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Scientific Correspondence

Unusual neuropathological features and increased brain aluminium in a resident of Camelford, UK

The possible role of aluminium in the pathogenesis of Alzheimer's disease (AD) has been hotly debated over the past few decades. Although the so-called 'aluminium hypothesis' was popular in the 1970s and 1980s, it has gradually fallen out of favour in the past few years possibly following a number of inconclusive and contradictory human environmental/clinical studies. Nevertheless, there have from time to time been reminders in the media of environmental accidents; these have prevented the topic from disappearing completely from public memory. One such accident occurred on July 6th 1988 in Camelford, Cornwall when 20 tonnes of aluminium sulphate was mistakenly discharged into the mains water supply. Twenty thousand people were exposed to concentrations of aluminium which were 500–3000 times the acceptable limit under European Union legislation. Over the subsequent years there have been UK government inquiries into the supposed environmental impact and occasional clinical follow-up studies documenting declining cerebral function in those exposed to the contaminated water but very little neuropathological data has been published. To our knowledge, the case described here is only the second neuropathological description.

In 1988 a 43-year-old man living in the Camelford area of Cornwall was exposed to high concentrations of aluminium in the water supply following the accidental discharge. Six years later (age 49) he presented with some memory problems which were put down to difficulty in concentration. A SPECT scan, however, was reported as normal. By the age of 55 he had definite memory problems and indeed 5 years later he was admitted to a nursing home. He had developed expressive dysphasia with dyspraxia, poor visuospatial skills, visual hallucinations and then myoclonic jerks. He died at the age of 69. It was stated briefly in the clinical notes that his mother probably had dementia. Consent was given for retention of the brain and therefore following a limited autopsy, the brain was removed and half was frozen and half fixed in formalin.

The fixed left half of the brain weighed 463 g and revealed cerebral atrophy. The half brain stem and cerebellum weighed 81 g. Coronal slicing of the half cerebrum confirmed cerebral atrophy, and there was considerable softening. The hippocampus was small and there was pallor to the locus coeruleus. The cerebellum appeared normal. The histology showed extensive and widespread hyperphosphorylated (HP) tau deposition (Figure 1**a**) in the form of neuritic plaques (Figure 1**b**), neurofibrillary tangles (Figure 1**b** inset) and neuropil threads and extensive Aβ protein in the form of plaques (both cored and diffuse) and amyloid angiopathy (Figure 1**c**). This therefore reached an AD modified Braak (BrainNet Europe (BNE)) stage of VI [1]. It also reached a Thal Aβ phase of 5 and a frequent CERAD neuritic plaque score [2]. In addition, there was widespread α-synuclein Lewy body pathology extending from the brain stem to involve frontal, temporal and parietal neocortex (Figure 1**d**), thereby reaching a diffuse neocortical stage of Dementia with Lewy bodies according to McKeith criteria [3]. A number of α-synuclein positive (non-Papp-Lantos) glial cytoplasmic inclusions (GCIs) (mainly astrocytic) were seen in the cingulate, parietal and temporal cortex (Figure 1**d** inset). There was also TDP-43 pathology in the limbic system and extending into the neocortex (as demonstrated by antibodies to phosphorylated (p)-TDP-43). The TDP-43 pathology was seen in the form of neuronal cytoplasmic inclusions (NCIs) (Figures 1**e** and **f**), thin neurites (Figure 1**f**) and very occasional neuronal intra-nuclear inclusions (NII) (Figure 1**g**). There were also occasional GCIs. The hippocampus, frontal, parietal, motor and temporal cortices showed TDP-43 pathology together with the basal ganglia, but not the medulla or occipital cortex. The pTDP-43 staining pattern most closely matched type A [4]. Therefore there was AD pathology, Lewy body pathology and TDP-43 pathology in the brain. Whilst this combined pathology with this intensity is somewhat unusual it is certainly not unique. The pathological findings are summarized in Table S1. Subsequent analysis revealed elevated

