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# Alterations in angiotensin $AT_1$ and $AT_2$ receptor subtype levels in brain regions from patients with neurodegenerative disorders

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**Abstract**

The present studies assessed the levels of [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II-labelled angiotensin $AT_1$ and $AT_2$ receptor recognition sites in homogenates of various brain areas (including caudate nucleus, putamen, substantia nigra, hippocampus, frontal cortex, temporal cortex and cerebellum) from patients with clinically diagnosed Parkinson's disease, Huntington's disease and Alzheimer's disease and those from age-, sex- and post-mortem delay-matched neurologically and psychiatrically normal patients. Radiolabelled angiotensin $AT_1$ receptor recognition site levels were significantly decreased by approximately 70%, 70% and 90% in the caudate nucleus, putamen and substantia nigra, respectively, from patients with Parkinson's disease relative to matched controls. Furthermore, radiolabelled angiotensin $AT_2$ receptor levels were decreased by some 60% in the caudate nucleus of patients with Parkinson's disease relative to control patients. In brain tissue homogenates from patients with Huntington's disease, the angiotensin $AT_1$ receptor recognition site levels were decreased by approximately 30% in putamen relative to the control patients whilst angiotensin $AT_2$ receptor levels were increased by some 90% in the caudate nucleus relative to the control patients. In brain tissue homogenates from patients with Alzheimer disease, the angiotensin $AT_2$ receptor recognition site levels were significantly increased by approximately 200% in the temporal cortex relative to the control patients. The present results indicate that the reduction of angiotensin $AT_1$ and/or $AT_2$ receptor recognition site levels in the caudate nucleus, putamen and substantia nigra correlates with the principal neuropathology associated with Parkinson's disease and as such indicates that at least a significant population of angiotensin $AT_1$ and $AT_2$ receptors are located on the human dopaminergic nigrostriatal pathway. In addition, the marked increase in the levels of angiotensin $AT_2$ receptor recognition sites in temporal cortex from patients with Alzheimer's disease correlates with some other markers associated with the renin-angiotensin system previously investigated in tissue from patients with this neurological disease.

*Keywords:* Parkinson's disease; Huntington's disease; Alzheimer's disease; Losartan; PD123177; Angiotensin $AT_1$ receptor; Angiotensin $AT_2$ receptor; [$^{125}$I][Sar$^1$,Ile$^8$]Angiotensin II; Brain, human

## 1. Introduction

The circulating octapeptide hormone angiotensin II serves an important role in the control blood pressure in both experimental animals and man (Peach, 1977). In addition to a peripheral site of action, angiotensin II also modulates components of the cardiovascular system via an interaction within the central nervous system (CNS) (Unger et al., 1988). It is also generally accepted that angiotensin II displays other physiological roles in the CNS (e.g. modulation of drinking behaviour and hormone release; e.g. Phillips, 1987; Reid and Rubin, 1987; Bottari et al., 1993). Furthermore, recent evidence indicates that manipulation of the renin-angiotensin system can modify levels of anxiety and cognitive performance in experimental animals (Barnes et al., 1990b, c; Barnes et al., 1991a, b; Costall et al., 1990). These behavioural responses presumably reflect an interaction within the CNS.

Previously, components of the central angiotensin system have been shown to be altered in patients with neurodegenerative diseases. For example, in Parkinson's disease, levels of angiotensin II receptors (angiotensin AT receptors) were markedly decreased in the human caudate nucleus, putamen and substantia nigra (Allen et al., 1992), although the binding studies were performed without the use of pharmacological agents to discriminate between angiotensin $AT_1$ ($AT_1$) and angiotensin $AT_2$ ($AT_2$) receptors. In addition to alteration in receptor levels, angiotensin-converting enzyme activity was significantly de-

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creased in the striatum from patients with Huntington's disease (Arregui et al., 1979). Uncharacteristic of a neurodegenerative disease, it has been demonstrated that angiotensin-converting enzyme activity and density are significantly increased in the temporal cortex from patients with Alzheimer's disease (Arregui et al., 1982; Barnes et al., 1991c), although the role of the angiotensin systems in Alzheimer's disease remains uncertain (Weiner et al., 1992; Koning et al., 1993).

