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Neocortical levels of lithium are increased in bipolar disorder

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Salts of lithium, present in the human diet at trace levels,[1] are used to manage depressive, manic and psychotic symptoms in bipolar disorder and related conditions.[2,3] Although a neurological function for lithium was first proposed over 60 years ago based on its anti-manic properties,[2] surprisingly, a significant pool of lithium has never been identified in the brain.[4,5] In this study, with quadrupole inductively coupled plasma-mass spectroscopy, lithium is identified as a physiological trace element in the human neocortex. Moreover, cortical levels of lithium are found to be elevated in bipolar subjects with no past history of lithium pharmacotherapy.

The Harvard Brain Tissue and Resource Center at McLean Hospital provided fresh-frozen blocks of post-mortem neocortex (brodmann area 7) from 39 subjects, including 36 subjects from the McLean-66 cohort, with clinical and pathological diagnoses as described (http://national_databank.mclean.harvard.edu). Specimens derived from 15 subjects with bipolar disorder, 11 subjects with schizophrenia and 12 normal controls, matched for age, gender and post-mortem interval (Table 1). Neuropathology for all subjects was negative for an infectious, neurodegenerative or acute vascular process. For all psychiatric cases, no evidence was found for a past history of lithium treatment, as determined by a board-certified psychiatrist after review of all available medical records, laboratory results and questionnaires completed by next of kin. Although the psychopharmacologic histories were comprehensive, remote lithium trials could not be completely ruled out for psychiatric subjects, which is a limitation of the study.

In all, 200 to 300 μg wet-weight from each cortical specimen was transferred to a 1.5 ml polypropylene eppendorf tube. Plastic cutting surfaces, latex gloves, and stainless steel blades and spatulas that contacted specimens were thoroughly pre-rinsed with millipore water, and six empty eppendorf tubes were included as controls. Samples were prepared as described[6] and analyzed using an Agilent 7500CE quadrupole (Palo Alto, CA, USA) inductively coupled plasma-mass spectrometer (UC Davis, Mass Spectroscopy Core Facility). Samples were introduced with a MicroMist nebulizer (Glass Expansion, Pocasset, MA, USA) into a temperature-controlled spray chamber that was tuned to beryllium with the following settings: helium as the collision cell gas delivered at $4.6\,\mathrm{ml\,min^{-1}}$; radio frequency power of 1550 W; matching 1.6 V carrier gas at $1.03\,\mathrm{l\,min^{-1}}$; make-up gas at $0.17\,\mathrm{l\,min^{-1}}$; nebulizer pump at 0.11 r.p.s. Certiprep Me2, Sc and Y instrument and internal standards (SPEX CertiPrep Metuchen, NJ, USA) were analyzed from 0.25 ppb to 100 ppb. For quality control, NIST standard 1643E (Gaithersburg MD, USA) was analyzed at 10 and 100 ppb after every twelfth sample. Under these conditions, the detection limit for lithium was 3.89×10^{-3} ppb and the background equivalent concentration was 1.14×10^{-2} ppb. Statistical analysis was performed with one-way analysis of variance and Tukey's range test, after one subject from the bipolar group was excluded due to a lithium level greater than two standard deviations above the mean. Lithium levels are reported as $\mu g\,kg^{-1}$ dry-weight, with wet-weights used for calculating molar equivalence.

Cortical lithium levels (Figure 1) were elevated twofold ($P<0.01$) in bipolar disorder ($21.2 \pm 11.3\,\mu g\,kg^{-1}$) as compared with schizophrenic ($12.5 \pm 10.2\,\mu g\,kg^{-1}$) and normal controls ($11.5 \pm 5.3\,\mu g\,kg^{-1}$). The average lithium content across all 38 specimens analyzed was $15.2 \pm 10.27\,\mu g\,kg^{-1}$, with a molar equivalence of $0.38 \pm 0.24\,\mu M$.