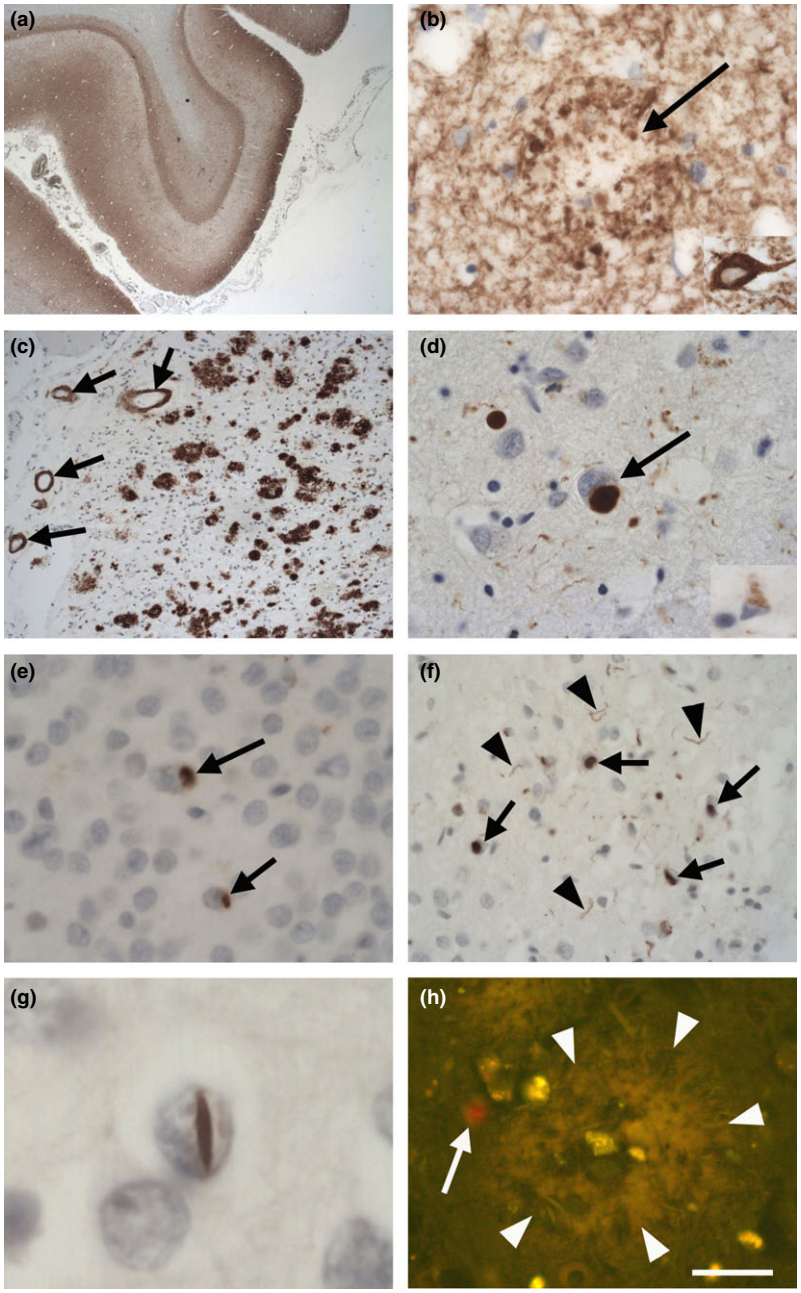

Figure 1. Immunohistochemical features in the brain. (**a**) There was extensive HP-Tau immunopositivity throughout the neocortex including here in the occipital cortex. (**b**) There were also numerous neuritic plaques (arrow) throughout the neocortex such as here in the temporal lobe and there were also abundant neurofibrillary tangles (inset) as seen here in the frontal lobe. Anti -HP-Tau in (**a**), (**b**) and (**b** inset). (**c**) There was Aβ deposition in the cerebral cortex in the form of plaques as seen here, together with significant amyloid angiopathy (arrows). Anti-Aβ. (**d**) Numerous Lewy bodies were evident in the brain stem and limbic region but also here (arrow) in the parietal cortex. A number of α-synuclein immunopositive glial (mainly astrocytic) cytoplasmic inclusions (inset) were also evident. Anti-α-synuclein. (**e**)-(**g**) The immunohistochemistry for phosphorylated (p)TDP-43 revealed neuronal cytoplasmic inclusions (NCIs) in (**e**) the dentate gyrus of the hippocampus (arrows) and the neocortex (**f**) such as here in the temporal cortex (arrows). In addition, neurites were seen (**f**) (arrowheads). Very occasional neuronal intranuclear inclusions were seen in the frontal lobe (**g**). Anti-pTDP-43 in (**e**)-(**g**). (**h**) Fluorescent image from the occipital cortex and stained with lumogallion revealing an orange- coloured deposit corresponding to aluminium (arrow) adjacent to a plaque (defined by arrowheads). Scale Bar: (**a**)- 2000μm, (**b**) and (**b** inset)- 40 μm, (**c**) -250 μm, (**d**) and (**h**)-35 μm, (**d** inset)-15 μm, (**e**) -25 μm (**f**)- 55 μm, (**g**) -10 μm.