With the availability of highly selective ligands for angiotensin $AT_1$ and $AT_2$ receptors (Timmermans et al., 1991), the present studies investigated the modifications in angiotensin $AT_1$ and $AT_2$ receptor levels in various neurodegenerative diseases. Such investigation will also provide evidence as to the cellular location of angiotensin receptor subtypes in the human brain. These studies follow the demonstration that angiotensin $AT_1$ and $AT_2$ receptors are widely distributed throughout animal (Gehlert et al., 1991; Rowe et al., 1992) and human brain (Barnes et al., 1993) although clear species differences exist in the relative levels of expression in different regions (e.g. see Barnes et al., 1993).

## 2. Materials and methods

### 2.1. Source of tissue

Human brain tissues were obtained from male and female patients with clinically and neuropathologically diagnosed Parkinson's disease, Huntington's disease or Alzheimer's disease and age-, sex- and post-mortem delayed-matched patients who had died without a neurological or psychiatric disorder (Table 1; four tissues (one caudate nucleus, one putamen, two substantia nigra) were from patients where the clinical diagnosis of idiopathic Parkinson's disease was not neuropathologically confirmed, data from these tissues, however, were within the range of the data generated from tissue from patients with neuropathologically confirmed Parkinson's disease). After removal of the brain at autopsy, individual hemispheres were frozen at −20°C before they were sliced (approximately 1 cm) using an electric meat slicer. The individual brain nuclei were then dissected using a scalpel and chopped (using a scalpel) over a cooling tray before being stored in Eppendorf tubes at −80°C until assay. All the tissues used in the present study were from the Cambridge MRC Brain Bank (Cambridge, UK).

Table 1
Details of the patients for the [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding studies

| Group | Sex | Age (years) | PM delay (h) |
|---|---|---|---|
| *Caudate* | | | |
| Control | 8M, 2F | 71 ± 5 | 60 ± 4 |
| Parkinson's disease | 5M, 5F | 74 ± 3 | 60 ± 5 |
| Control | 7M, 3F | 55 ± 3 | 40 ± 6 |
| Huntington's disease | 5M, 5F | 54 ± 3 | 46 ± 7 |
| Control | 6M, 4F | 77 ± 2 | 36 ± 8 |
| Alzheimer's disease | 5M, 5F | 77 ± 2 | 35 ± 8 |
| *Putamen* | | | |
| Control | 8M, 2F | 75 ± 2 | 55 ± 7 |
| Parkinson's disease | 4M, 6F | 74 ± 3 | 56 ± 6 |
| Control | 8M, 2F | 59 ± 4 | 38 ± 6 |
| Huntington's disease | 5M, 5F | 59 ± 4 | 38 ± 6 |
| Control | 4M, 6F | 79 ± 2 | 33 ± 9 |
| Alzheimer's disease | 2M, 8F | 80 ± 2 | 28 ± 6 |
| *Substantia nigra* | | | |
| Control | 8M, 2F | 73 ± 4 | 56 ± 6 |
| Parkinson's disease | 7M, 3F | 71 ± 4 | 51 ± 6 |
| Control | 5M, 5F | 58 ± 4 | 33 ± 5 |
| Huntington's disease | 7M, 3F | 56 ± 4 | 34 ± 5 |
| Control | 6M, 4F | 79 ± 2 | 22 ± 3 |
| Alzheimer's disease | 2M, 8F | 78 ± 2 | 22 ± 3 |
| *Temporal cortex (Brodmann area 21)* | | | |
| Control | 3M, 3F | 82 ± 1 | 28 ± 3 |
| Alzheimer's disease | 3M, 3F | 83 ± 1 | 24 ± 4 |
| *Temporal cortex (Brodmann area 22)* | | | |
| Control | 7M, 3F | 79 ± 2 | 38 ± 4 |
| Alzheimer's disease | 4M, 6F | 79 ± 2 | 38 ± 4 |
| *Cerebellum* | | | |
| Control | 3M, 2F | 71 ± 5 | 21 ± 4 |
| Parkinson's disease | 3M, 2F | 71 ± 7 | 21 ± 5 |
| Control | 4M, 4F | 77 ± 3 | 27 ± 3 |
| Alzheimer's disease | 2M,6F | 77 ± 3 | 26 ± 7 |
| *Hippocampus* | | | |
| Control | 4M, 3F | 71 ± 2 | 33 ± 8 |
| Alzheimer's disease | 5M, 2F | 75 ± 2 | 37 ± 7 |
| *Frontal cortex (Brodmann area 10)* | | | |
| Control | 4M, 2F | 78 ± 2 | 40 ± 9 |
| Alzheimer's disease | 1M, 5F | 77 ± 3 | 35 ± 7 |

M, male; F, female. For age and post-mortem (PM) delay, data represent the means ± S.E.M.