The lithium levels reported in this study for the brain are comparable to plasma reference values[7] and three orders of magnitude lower than levels achieved in clinical pharmacology ($\sim$0.6 to 1.2 mEq/L).[8] As

Table 1 Investigated group characteristics

	Bipolar disorder	*Schizophrenia*	*Normal controls*	P
No. cases	15	11	12	—
Age, (years)	63.7 ± 18.2	59.2 ± 8.9	62.3 ± 14.9	n.s.[a]
PMI	22.6 ± 9.6	20.7 ± 4.4	19.1 ± 6.2	n.s.[a]
% male/female	60/40	73/27	70/30	n.s.[b]

Abbreviations: n.s. = non-significant ($P<0.05$); PMI = post-mortem interval.
Values are expressed as mean ± s.d. unless otherwise indicated.
[a]One-way ANOVA.
[b]χ^2-test.

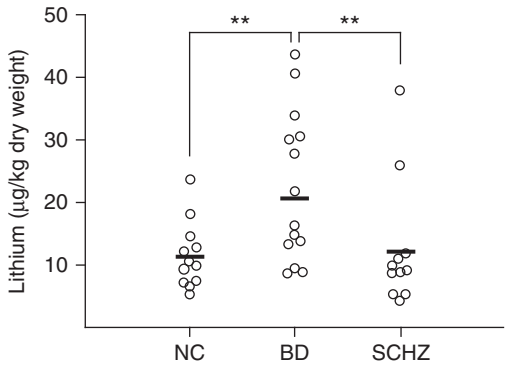

Figure 1 Neocortical lithium levels are elevated in bipolar disorder. Bars indicate the mean for each group, with each subject represented by an open circle. $^{**}P < 0.01$. Abbreviations: NC = normal control; BD = bipolar disorder; SCHZ = schizophrenia.

research into the neurobiology of lithium has been driven by its clinical success as a mood stabilizer, diverse biological effects of lithium are characterized at pharmacologically relevant concentrations.[8] In contrast, lithium has rarely been studied in its physiological concentration range, and no constitutive function is known.[3,8]

A functional role for lithium in the brain is supported in this study by the unexpected finding of elevated cortical lithium in bipolar disorder. To explain this finding and its possible significance, further study is required. In this report, the possibility is raised that cortical lithium may be elevated in bipolar disorder as a compensatory response against a deficient lithium-dependent pathway, underlying the disorder. Alternatively, elevated cortical lithium levels in bipolar disorder could be caused by pathological sequestration of lithium, with secondary deficiency in lithium bioavailability. Compensatory responses and abnormal metal sequestration are both common in neurological diseases.[9]

In summary, these data confirm lithium as a neurophysiological trace metal, and for the first time reveal that cortical lithium levels are elevated in bipolar disorder. Consequently, a role for lithium and possible substrates may need to be considered in the neurobiology of mood and cognition and the etiology of bipolar disorder.

Conflict of interest

The author declares no conflict of interest.

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CCDC22: a novel candidate gene for syndromic X-linked intellectual disability

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X-linked intellectual disability (XLID), defined as clinical ID combined with a pedigree consistent with X-linked inheritance, is a genetically heterogeneous condition that affects more than 10% of males with ID. Currently there are at least 92 genes known to cause XLID,[1–3] yet a large proportion of XLID cases remain unexplained, as each of the XLID genes identified so far only accounts for a small fraction (<1%) of affected individuals. Given that about one third of mutations affect gene expression levels,[4] we reasoned that transcriptome profiling of lymphoblast cell lines from XLID patients may highlight genes harboring disease-causing mutations and may be an efficient follow-up method for rare sequence variants of unknown functional significance.

We analyzed expression profiles of lymphoblast cell lines from 64 XLID patients, including 13 cases that were part of a recent X-chromosome exon re-sequencing study[5] (Supplementary Methods, Supplementary Table 1). We found polyglutamine-binding protein 1 (*PQBP1*), a gene previously implicated in XLID,[6,7] to be significantly downregulated in two cases (Supplementary Table 2), and confirmed an exon 4 (AG)2 deletion as the cause of mRNA downregulation in both instances. *PQBP1* mutations cause a sydromic form of XLID commonly referred to as Renpenning syndrome.[8] The specific mutation we describe here has been previously shown to cause XLID,[6] and has also been proven to decrease mRNA levels by nonsense-mediated mRNA decay,[7] thus being likely to be detected by assessment of gene expression. The cases for which we identified *PQBP1* mutations were not part of the cohort studied by Tarpey *et al.* (Supplementary Table 1).