aluminium levels in the brain especially the parietal and occipital lobes where there were levels up to 5.58 and 4.45 μg/g dry weight respectively. The median control level derived from previous work being 1.02 μg/g dry weight [5], and those values above 2.0 considered pathologically concerning. Furthermore, aluminium was able to be visualized as orange deposits with a novel fluorescent method using lumogallion (Figure 1**h**) and this was described in a previous publication [6]. These deposits, although mainly extracellular did not appear to consistently correspond to any obvious pathological features [6]. DNA extraction was carried out from the frozen tissue and analysis showed no evidence of pathological mutations for β-Amyloid precursor protein, presenilin 1 or 2 or progranulin. Obviously this does not completely rule out an underlying genetic cause. The Apolipoprotein E (APOE) genotype was found to be ε4/4.

To our knowledge, there has only been one previously published account of the neuropathology in a brain from a patient with cognitive decline who was living in the Camelford area at the same time as the patient described here [7]. The woman was aged 44 when the accident occurred, 58 when investigated for mental decline and she died a year later. Interestingly, as in our case she had suffered visual hallucinations, and dysphasia but also had hypertonia. Neuropathologically there were few neuritic plaques or neurofibrillary tangles; the main abnormality being amyloid angiopathy. There was, however, Lewy body pathology as in our case but here it was confined to brain stem and limbic system. The APOE genotype in the previous case was ε4/4 as in our case. This combination is associated with a greater risk for developing sporadic AD [8].

Three UK Government reports dealt with the Camelford accident between 1989 and 2013 and the final conclusion stated that there was no evidence to suggest a link between the incident and delayed health effects [9]. The interim findings had been disputed by a separate clinical study in 1999 which suggested an association of aluminium exposure with considerable damage to cerebral function [10]. This study itself was, however, criticized for the manner of selection of cases [11]. Whilst some separate environmental studies had shown an association between dementia and high levels of aluminium in the water supply [12] other investigations revealed no such link [13]. It is known that patients on renal dialysis can develop an encephalopathy probably due to aluminium in the water but neuropathologically this does not show typical AD pathology and experimentally at the ultrastructural level there are collections of straight rather than AD type helical filaments [14,15]. One investigation showed that the gastrointestinal absorption of aluminium was greater in patients with AD than controls suggesting that the elevated brain levels of the metal could be the effect rather than cause of the disease [16], although it could be argued that the resulting increase in cerebral aluminium may also exacerbate the disease process. To complicate the issue further a more recent study has shown very high brain aluminium levels in cases of familial AD [17]. In addition, there have been occasional suggested links between aluminium and Lewy body pathology [18,19], but none as yet reported between TDP-43 pathology and aluminium.

Aluminium is a relatively common metal found on the earth's crust. It is also a known neurotoxin and animal experiments have revealed neurofibrillary changes in the brains of rabbits, but often no or few plaques but these deposits may be species specific [20]. There has been scepticism as to how close this process resembles human AD, but there have been some immunohistochemical similarities demonstrated such as the presence of hyperphosphorylated tau in animal models [21,22]. The possible mechanisms suggested for aluminium neurotoxicity include: enhancing oxidative stress, interfering with gene expression, affecting cholinergic function, excitotoxicity, inducing apoptosis, affecting the inflammatory response, increasing oligomerization of Aβ, and inducing tangle formation [23,24]. Nevertheless, there is a body of opinion which argues strongly against the so-called 'aluminium hypothesis' in AD, stating lack of consistent reliable and reproducible evidence [25]. Unfortunately the debate often becomes overheated because it not only involves scientific issues but also encompasses environmental politics. An additional complication in this particular incident was that the sulphate component appeared to cause the water supply to become acidic (down to pH 3.9), which in turn (presumably by dissolving pipe linings) elevated the concentrations of other metals such as copper, iron and lead [9,26]. These metals were not measured in the brain tissue, but it is possible that they contributed to the neurodegenerative disease process since they all (especially lead) have been implicated as neurotoxins.

