Annette Heinrich

Molecular mechanisms of the effect of the mood stabilizer lithium on cAMP-induced CREB transcriptional activity

Lithium salts are used now for 60 years to treat bipolar disorder, a chronic, severe and often life-threatening illness characterized by recurrent phases of the two opposing mood states of depression and mania. The mechanisms accounting for the clinical efficacy are not completely understood. Chronic treatment with lithium is required to establish mood stabilization, suggesting the involvement of processes of neural plasticity. CREB (cAMP response element binding protein) is a transcription factor known to mediate neuronal adaptation. Recently, the CREB-coactivator TORC (transducer of regulated CREB) has been identified as a novel target of lithium and was shown to confer an enhancement by lithium of cAMP-induced CREB-directed gene transcription. TORC is sequestered in the cytoplasm and its nuclear translocation controls CREB activity by interacting with the basic leucine zipper of CREB. In the present study, the molecular mechanism was investigated by which lithium stimulates cAMP-induced CRE/CREB-mediated gene transcription through TORC1.

For this purpose, the effect of lithium on the nuclear localization of TORC was investigated in HIT-T15 cells by immunofluorescence and the effect on TORC transcriptional activity was examined by luciferase reporter-gene assays. The interaction between TORC and CREB was investigated in vitro in a GST pull-down assay, in vivo in a mammalian two hybrid assay and at the promoter by chromatin immunoprecipitation. Lithium did affect neither the nuclear translocation nor the intrinsic transcriptional activity of TORC proteins in HIT-T15 cells, but increased the oligomerization of TORC1 and the association of TORC with CREB. When heterologously expressed in HIT-T15 cells, all three human TORC isoforms mediated the lithium-induced enhancement of cAMP-dependent CRE/CREB-directed transcriptional activity. TORC1 was identified to be isoform predominantly expressed endogenously in HIT-T15 cells. As a cation, lithium facilitated directly the interaction between CREB and TORC1 in a concentration-dependent manner. The data support the notion that, once TORC has been shifted by cAMP into the nucleus, lithium facilitates the interaction between the CREB bZip and TORC1, thereby enhancing CREB-directed transcription. In contrast to lithium, magnesium strongly inhibited the CREB-TORC1 interaction which was attenuated by lithium. This effect could be considered as therapeutically relevant for the neuroprotective action of lithium possibly in that lithium counteracts excitotoxic effects of magnesium. The amino acid K290 of CREB, known to mediate the binding of a magnesium ion to the CREB bZip, appears not to be required for the effect of lithium as CREB-K290 mutants were inducible by lithium.

In addition to its action on artificial promoters, lithium was shown to enhance the cAMP-induced gene transcription at CRE/CREB-dependent human native promoters of the cfos-, BDNF(exonIV)- and NR4A2-genes through TORC1. Thus, a physiological relevance of the presently described effect is suggested and implicates a contribution of lithium to neuroplasticity. Taken together the results of the present study provide a novel mechanism of lithium action that may contribute to the therapeutic effect of lithium in the treatment of bipolar disorder.