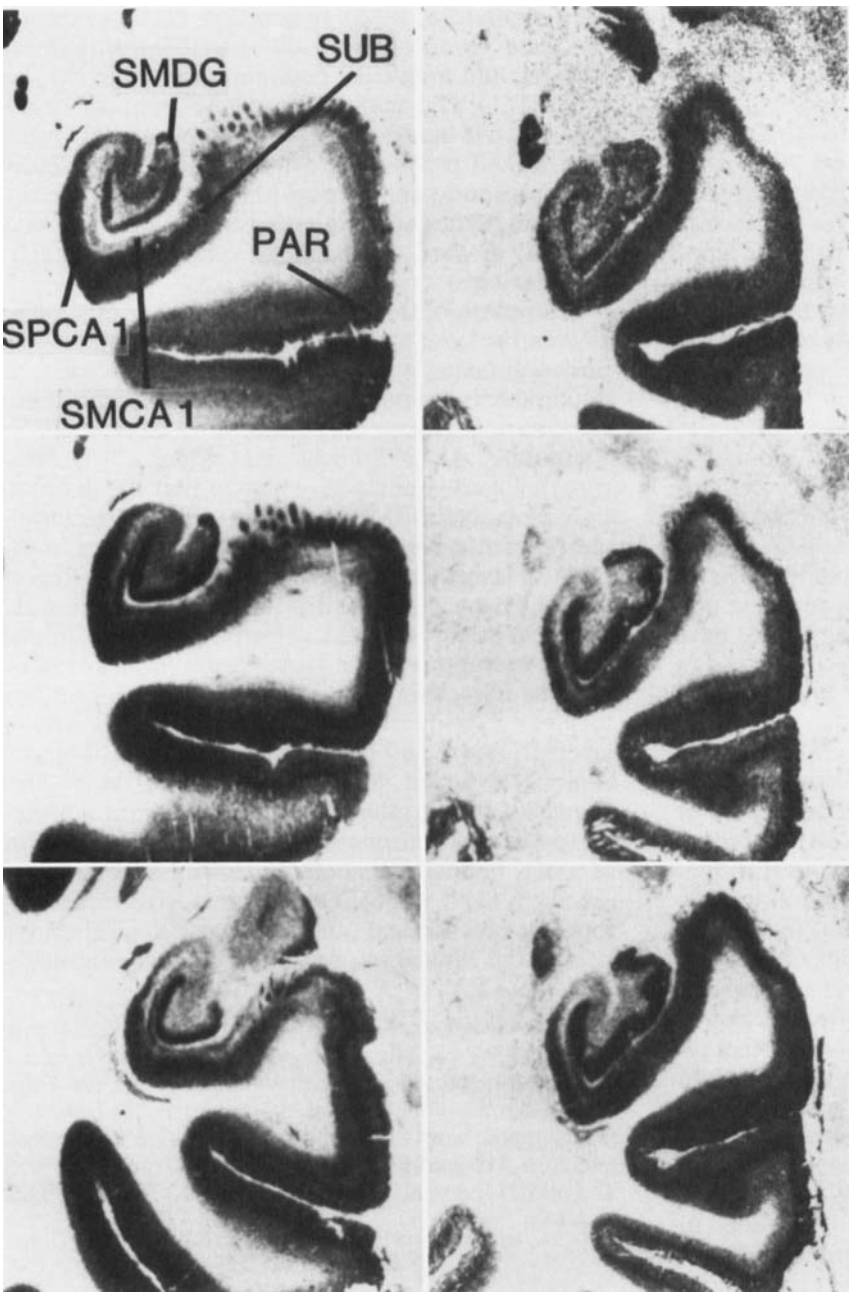

FIG. 3. Autoradiographs of [^{3}H]QNB binding to muscarinic receptors (**top**), [^{3}H]flunitrazepam binding to benzodiazepine receptors (**middle**), and [^{3}H]-muscimol binding to $GABA_A$ receptors (**lower**) in control (**left**) and DAT brains (**right**). Only binding to benzodiazepine receptors in parahippocampal cortex was significantly reduced in DAT (see Table 1). Abbreviations as in Fig. 1.

DISCUSSION

Glutamate exerts its excitatory effects through at least three distinct receptor types defined electrophysiologically (Watkins and Evans, 1981). These receptors are named for NMDA, QA, and kainate, the most specific agonists at each of these receptors. We have previously demonstrated distinct populations of glutamate binding sites that are sensitive to NMDA, QA, and kainate and that appear to correspond to the physiologically defined NMDA, QA, and kainate receptors (Greenamyre et al., 1985*a*). There is strong evidence to suggest that the NMDA-sensitive sites represent the NMDA receptors; these sites have a pharmacological profile and anatomical distribution expected of the NMDA receptor. Definitive identification of the QA-sensitive site as the physiologically relevant QA receptor awaits development of specific drugs to act at this receptor. However, it has been shown that the QA-sensitive site is a neuronal site with a distribution consistent with the physiological receptor (Greenamyre et al., 1984, and 1985*a;* Olson et al., 1985).