### 2.2. [$^{125}$I][Sar$^1$,Ile$^8$]Angiotensin II radioligand binding

Individual brain tissues were homogenized (polytron setting 7, 10 s) in ice-cold incubation buffer (mM: sodium chloride 150; sodium dihydrogen phosphate 50; magnesium chloride 10; ethylene glycol-bis(β-amino-ethyl-ether)*N*,*N*,*N*′,*N*′-tetra-acetic acid (EGTA) 5% and 0.4% w/v bovine serum albumin, final pH 7.4) and centrifuged (48 000 × *g*, 4°C, 10 min). The pellet was gently resuspended in incubation buffer and recentrifuged (48 000 × *g*, 4°C, 10 min). The pellet was gently resuspended in incubation buffer to generate the binding homogenate at a concentration of 100 mg original wet weight/ml. The binding homogenate was prepared immediately prior to assay. For [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding, test-tubes in triplicate contained 50 μl competing compound (final concen-

tration 1.0 μM of either losartan or PD123177) or vehicle (incubation buffer) and 50 μl [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II (final concentration 0.1 nM). Brain homogenate (250 μl) was added to initiate binding which was allowed to proceed for 90 min at 25°C before termination by rapid filtration through pre-wet (0.1% v/v polyethyleneimine in incubation buffer) GF/B filters followed by washing (8 s) with ice-cold incubation buffer. Radioactivity remaining on the filters was quantified using a gamma-counter. Protein content of the brain homogenates was assayed by the Bio-Rad Coomassie Brilliant Blue method (Bradford, 1976) using bovine serum albumin as the standard.

### 2.3. Drugs

Losartan (Dup 753; (2-*n*-butyl-4-chloro-5-hydroxymethyl-1-[2-(1*H*-tetrazol-5-yl)biphenyl-4-yl) methyl] imidazole; DuPont Merck Pharmaceuticals) and PD 123177 (1-(4-amino-3-methylphenyl)-methyl-5-diphenyl-acetyl-4,5,6,7-tetrahydro-1*H*-imidazo[4,5-*C*] pyridine-6-carboxylic acid; Dupont Merck Pharmaceuticals) were dissolved in a minimum quantity of distilled water and diluted to volume in incubation buffer. [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II (2200 Ci/mmol; NEN) was dissolved in incubation buffer. All drugs and reagents were used as recieved.

## 3. Results

### 3.1. Alterations in the levels of angiotensin $AT_1$ and $AT_2$ receptor recognition sites in brain tissues from patients with Parkinson's disease

In caudate nucleus homogenates from matched patients who had died without a neurological or psychiatric disorder, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II (0.1 nM) specific angiotensin $AT_1$ (defined by losartan, 1.0 μM) and $AT_2$ (defined by PD123177, 1.0 μM) receptor binding represented approximately 75% (e.g. total binding = 21 943 dpm, specific binding = 16 458 dpm, data from representive experiment; mean of triplicate determination) and 10% (e.g. total = 21 943 dpm, specific = 2017 dpm, data from representive experiment; mean of triplicate determination) of total binding, respectively. In caudate nucleus homogenates from patients with Parkinson's disease, specific [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding to angiotensin $AT_1$ and $AT_2$ receptors was significantly decreased by approximately 70% and 60%, respectively, when compared to the levels in tissues from matched control patients (Fig. 1). In putamen homogenates from matched patients without a neurological or psychiatric disorder, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II (0.1 nM) specific angiotensin $AT_1$ and $AT_2$ receptor binding represented approximately 70% (e.g. total

