Eur Arch Psychiatr Neurol Sci (1989) 239:177–179

European Archives of Psychiatry and Neurological Sciences

Brain Quinolinic Acid in Alzheimer's Dementia

E. Sofic[1], J. Halket[2], A. Przyborowska[2], P. Riederer[1], H. Beckmann[1], M. Sandler[2], and K. Jellinger[3]

[1]Department of Psychiatry, Clinical Neurochemistry, University of Würzburg, Würzburg, Federal Republic of Germany
[2]Department of Chemical Pathology, Queen Charlotte's Hospital, London, England
[3]Ludwig Boltzmann Institute of Clinical Neurobiology, Lainz Hospital, Vienna, Austria

Summary. Quinolinic acid (QA) content was measured in postmortem frontal and temporal cortex, putamen and cerebellum obtained from patients with senile dementia of Alzheimer type (SDAT), Huntington's disease (HD) and controls, using a gas chromatography/mass spectrometry method. There were no significant group differences in QA content of any of the regions examined. The data do not support the hypothesis that an accumulation of QA plays a role in neuronal degeneration occurring in the frontal and temporal cortex, putamen and cerebellum of patients with SDAT.

Key words: Quinolinic acid – Senile dementia of Alzheimer type – Huntington's disease – Neurotoxin – Dementia – Postmortem brain – Gas chromatography/mass spectrometry

Introduction

An excess of the tryptophan metabolite quinolinic acid (QA) in the brain has been hypothetically related to the pathogenesis of senile dementia of Alzheimer type (SDAT), Huntington's disease (HD) and epilepsy (Schwarcz et al. 1984; Feldblum et al. 1988). Furthermore, an involvement of QA in the neuronal cell loss occurring during aging might also be possible. In patients affected by SDAT a degeneration of neurons has been shown in the nucleus basalis of Meynert (Whitehouse et al. 1982), hippocampus (Jellinger and Riederer 1984; Ball et al. 1985; Hyman et al. 1984) and in various cortical areas (Mountjoy et al. 1983; Terry et al. 1981). The pathogenesis of this degeneration is unknown. One possibility would be a substantial increase of the endogenous neurotoxic tryptophan metabolite QA.

QA, a glutamate receptor agonist, causes a selective degeneration of intrinsic neurons when injected into the rat striatum (Beal et al. 1986). Lesions produced by intrastriatal injections of QA resemble neuropathological and neurochemical changes that are characteristic of HD: a depletion of neurotransmitters contained within striatal spiny neurons, e.g. γ-aminobutyric acid, reduction in the number of cholinergic neurons, while dopamine is unaffected (Schwarcz et al. 1983, 1984; Beal et al. 1986). Quinolinate's intermediate biosynthetic enzyme, 3-hydroxyanthranilate oxygenase (EC 1.13.11.6) has now been detected in brain tissue and can be expected to control the metabolism of QA (Foster et al. 1985, 1986). The activity of 3-hydroxyanthranilate oxygenase is increased in HD brains as compared with controls (Schwarcz et al. 1988). Furthermore, accumulation of QA could be due to a deficiency in its degradation. Quinolinic-phosphoribosyl-transferase is the first degradative enzyme of QA. The activity of this enzyme is decreased in epileptic human brain tissue (Feldblum et al. 1988).

In the present study we have examined the content of QA in various brain regions of patients with SDAT, HD and controls.

Materials and Methods

At autopsy the brain was divided by sagital section. One half brain was deep-frozen for biochemical assay. The other half brain was fixed in buffered formalin. Multiple blocks were embedded in paraffin and the preparation of sections followed routine methods (Khachaturian 1985). The samples for bio-

Offprint requests to: E. Sofic, Clinical Neurochemistry, Department of Psychiatry, University of Würzburg, Füchsleinstrasse 15, D-8700 Würzburg, Federal Republic of Germany

Table 1. Quinolinic acid content in the frontal and temporal cortex, putamen and cerebellum of human subjects

Age	Sex	Neuropathological diagnosis	Frontal cortex	Temporal cortex	Putamen	Cerebellum
79.7 ± 3.4	2 M, 2 F	Controls	0.77 ± 0.37	0.34 ± 0.10	0.41 ± 0.11	0.58 ± 0.18
82.5 ± 4.9	2 M, 2 F	SDAT	0.54 ± 0.16	0.91 ± 0.32	0.58 ± 0.13	0.76 ± 0.20
41	F		0.11	ND	0.27	0.36
35	F	HD	0.71	ND	0.20	0.45

The content of quinolinic acid is expressed as nmol/g tissue. Results are expressed as means ± SEM, except for HD where individual data points are given. ND = Not determined

chemical assays were removed using the atlas of Nieuwenhuys et al. (1981).