In the present study, we have investigated NMDA- and QA-sensitive glutamate binding sites in the hippocampus of patients dying with DAT as compared

to age-matched controls. Under the assay conditions used, there was no binding to kainate-sensitive sites. The reduction in glutamate binding in hippocampi of DAT patients was accounted for by losses of both NMDA- and QA-sensitive binding sites; NMDA-sensitive receptors were reduced by almost 90% in the stratum moleculare of CA1. This profound loss of NMDA- and QA-sensitive receptors was not accompanied by similar reductions in other receptor types. Muscarinic cholinergic receptors were unchanged in the hippocampus of DAT patients and benzodiazepine receptors were reduced significantly only in parahippocampal cortex, a site of severe pathology in DAT (Hyman et al., 1984). The decreases in $GABA_A$ receptors are statistically nonsignificant and are relatively modest (30%) compared to the large reductions in NMDA-sensitive and QA-sensitive receptors. Furthermore, there is evidence that the third class of glutamate receptor, the kainate receptor, is not decreased in the stratum moleculare of the dentate gyrus in DAT, despite large changes in NMDA-sensitive and QA-sensitive binding sites. Geddes et al. (1985) have reported an expansion of the kainate receptor field in the stratum moleculare of the dentate gyrus in DAT brains, perhaps in response to the deafferentation of this region described by Hyman et al. (1984). It is also unlikely that the loss of NMDA-sensitive and QA-sensitive receptors in DAT is merely a reflection of chronic neurological disease since cortical and hippocampal glutamate binding are normal in Huntington's disease (Greenamyre et al., 1985*b* and unpublished data). Moreover, glutamate binding in caudate, putamen, claustrum, and nucleus basalis of Meynert is unchanged in DAT (Greenamyre et al., 1985*b*). One possible explanation for the differential loss of QA- and NMDA-sensitive sites compared to other receptors is that cortical efferents to hippocampus and CA3 efferents to CA1 are thought to synapse on the dendritic spines of the hippocampal neurons and not on proximal dendritic shafts or cell somata (Lorente de No, 1934; Hjorth-Simonsen, 1973; Van Hoesen and Pandya, 1975; Swanson et al., 1978). Since in DAT, these neurons lose their distal spines and dendrites (Mehraein et al., 1975; Mann et al., 1986), receptors on these areas of the neurons would be preferentially lost.

Evidence suggests that NMDA-sensitive and QA-sensitive receptors are important in synaptic transmission in neocortex and hippocampus (Crunelli et al., 1982; Coan and Collingridge, 1985; Thomson et al., 1985) and it is well known that the hippocampus plays an important role in learning and memory (Victor et al., 1961; Horel, 1978). Therefore, it might be expected that abnormalities in glutamatergic transmission would disrupt hippocampal function, and hence impair new memory formation. In support of this hypothesis, it has been shown that blockade of hippocampal NMDA receptors prevents the development of long-term potentiation, a model system used widely for studying learning and memory (for a review see Collingridge, 1985). In addition, NMDA receptor blockade reversibly impairs spatial discrimination learning and long-term potentiation in vivo (Morris et al., 1986). The marked loss of NMDA-sensitive and QA-sensitive binding sites in the hippocampal formation in DAT reported here makes it likely that neurotransmission to and through the hippocampus is impaired in Alzheimer's patients. Such impairment may explain, in part, the memory deficits exhibited by these patients.

Disruption of the glutamatergic system might also explain the symptoms of cortical disconnection—agnosias, apraxias, and aphasia—seen in DAT patients. Glutamate is the putative transmitter of corticocortical association pathways (Clark and Collins, 1976; Fonnum et al., 1981; Hicks and Geddes, 1981). Neuropathological studies have shown that the distribution of plaques in DAT correlates well with the termination of corticocortical association pathways in superficial layers and layers V and VI of cortex (Rogers and Morrison, 1985; Pearson et al., 1985; Esiri et al., 1986). We demonstrated a relatively selective loss of NMDA receptors in the same cortical layers of DAT patients (Greenamyre et al., 1985*b*). Neurofibrillary tangles are primarily localized in pyramidal cells of layers III and V and in CA1 and subiculum of hippocampal formation. Recent studies in our laboratory have shown that many glutamate-like immunoreactive pyramidal neurons in the hippocampal formation of DAT brains also contain neurofibrillary tangles (Maragos et al., 1986). Thus, there appear to be anatomical, biochemical, and neuropathological correlates to the clinical impression of cortical disconnection in DAT.

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