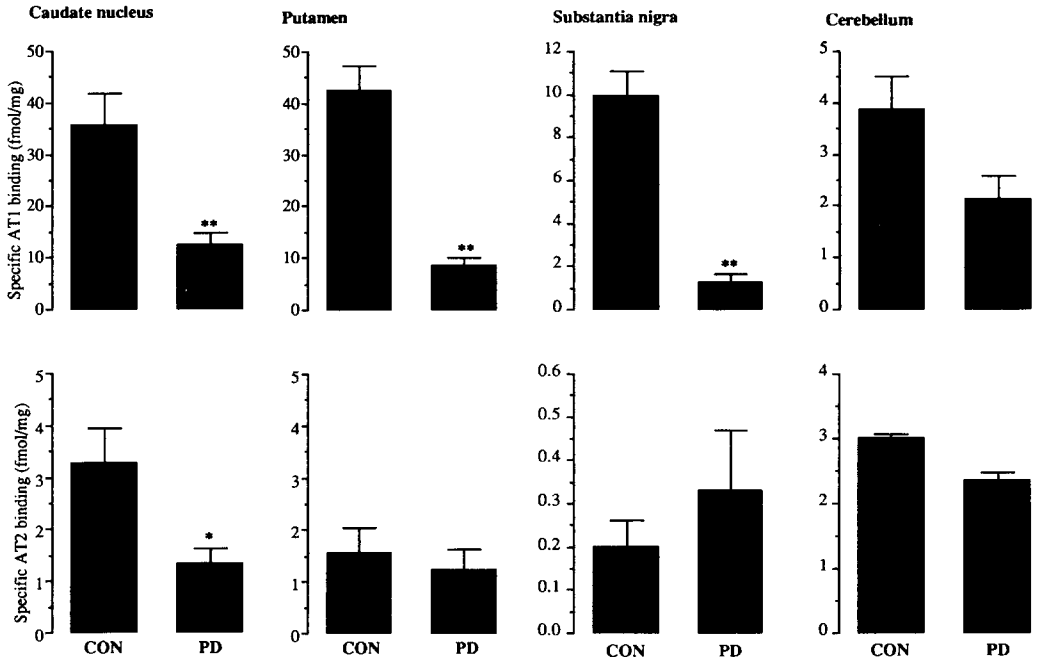


Fig. 1. Angiotensin $AT_1$ and $AT_2$ receptor recognition site levels in homogenates of caudate nucleus, putamen, substantia nigra and cerebellum from patients with Parkinson's disease (PD) and matched control patients (CON) who had died without a neurological or psychiatric disorder. Data represent the means ± S.E.M., $n = 5–10$, * $P < 0.05$, ** $P < 0.001$ (Student's *t*-test).

= 37 750 dpm, specific = 26 425 dpm, data from representive experiment; mean of triplicate determination) and 5% (e.g. total = 37 750 dpm, specific = 2342 dpm, data from representive experiment; mean of triplicate determination) of total binding, respectively. In putamen homogenates from patients with Parkinson's disease, specific [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding to angiotensin $AT_1$ receptors was significantly decreased by approximately 70%, whereas angiotensin $AT_2$ receptors levels were unaltered, when compared to the levels in tissues from matched control patients. In substantia nigra homogenates from both neurologically and psychiatrically normal matched patients, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II (0.1 nM) specific angiotensin $AT_1$ and $AT_2$ receptor binding represented approximately 80% (e.g. total = 9558 dpm, specific = 7646 dpm, data from representive experiment; mean of triplicate determination) and 7% (e.g. total = 9558 dpm, specific = 700 dpm, data from representive experiment; mean of triplicate determination) of total binding, respectively. In substantia nigra homogenates from patients with Parkinson's disease, specific [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding to angiotensin $AT_1$ receptors was significantly decreased by approximately 90%, when compared to the levels in tissues from matched control patients, whereas angiotensin $AT_2$ receptor specific binding was not significantly altered (Fig. 1). In cerebellum homogenates from matched patients who had died without a neurological or psychiatric disorder, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II (1.0 $\mu$M) specific angiotensin $AT_1$ and $AT_2$ receptor binding represented approximately 48% (e.g. total = 4515 dpm, specific = 2167 dpm, data from representive experiment; mean of triplicate determination) and 45% (e.g. total = 4515 dpm, specific = 2043 dpm, data from representive experiment; mean of triplicate determination) of total binding, respectively. In homogenates of cerebellum from patients with Parkinson's disease, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding to angiotensin $AT_1$ and $AT_2$ receptors tended to be reduced relative to values from tissue from control patients although this did not reach statistical significance (Fig. 1).