Brain tissue (frontal and temporal cortex, putamen and cerebellum) from four patients with SDAT, two patients with HD and four neurologically normal control subjects (Table 1) was obtained; tissue samples were stored again at −80°C in airtight plastic tubes and assayed within 3 weeks.

Postmortem time in all cases was between 3 and 15 h. Our samples were analysed in part in parallel with those of Reynolds et al. (1988) and have been identified by pathological and neuropathological examination and additional studies of the case histories.

Controls had died without any evidence of neurological or psychiatric disease. All brains were examined histopathologically by routine staining methods and were seen by a neuropathologist. The histopathological findings in all four controls consisted of non-specific senile changes. Drug treatment consisted of cardiovascular active drugs and antibiotics only. The cause of death was pneumonia and oedema of the lung ($n = 1$), myocardial infarction ($n = 1$), carcinoma ($n = 1$) and cardiac arrest after diabetes mellitus ($n = 1$). In all cases of SDAT the histopathological examination showed severe SDAT with accentuation in the hippocampus. Histopathological changes in the occipital cortex ($n = 1$) and amyloid angiopathy were notable in a single patient. Two patients with SDAT were drug-free, while in two other patients neuroleptic drug treatment had been withdrawn 6 weeks before death. The cause of death (SDAT group) was bronchopneumonia ($n = 1$), pneumonia and hypertensive heart disease ($n = 2$) and oedema of the lung ($n = 1$).

The two cases of HD on histopathological examination showed severe atrophy of the striatum, generally accompanied by cortical cell loss. Drug treatment had consisted of antibiotics and neuroleptics. The cause of death (HD group) was bronchopneumonia ($n = 1$) and aspiration ($n = 1$).

QA was isolated and measured using a modification of previous methods (Wolfensberger et al. 1983; Moroni et al. 1986a, b). Using 2,4-pyridine dicarboxylic acid as the internal standard, tissue (300 mg) was homogenized in 0.5 ml of 0.3 *M* formic acid and centrifuged at 12000 *g* for 3 min. The pellet was resuspended in 0.5 ml phosphate buffer (0.1 *M*, pH 9.4) and recentrifuged. The combined supernatants were centrifuged at 50000 *g* for 10 min and applied to a prewashed strong anion exchange Bond Elut sorbent column. After washing with consecutive 2-ml volumes of water, 0.1 *M* formic acid and 25%, 50% and 80% methanol, QA and its internal standard were eluted with 1 ml formic acid/methanol (20:80/vol/vol). This mixture was dried in a stream of nitrogen and treated by heating for 90 min at 110°C with 50 µl each of hexafluoroisopropanol and trifluoroacetic anhydride. This was dried under nitrogen and the derivatives dissolved in 50 µl heptane prior to injection of 1 µl into the gas chromatograph. This employed a 15 m × 0.32 mm chemically bonded OV1 column at 120°C and mass spectrometer (Hewlett Packard mass selective detector) to determine ions at m/z 272, 300 and 448.

Concentrations of QA were calculated from the ratios of peak areas of QA and internal standard (respective retention times 5.6 and 4.8 min) by comparison with results from a calibration curve obtained by addition of QA (10–100 ng) to aliquots of a tissue homogenate and taken through the above procedure.

For statistical comparisons Student's *t*-test and the Wilcoxon rank sum test were performed.

Results

Individual values of QA content in tissues are shown in Table 1. No significant difference is apparent in QA concentrations between the three groups in the frontal and temporal cortex, putamen or the cerebellum.

Discussion

The results indicate that the concentration of QA in postmortem human cortex, putamen and cerebellum in SDAT is unchanged compared with controls. These data do not support the speculative link between QA and SDAT. However, our findings are in conformity with the results of Moroni et al. (1986a), who reported no changes in cortical QA content in SDAT. Furthermore, no correlation was found between the severity of Alzheimer changes and QA content in the cortex (Moroni et al. 1986a).

Recently, no changes in the content of quinolinate in cerebrospinal fluid (Schwarcz et al. 1988) or in urine (Heyes et al. 1985) were reported in HD samples. Our preliminary data in HD are limited by the fact that they could not be compared with age-matched controls. However, there is no evidence that QA undergoes age-dependent changes (Moroni et al. 1986b). In that respect, the data of our two cases are in line with those in the literature (Reynolds et al. 1988).

