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Effects of a Chronic Lithium Treatment on Central Dopamine Neurotransporters

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ABSTRACT. The objectives of this study were to characterize the effects of a chronic lithium (Li^+) treatment on dopamine (DA) uptake sites, as well as on the levels of mRNA encoding for these transporters, and to determine the eventual reversibility of the treatment. Quantitative autoradiography was carried out on sections from rat brain using 3β-(4-[^{125}I]iodophenyl)tropane-2β-carboxylic acid isopropyl ester ([^{125}I]RT1-121) to label DA transporters, and mRNA levels were measured by *in situ* hybridization. Following chronic Li^+ treatment (28 days), the labelling to DA transporters increased (60–90%) in all sections of the rostral and caudal neostriatum, whereas no alteration was observed in the other regions studied, namely the substantia nigra, the ventral tegmental area, and the dorsal raphe nucleus. These effects were reversed completely following a withdrawal period of 2 days without Li^+. Also, there were no modifications in the labelling of DA transporters after only 2 days of Li^+ treatment. In addition, we measured the levels of mRNA encoding for DA transporters in the substantia nigra and the ventral tegmental area; however, no alterations were observed following a chronic Li^+ treatment, and thus the hypothesis of an increased synthesis is not supported. This could mean that the Li^+ treatment modified the affinity of DA transporters for the radioligand, possibly a consequence of conformational changes induced by the disruption of the nerve terminal membrane environment; however, a modification in the number of transporters could not be ruled out. The results of this study further support the hypothesis of the implication of central dopaminergic transmission in the pathology and treatment of affective disorders. BIOCHEM PHARMACOL **54**;3:391–397, 1997. © 1997 Elsevier Science Inc.

KEY WORDS. lithium; dopamine; uptake sites; transporters; *in situ* hydridization; autoradiography

The role of central DA§ transmission in mood disorders is well established. Indeed, numerous studies have shown that this transmission is increased in mania, the selective stimulation of the central DA activity precipitating manic episodes in patients with bipolar disorder [1–4] and the reduction of this activity ameliorating the symptoms of mania [5–7], and is decreased in depression. However, the exact mechanism by which these alterations in DA synaptic activity are accomplished remains unclear. One psychoactive agent of therapeutic relevance in the treatment of affective disorders is Li^+, which was first introduced in psychiatry by Cade [8]. Many authors have demonstrated the efficiency of Li^+ in the treatment and prophylaxis of mania [9–11], and numerous studies have been carried out in an attempt to understand its mechanism of action. Schildkraut and colleagues [12, 13] have proposed, while investigating the effects of Li^+ on the turnover of monoamines, an important role for these neurotransmitters in psychiatric disorders, but the mechanisms underlying the therapeutic effects of Li^+ still remain elusive. Previous studies have documented effects of Li^+ on central DA transmission [14, 15]. More recently, we have shown that a chronic Li^+ treatment alters the coupling efficacy between DA D_1 and D_2 receptors and their associated G proteins, and the activity of the enzyme adenylyl cyclase stimulated by DA was found to be reduced by more than 50% in these conditions, suggesting a direct action by the treatment on the G proteins themselves [16]. It was, therefore, deemed of interest to further investigate the effects of a chronic Li^+ treatment on central DA transmission.

The inactivation of DA in the synapse is accomplished mainly by an uptake mechanism, and the biological structure responsible for this phenomenon is a DA transporter or DA uptake site. This transmembrane protein has been well characterized [17–20], and, fortunately, a ligand that binds very selectively and specifically to these transporters, namely [^{125}I]RT1-121, has been made available, permitting a detailed study of DA uptake sites [21, 22]. Moreover,

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§ *Abbreviations*: CSF, cerebrospinal fluid; DA, dopamine, dopaminergic; dRN, dorsal raphe nucleus; HVA, homovanillic acid; [^{125}I]RT1-121, 3β-(4-[^{125}I]iodophenyl)tropane-2β-carboxylic acid isopropyl ester; SN, substantia nigra; and VTA, ventral tegmental area.

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Giros and coworkers [23] have demonstrated, using a strain of mice lacking the DA transporter, that this protein is the most crucial component regulating DA transmission.

Therefore, the aim of the present study was to characterize the effects of a chronic Li^+ treatment on central DA neurotransmission, particularly by studying the transporters responsible for DA uptake, and to determine if the eventual effects could be reversed following a withdrawal for 48 hr from the treatment, i.e. without Li^+ administration. This was accomplished with the novel ligand [^{125}I]RT1-121 to label DA uptake sites in sections of rat brain, and by *in situ* hybridization experiments with a cRNA probe to investigate the rate of synthesis of DA transporters.