### 3.2. Alteration in the levels of angiotensin $AT_1$ and $AT_2$ recognition sites in brain tissue from patients with Huntington's disease

In caudate nucleus homogenates from both neurologically and psychiatrically normal matched patients and those with Huntington's disease, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II (0.1 nM) specific angiotensin $AT_1$ (defined by losartan, 1.0 $\mu$M) and $AT_2$ (defined by PD123177, 1.0 $\mu$M) receptor binding represented approximately 70% (e.g. total = 17 888 dpm, specific = 10 733 dpm, data from representive experiment; mean of triplicate determination) and 4% (e.g. total = 17 888 dpm, specific = 704 dpm, data from representive experiment; mean of triplicate determination) of the total binding, respectively. In the caudate nucleus homogenates from patients with Huntington's disease, specific [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding to angiotensin $AT_2$ receptor was significantly increased by approximately 90% compared to the values obtained from the matched control patients, whilst specific angiotensin $AT_1$ receptor binding levels were unaltered. In putamen homogenates

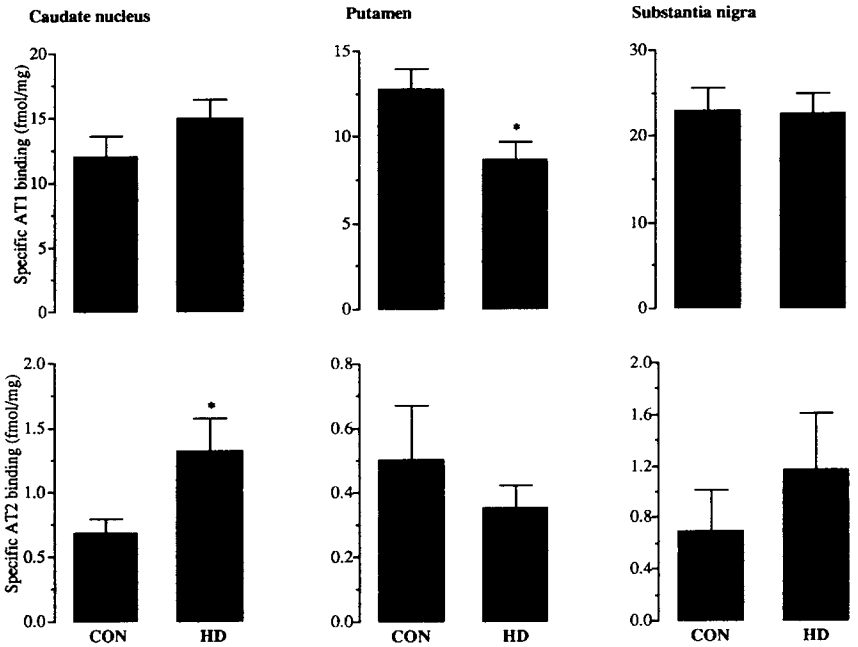


Fig. 2. Angiotensin $AT_1$ and $AT_2$ receptor recognition site levels in homogenates of caudate nucleus, putamen and substantia nigra from patients with Huntington's disease (HD) and matched control patients (CON) who had died without a neurological or psychiatric disorder. Data represent the means ± S.E.M., $n = 10$, * $P < 0.05$ (Student's $t$-test).

from matched patients who had died without a neurological or psychiatric disorder, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II specific angiotensin $AT_1$ and $AT_2$ receptor binding represented approximately 70% (e.g. total = 11 957 dpm, specific = 8370 dpm, data from representive experiment; mean of triplicate determination) and 5% (e.g. total = 11957 dpm, specific = 573 dpm, data from representive experiment; mean of triplicate determination) of the total binding, respectively. In putamen homogenates from patients with Huntington's disease, specific [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding to angiotensin $AT_1$ receptor was significantly reduced by approximately 30% whilst specific angiotensin $AT_2$ binding was also reduced although this did not reach statistical significance, when compared to the levels in tissue from matched control patients. In substantia nigra homogenates from matched patients without a neuro-