To our knowledge, this is only the second neuropathological description from a patient who had a neurodegenerative disease and had been a Camelford resident in 1988. It is very surprising that there have been so few such follow-up studies. Apart from the relatively early psychometric study described [10], there have to our knowledge been no full independent epidemiological analyses into the prevalence of early onset neurodegenerative diseases in this cohort compared to a similar rural population. Such investigations were recommended in the conclusions to the 2013 Government report [9]. To echo the appeal from the one previously published neuropathological case, it is important to undertake brain examination of others exposed to the drinking water in Camelford at that time, whenever there is consent and the opportunity arises. The unusual mixed pathology in the brain and elevated cerebral aluminium levels in this latest case certainly does not prove a role for aluminium in the pathogenesis of AD or dementia in general. Indeed the predominantly different pathologies in the two cases studied so far could argue against this. Similarly the APOE ε4/4 genotype in both could just point to an increased risk in developing sporadic AD. Nevertheless, one patient did not actually develop high stage AD, the other described here had a complex neuropathological profile and both had Lewy body pathology and elevated aluminium levels. It could therefore at least be argued that this case keeps the 'aluminium hypothesis' debate alive.

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Author contributions

AK and MA dissected the brain and examined the histology and immunohistochemistry. AK, CT, SAS, AH, MA and CE prepared the manuscript. CE and AM analysed the brain for aluminium. CT collected the clinical history prepared tissue for aluminium analysis. AH examined for AD mutations.

Ethical approval

There was ethical approval provided via the London Neurodegenerative Diseases Brain Bank (08/MRE09/38 + 5).

Conflict of interest

The authors declare they have no conflict of interest.

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References

1 Alafuzoff I, Arzberger T, Al-Sarraj S, Bodi I, Bogdanovic N, Braak H, Bugiani O, Del-Tredici K, Ferrer I, Gelpi E, Giaccone G, Graeber MB, Ince P, Kamphorst W, King A, Korkolopoulou P, Kovacs GG, Larionov S, Meyronet D, Monoranu C, Parchi P, Patsouris E, Roggendorf W, Seilhean D, Tagliavini F, Stadelmann C, Streichenberger N, Thal DR, Wharton SB, Kretzschmar H. Staging of neurofibrillary pathology in Alzheimer's disease: a study of the BrainNet Europe Consortium. *Brain Pathol* 2008; **18**: 484–96

2 Hyman BT, Phelps CH, Beach TG, Bigio EH, Cairns NJ, Carrillo MC, Dickson DW, Duyckaerts C, Frosch MP, Masliah E, Mirra SS, Nelson PT, Schneider JA, Thal DR, Thies B, Trojanowski JQ, Vinters HV, Montine TJ. National Institute on Aging-Alzheimer's Association guidelines for the neuropathologic assessment of Alzheimer's disease. *Alzheimers Dement* 2012; **8**: 1–13

3 McKeith IG, Dickson DW, Lowe J, Emre M, O'Brien JT, Feldman H, Cummings J, Duda JE, Lippa C, Perry EK, Aarsland D, Arai H, Ballard CG, Boeve B, Burn DJ, Costa D, Del ST, Dubois B, Galasko D, Gauthier S, Goetz CG, Gomez-Tortosa E, Halliday G, Hansen LA, Hardy J, Iwatsubo T, Kalaria RN, Kaufer D, Kenny RA, Korczyn A, Kosaka K, Lee VM, Lees A, Litvan I, Londos E, Lopez OL, Minoshima S, Mizuno Y, Molina JA, Mukaetova-Ladinska EB, Pasquier F, Perry RH, Schulz JB, Trojanowski JQ, Yamada M. Diagnosis and management of dementia with Lewy bodies: third report of the DLB Consortium. *Neurology* 2005; **65**: 1863–72

4 Mackenzie IR, Neumann M, Baborie A, Sampathu DM, Du PD, Jaros E, Perry RH, Trojanowski JQ, Mann DM. Lee VM A harmonized classification system for FTLD-TDP pathology. *Acta Neuropathol* 2011; **122**: 111–3

5 House E, Esiri M, Forster G, Ince PG, Exley C. Aluminium, iron and copper in human brain tissues donated to the medical research council's cognitive function and ageing study. *Metallomics* 2012; **4**: 56–65

6 Mirza A, King A, Troakes C, Exley C. The identification of aluminum in human brain tissue using lumogallion and fluorescence microscopy. *J Alzheimers Dis* 2016; **54**: 1333–8