In conclusion, our data do not support the hypothesis that an accumulation of QA plays an important

role in the neuronal degeneration occurring in the course of SDAT.

References

Ball MJ, Hachinski V, Fox A, Kirshen AJ, Fisman M, Blume W, Kral VA, Fox H (1985) A new definition of Alzheimer's disease: a hippocampal dementia. Lancet I: 14–16

Beal MF, Kowal NW, Ellison DW, Mazurek MF, Schwartz KJ, Martin JB (1986) Replication of the neurochemical characteristics of Huntington's disease by quinolinic acid. Nature: 321: 168–171

Feldblum S, Rougier A, Loiseau H, Loiseau P, Cohadon F, Morselli PL, Lloyd KG (1988) Quinolinic-phosphoribosyltransferase activity is decreased in epileptic human brain tissue. Epilepsia 29: 523–529

Foster AC, Zinkand WC, Schwarcz R (1985) Quinolinic-acid phosphoribosyltransferase in rat brain. J Neurochem 44: 446–454

Foster AC, Zinkand WC, Schwarcz R (1986) Synthesis of quinolinic acid by 3-hydroxyantranilic acid oxygenase in rat brain tissue in vitro. J Neurochem 47: 23–30

Heyes MP, Garnett ES, Brown RR (1985) Normal excretion of quinolinic acid in Huntington's disease. Life Sci 37: 1811–1816

Hyman BT, Van Hoesen GW, Damasio AR, Barnes CL (1984) Alzheimer's disease: cell-specific pathology isolates the hippocampal formation. Science 225: 1168–1170

Jellinger K, Riederer P (1984) Dementia in Parkinson's disease and (Pre)senile dementia of Alzheimer type: morphological aspect and changes in the intracerebral MAO activity. In: Hassler RG, Christ JF (eds) Advances in neurology, vol 40. Raven Press, New York, pp 199–210

Khachaturian Zaven S (1985) Diagnosis of Alzheimer's disease. Arch Neurol 42: 1097–1105

Moroni F, Lombardi G, Robitaille Y, Etienne P (1986a) Senile dementia and Alzheimer's disease: lack of changes of the cortical content of quinolinic acid. Neurobiol Aging 7: 249–253

Moroni F, Lombardi G, Carla V, Lal S, Etienne P, Nair NPV (1986b) Increase in the content of quinolinic acid in cerebrospinal fluid and frontal cortex of patients with hepatic failure. J Neurochem 47: 1667–1671

Mountjoy CQ, Roth M, Evans NJR, Evans HM (1983) Cortical neuronal counts in normal elderly controls and demented patients. Neurobiol Aging 4: 1–11

Nieuwenhuys R, Voogd J, van Huijzen C (1981) The human central nervous system. A synopsis and atlas. Springer, Berlin Heidelberg New York

Reynolds GP, Pearson SJ, Halket J, Sandler M (1988) Brain quinolinic acid in Huntington's disease. J Neurochem 50: 1959–1960

Schwarcz R, Whetsell WO Jr, Mangano RM (1983) Quinolinic acid: An endogenous metabolite that produces axon-sparing lesions in rat brain. Science 219: 316–318

Schwarcz R, Foster AC, French ED, Whetsell WO Jr, Köhler C (1984) Exitotoxic models for neurodegenerative disorders. Life Sci 35: 19–32

Schwarcz R, Okuno E, White RJ, Bird ED, Whetsell WO Jr (1988) 3-Hydroxyantranilate oxygenase activity is increased in the brains of Huntington disease victims. Proc Natl Acad Sci USA, Neurobiology 85: 4079–4081

Terry RD, Peck A, Deteresa R, Schechter R, Horoupian DS (1981) Some morphometric aspects of the brain in senile dementia of the Alzheimer type. Ann Neurol 10: 184–192

Whitehouse PJ, Price DL, Struble RG, Clark AW, Coyle JT, De Long MR (1982) Alzheimer's disease and senile dementia: loss of neurons in the basal forebrain. Science 215: 1237–1239

Wolfensberger M, Amsler U, Cuenod M, Foster AC, Whetsell WO, Schwarcz R (1983) Identification of quinolinic acid in rat and human brain tissue. Neurosci Lett 41: 247–252

Received January 30, 1989