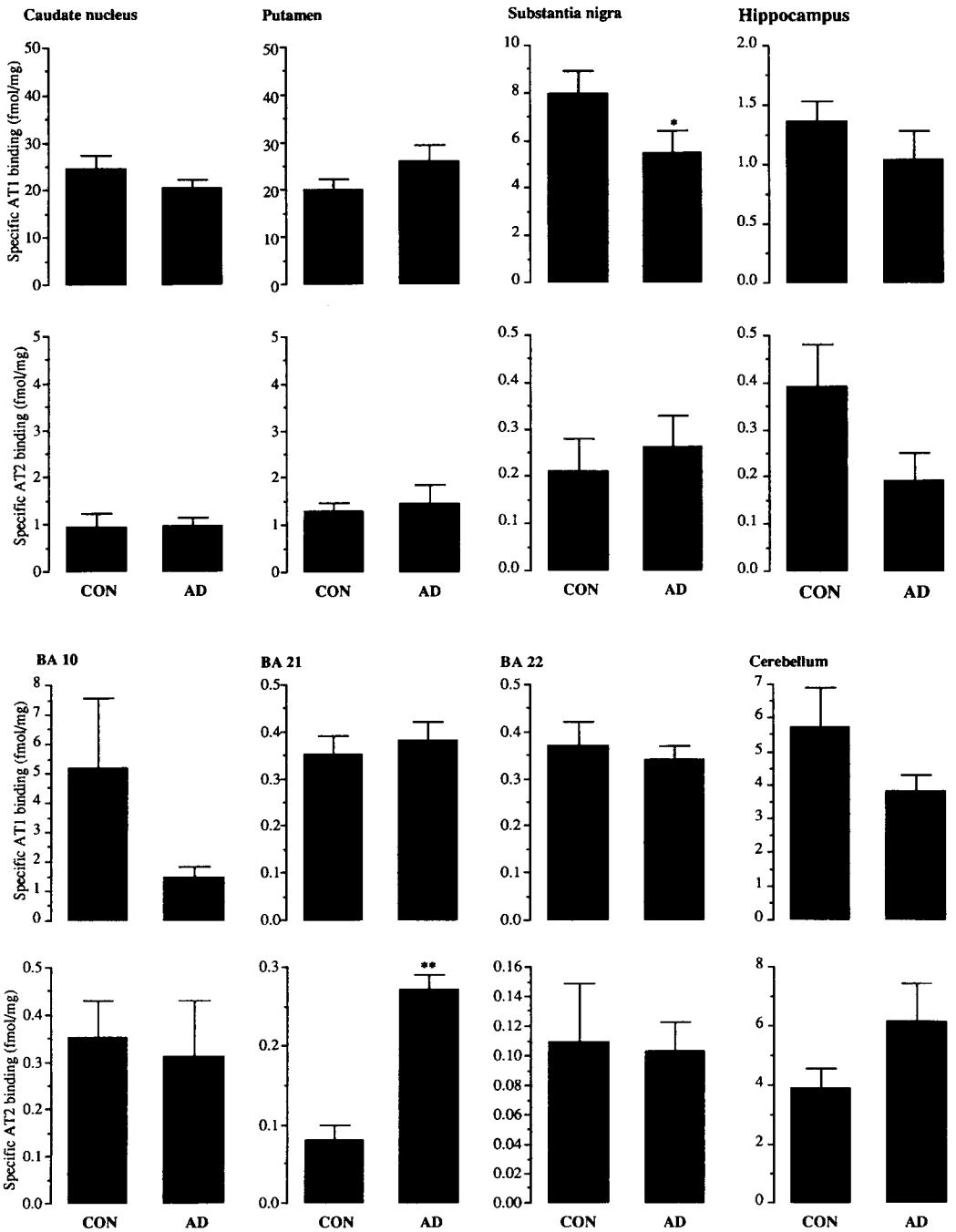


Fig. 3. Angiotensin $AT_1$ and $AT_2$ receptor recognition site levels in homogenates of caudate nucleus, putamen, substantia nigra, hippocampus, cerebellum, frontal cortex (BA 10) and temporal cortex (BA 21 and BA 22) from patients with Alzheimer's disease (AD) and matched control patients (CON) who had died without a neurological or psychiatric disorder. Data represent the means ± S.E.M., $n = 6–10$, $^*$ $P < 0.05$ and $^{**}$ $P < 0.001$ (Student's $t$-test).

logical or psychiatric disorder, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II specific angiotensin $AT_1$ and $AT_2$ receptor binding represented approximately 80% (e.g. total = 14425 dpm, specific = 11540 dpm, data from representive experiment; mean of triplicate determination) and 5% (e.g. total = 14425 dpm, specific = 682 dpm, data from representive experiment; mean of triplicate determination) of the total binding, respectively. In substantia nigra homogenates from patients with Huntington's disease, specific angiotensin $AT_1$ and $AT_2$ receptor binding levels were not significantly different to levels in tissue from control patients (Fig. 2).