7 Exley C, Esiri MM. Severe cerebral congophilic angiopathy coincident with increased brain aluminium in a resident of Camelford, Cornwall, UK. *J Neurol Neurosurg Psychiatry* 2006; **77**: 877–9

8 Poirier J, Davignon J, Bouthillier D, Kogan S, Bertrand P, Gauthier S. Apolipoprotein E polymorphism and Alzheimer's disease. *Lancet* 1993; **342**: 697–9

9 Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment. Subgroup Report on the Lowermoor Water Pollution Incident 2013. https://cot food gov uk/sites/default/files/cot/lwpiapp811 pdf

10 Altmann P, Cunningham J, Dhanesha U, Ballard M, Thompson J, Marsh F. Disturbance of cerebral function in people exposed to drinking water contaminated with aluminium sulphate: retrospective study of the Camelford water incident. *BMJ* 1999; **319**: 807–11

11 Murray V, Goodfellow F, Bogle R. Cerebral dysfunction after water pollution incident in Camelford. Inappropriate study, inappropriate conclusions. *BMJ* 2000; **320**: 1338

12 McLachlan DR, Bergeron C, Smith JE, Boomer D, Rifat SL. Risk for neuropathologically confirmed Alzheimer's disease and residual aluminum in municipal drinking water employing weighted residential histories. *Neurology* 1996; **46**: 401–5

13 Wettstein A, Aeppli J, Gautschi K, Peters M. Failure to find a relationship between mnestic skills of octogenarians and aluminum in drinking water. *Int Arch Occup Environ Health* 1991; **63**: 97–103

14 Burks JS, Alfrey AC, Huddlestone J, Norenberg MD, Lewin E. A fatal encephalopathy in chronic haemodialysis patients. *Lancet* 1976; **1**: 764–8

15 Wisniewski HM, Shek JW, Gruca S, Sturman JA. Aluminum-induced neurofibrillary changes in axons and dendrites. *Acta Neuropathol* 1984; **63**: 190–7

16 Moore PB, Day JP, Taylor GA, Ferrier IN, Fifield LK, Edwardson JA. Absorption of aluminium-26 in Alzheimer's disease, measured using accelerator mass spectrometry. *Dement Geriatr Cogn Disord* 2000; **11**: 66–9

17 Mirza A, King A, Troakes C, Exley C. Aluminium in brain tissue in familial Alzheimer's disease. *J Trace Elem Med Biol* 2017; **40**: 30–6

18 Saberzadeh J, Arabsolghar R, Takhshid MA. Alpha synuclein protein is involved in Aluminum-induced cell death and oxidative stress in PC12 cells. *Brain Res* 2016; **1635**: 153–60

19 Uversky VN, Li J, Fink AL. Metal-triggered structural transformations, aggregation, and fibrillation of human alpha-synuclein. A possible molecular NK between Parkinson's disease and heavy metal exposure. *J Biol Chem* 2001; **276**: 44284–96

20 Savory J, Herman MM, Ghribi O. Mechanisms of aluminum-induced neurodegeneration in animals: Implications for Alzheimer's disease. *J Alzheimers Dis* 2006; **10**: 135–44

21 Muma NA, Singer SM. Aluminum-induced neuropathology: transient changes in microtubule-associated proteins. *Neurotoxicol Teratol* 1996; **18**: 679–90

22 Singer SM, Chambers CB, Newfry GA, Norlund MA, Muma NA. Tau in aluminum-induced neurofibrillary tangles. *Neurotoxicology* 1997; **18**: 63–76

23 Maya S, Prakash T, Madhu KD, Goli D. Multifaceted effects of aluminium in neurodegenerative diseases: a review. *Biomed Pharmacother* 2016; **83**: 746–54

24 Kandimalla R, Vallamkondu J, Corgiat EB, Gill KD. Understanding aspects of aluminum exposure in Alzheimer's disease development. *Brain Pathol* 2016; **26**: 139–54

25 Lidsky TI. Is the aluminum hypothesis dead? *J Occup Environ Med* 2014; **56**: S73–9

26 Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment, Subgroup Report on the Lowermoor Water Pollution Incident 2013. Appendices 6 to 9. https://cot food gov uk/cotreports/cotwgreports/lowermoorreport

Supporting information

Additional Supporting Information may be found in the online version of this article at the publisher's web-site:

Table S1. Illustrating the immunohistochemical features in different regions of the brain in this case

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