MATERIALS AND METHODS

Animals

This study was conducted with adult male Sprague–Dawley rats from Charles River (Montreal, Quebec), individually housed in clear plastic cages with wood-chip bedding, and maintained on a normal dark/light cycle. All the procedures concerning the use of animals were strictly in accord with the Canadian Council on Animal Care *Guide to the Care and Use of Experimental Animals*, and all the protocols were approved by the Comité de Déontologie de l'expérimentation animale of the Université de Montréal. The animals received LiCl orally (1 g/L) in their drinking water, for a period of 2 or 28 days. Such a chronic Li^+ treatment (28 days) results in an average concentration of about 1 mEq/L in plasma [24]. Following the 2 or 28 days of treatment with Li^+, with or without 48 hr of withdrawal from Li^+ administration, the rats were decapitated, and their brains were quickly removed and frozen at −80°. Since absolute values for the parameters studied in this report could change throughout the year due to seasonal variations, every group tested was compared with control animals specific for that group, i.e. each treated animal had a littermate as its control.

Autoradiographic Studies

The brains were cut serially with a cryostat; the 20-μm thick transverse sections were mounted on gelatin/chrome alum-coated slides, and kept at −80° until used. The autoradiographies were carried out as described previously [21, 22]. Briefly, the tissue sections (three slides per animal, i.e. two for total binding and one for the non-specific labelling) were preincubated for 30 min at 25° in a phosphate buffer (10.1 mM Na_2HPO_4, 1.8 mM KH_2PO_4, 137 mM NaCl, and 2 mM KCl at pH 7.4), and then were transferred to the incubation buffer containing 20 pM [^{125}I]RT1-121 (NEN/DuPont, Boston, MA; sp. act. 2200 Ci/mmol) for 60 min at room temperature. Non-specific binding was determined by adding nomifensine maleate (Hoechst-Roussel; 200 pM, final concentration). The slides were then washed in ice-cold buffer (2 × 20 min), rinsed in cold distilled water, and dried under a stream of air. The sections and [^{125}I]Microscales™ standards (Amersham, Arlington Heights, IL) were exposed to tritium-sensitive [^{3}H]Hyperfilm™ (Amersham). The films were developed after 40 hr of exposure. Slides from the same animals were stained with cresyl-violet to aid further in the localization of the anatomical regions.

Densitometric analyses of the films were performed with the MCID™ image analysis system (Imaging Research, Ste-Catherines, Ontario), and standard curves from the [^{125}I]Microscales™ were used to convert the optical density levels into femtomoles per milligram of protein. Anatomical structures were defined according to the atlas of Paxinos and Watson [25], and to direct observation of the cresyl-violet-stained sections. Multiple readings (10–40) were made for each region, depending on the extension of the anatomical structure considered, and taking into account the possible heterogeneity of labelling. The rostral neostriatum and the caudal portion of this structure were further arbitrarily subdivided into topographical zones. The location of the sampling areas is illustrated in Fig. 1.

Generation and Specificity of cRNA Probes to Be Used in the In Situ *Hybridizations*

A sequence encoding the entire rat dopamine transporter cDNA [17], provided by Dr. Marc Caron (Duke University, Durham, NC) was subcloned into the *Eco*RI site of pBluescript (Stratagene, La Jolla, CA). Uridine 5′-(α[^{35}S]thio) triphosphate-labelled antisense- or sense-strand RNA probes were prepared by *in vitro* transcription of linearized templates with the appropriate RNA polymerases, using the Riboprobe Gemini™ System II (Promega Corp., Madison, WI).

In Situ *Hybridization Histochemistry*

Brains were cut serially with a cryostat; the 20-μm thick transverse sections were mounted on superfrost-plus precleaned slides (Fisher Scientific, Ottawa, Ontario), and kept at −80° until used. The sections were air-dried and stored at room temperature overnight under vacuum with desiccant. The sections were fixed for 20 min at 4° in 4% formaldehyde prepared in 0.1 M sodium-phosphate buffer (PBS; pH 7.4), and then rinsed twice for 5 min in potassium-phosphate buffer (KPBS; pH 7.4) at room temperature. The slides were then immersed in a proteinase K solution (0.1 μg/mL) for 10 min at 37° and rinsed briefly in diethyl pyrocarbonate (DEPC)-treated water. The slides were incubated in a solution of 0.1 M triethanolamine (pH 8.0) for 3 min at room temperature. Then the slides were incubated in a fresh solution of 0.25% acetic anhydride in 0.1 M triethanolamine (pH 8.0) for 10 min at room temperature. The slide-mounted sections were rinsed for 5 min in 2× SSC (0.15 M NaCl/0.015 M sodium citrate), dehydrated in a series of ascending concentrations of ethanol, air-dried, and stored at room temperature for 1–2 hr under vacuum with desiccant.