### *3.3. Alteration in the levels of angiotensin $AT_1$ and $AT_2$ recognition sites in brain tissues from patients with Alzheimer's disease*

In substantia nigra homogenates from matched patients without a neurological or psychiatric disorder, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II (0.1 nM) specific angiotensin $AT_1$ (defined by losartan, 1.0 $\mu$M) and $AT_2$ (defined by PD123177, 1.0 $\mu$M) receptor binding represented approximately 70% (e.g. total = 7157 dpm, specific = 5010 dpm, data from representive experiment; mean of triplicate determination) and 3% (e.g. total = 7157 dpm, specific = 240 dpm, data from representive experiment; mean of triplicate determination) of the total binding, respectively. In substantia nigra homogenates from patients with Alzheimer's disease, specific [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II binding to angiotensin $AT_1$ receptor was significantly reduced by approximately 30% whilst specific angiotensin $AT_2$ receptor binding was similar to the levels in tissue from matched control patients (Fig. 3). In temporal cortex (Brodmann area 21) homogenates from patients who had died without a neurological or psychiatric disorders, [$^{125}$I][Sar$^1$,Ile$^8$] angiotensin II specific angiotensin $AT_1$ and $AT_2$ receptor binding represented approximately 15% (e.g. total = 3186 dpm, specific = 478 dpm, data from representive experiment; mean of triplicate determination) and 10% (e.g. total = 3186 dpm, specific = 302 dpm, data from representive experiment; mean of triplicate determination) of the total binding, respectively. In temporal cortex (Brodmann area 21) homogenates from patients with Alzheimer's disease, specific angiotensin $AT_2$ receptor binding was significantly increased by approximately 200% when compared to the levels in tissue from matched control patients, whilst specific angiotensin $AT_1$ receptor binding remained unaltered (Fig. 3).

In caudate nucleus, putamen, frontal cortex (Brodmann area 10), cerebellum and hippocampus homogenates from matched patients without a neurological or psychiatric disorder, [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II specific angiotensin $AT_1$ receptor binding represented approximately 70%, 65%, 30%, 45% and 48% of the total binding, respectively, and specific angiotensin $AT_2$ receptor binding represented approximately 2%, 5%, 7%, 35% and 30% of the total binding, respectively. The levels of both angiotensin $AT_1$ and $AT_2$ receptors in these brain regions were not significantly different in tissues from patients with Alzheimer's disease (Fig. 3) although angiotensin $AT_1$ receptor levels in the frontal cortex (Brodmann area 10) tended to be lower in tissue from patients with Alzheimer's disease (Fig. 3).

## 4. Discussion

Previous studies have demonstrated that the peptide angiotensin II analogue, [sarcosine$^1$,isoleucine$^8$]angiotensin ([Sar$^1$,Ile$^8$]angiotensin II) displays higher affinity for both angiotensin $AT_1$ and $AT_2$ receptors than the endogenous ligand angiotensin II (Schinke et al., 1991). Indeed, the radioligand [$^{125}$I][Sar$^1$,Ile$^8$]angiotensin II has been successfully applied to label angiotensin II receptors in various peripheral tissues (e.g. Schinke et al., 1991) and brain (e.g. Chang et al., 1990; Healey et al., 1986). In the present studies, this radioligand was used to label both angiotensin $AT_1$ and $AT_2$ receptor recognition sites in the brain tissues from matched control patients and patients with Parkinson's, Huntington's or Alzheimer's disease.

Radioligand binding techniques have identified angiotensin AT receptor recognition sites present throughout the human brain with relatively high levels associated with the nigro-striatal systems (e.g. Allen et al., 1992; Barnes et al., 1993). The present studies used the selective competing ligands, losartan and PD123177 (Timmermans et al., 1991), to define the angiotensin $AT_1$ and $AT_2$ receptor population, respectively and demonstrated significant reductions of angiotensin $AT_1$ receptors in the caudate nucleus, putamen and substantia nigra and a significant reduction of angiotensin $AT_2$ receptors in the caudate nucleus of patients with Parkinson's disease (although because of the relatively low levels of $AT_2$ receptor expression in the caudate nucleus, caution is necessary concerning the exact extent of the reduction in receptor levels). These findings are consistent with previous works demonstrating a reduction of unclassified angiotensin receptors in these nuclei (Allen et al., 1992).

Given the principle neurodegeneration in Parkinson's disease is a loss of the dopamine neurones within the nigro-striatal pathway (e.g. Price et al., 1979), it is tempting to speculate that the loss of angiotensin $AT_1$ and $AT_2$ receptors are associated with the loss of dopaminergic receptors; it must be appreciated, however, that other neurones also degenerate in this neurodegenerative disease (e.g. Jellinger, 1990) and therefore these systems may also contribute.

Functional data also implicate an association between the angiotensin system and the dopaminergic nigro-striatal system (Brown and Barnes, 1993; Mendelsohn et al., 1993; Simonnet and Giorsuieff-Chesselet, 1979; Steward et al., submitted), although some of these studies were performed

with rat tissues which often display levels of angiotensin AT receptors in the striatum below the limit of detection.

In the basal ganglia from patients with Huntington's disease, the levels of angiotensin II receptor were less altered relative to comparable tissues from patients with Parkinson's disease. Thus, the present study has demonstrated that angiotensin $AT_1$ receptors are reduced by some 35% in the putamen from patients with Huntington's disease whilst the levels of angiotensin $AT_1$ receptors in the caudate nucleus and substantia nigra are unaltered in this neurodegenerative disorder. In the caudate nucleus of patients with Huntington's disease, however, angiotensin $AT_2$ receptor levels were increased by some 90% (again caution is warranted in the precise level of this change due to the low level of angiotensin $AT_2$ receptor expression in this nucleus). It is generally accepted that the striatum from patients with Huntington's disease degenerates severely, but the ascending nigrostriatal system remains largely intact (e.g. Quarrel, 1991; Reynolds and Pearson, 1992). This supports one of the main findings of the present study that angiotensin II receptors in the basal ganglia are principally located on neurones that degenerate in Parkinson's disease (presumably dopamine neurones). Previous biochemical studies have demonstrated that angiotensin-converting enzyme activities were markedly reduced by some 60–90% in the caudate nucleus, putamen, globus pallidus and substantia nigra from patients with Huntington's disease (Arregui et al., 1979). Hence, if this angiotensin-converting enzyme activity is associated with degenerating neurones, then this may indicate that angiotensin II receptors are principally located post-synaptically to the degenerating neurones in Huntington's disease. The elevation in angiotensin $AT_2$ receptor levels in the caudate nucleus of patients with Huntington's disease may be associated with a compensatory increase in receptor density following the degeneration of these angiotensin II synthesising neurones.

To date, two studies have investigated the angiotensin system in the brain of patients with Alzheimer's disease (Arregui et al., 1982; Barnes et al., 1991c). Both studies investigated the activity or levels of angiotensin-converting enzyme in the brain and found elevations in either the activity or density of this enzyme in brain tissues from patients with Alzheimer's disease. Thus, the first study demonstrated that angiotensin-converting enzyme activities were significantly increased in frontal cortex, parahippocampal gyrus, medial hippocampus and caudate nucleus of patients with Alzheimer's disease (Arregui et al., 1982), whilst the second study demonstrated an elevation in the density of angiotensin-converting enzyme in the temporal cortex from patients with Alzheimer's disease (Barnes et al., 1991c). It is therefore of interest that in the present studies, angiotensin $AT_2$ receptor levels were increased by over 300% in temporal cortex of patients with Alzheimer's disease when compared with the matched control levels. Together with the previous studies, these findings suggest that the activity of the central angiotensin system is increased in patients with Alzheimer's disease. Given this association, it is of interest that a pharmacologically induced reduction in the activity of the angiotensin system e.g. via the administration of angiotensin-converting enzyme inhibitors, angiotensin $AT_1$ or $AT_2$ receptor antagonists, results in an improvement in the cognitive performance of laboratory animals (Barnes et al., 1990b, c, 1991a, b; Bartus et al., 1982). It may also be relavant that angiotensin II inhibits the neuronal release of acetylcholine in both rat and human cerebral cortex which may contribute to the cognitive deficit associated with this neurodegenerative disease (Barnes et al., 1989, 1990a; Bartus et al., 1982; Usinger et al., 1988). The importance of the abnormality in the angiotensin system in the temporal cortex is of special interest, since in Alzheimer's disease, the temporal cortex is rich in intraneuronal neurofibrillary tangles and extracellular amyloid plaques (Rogers and Morrison, 1985; Ball et al., 1985) which histologically characterise this neurodegenerative disease. It should be noted, however, that previous studies have failed to demonstrate a significant correlation between angiotensin-converting enzyme activity and the dysfunction in Alzheimer's disease (Koning et al., 1993; Weiner et al., 1992).

In summary, the present studies have investigated the levels of angiotensin $AT_1$ and $AT_2$ receptors in brain tissues from patients with neurodegenerative diseases compared to matched control patients. The principle findings suggest that significant population of both angiotensin $AT_1$ and $AT_2$ receptors within the basal ganglia are located on neurones that degenerate in Parkinson's disease (dopamine neurones?) and that angiotensin $AT_2$ receptor levels are elevated in the temporal cortex of patients with Alzheimer's disease.

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