For *in situ* hybridization, ^{35}S-labelled RNA probes for the

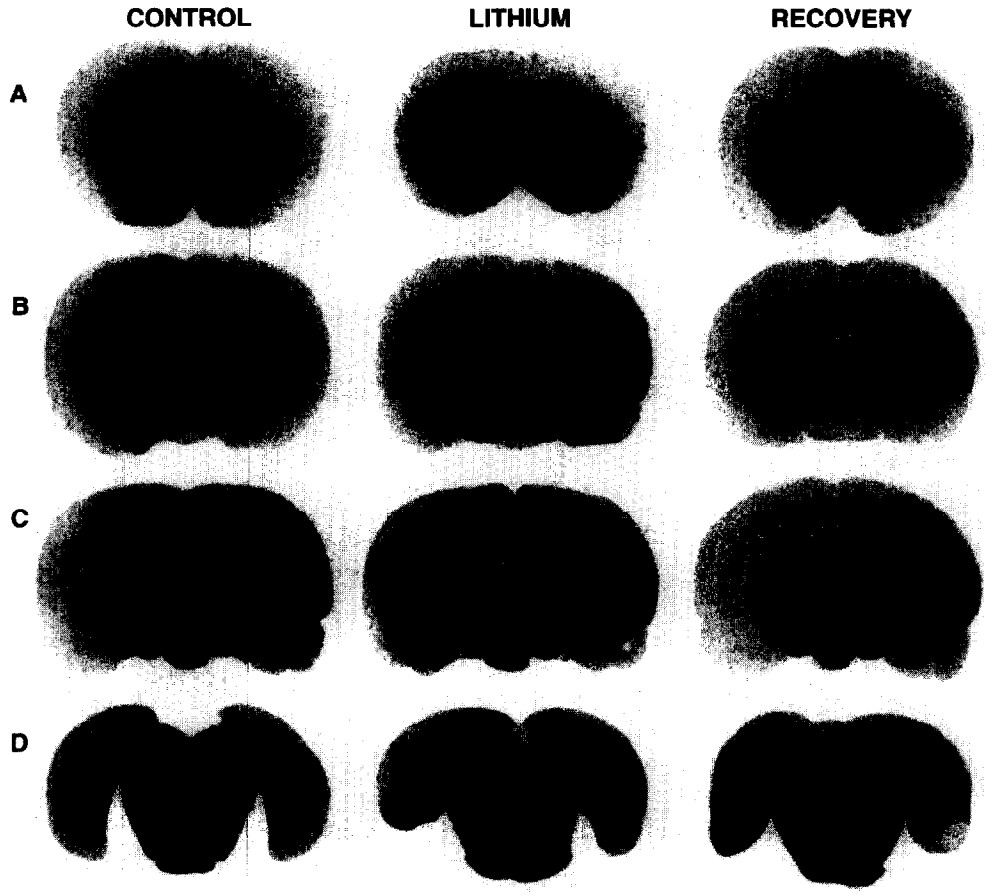

FIG. 1. Autoradiograms of total [^{125}I]RT1-121 labelling in coronal sections of brains from a control rat (left panels), an animal treated chronically (28 days) with lithium (center panels), and an animal that recovered for 48 hr from the chronic (28 days) lithium treatment. (A) Sections obtained from the most rostral portion of the brains, showing the labelling in the rostral neostriatum, that was divided into four quadrants: medio-dorsal, medio-ventral, latero-dorsal, and latero-ventral. (B) Sections showing the caudal neostriatum, divided into dorsal and ventral portions. Note in the neostriatum the dorsal to ventral, as well as the medial to lateral, increasing gradients of [^{125}I]RT1-121 labelling. (C) Sections illustrating the mesencephalon and the labelling in the substantia nigra and in the ventral tegmental area. (D) In these caudal sections of the midbrain, note the faint [^{125}I]RT1-121 labelling of the dorsal raphe.

dopamine transporter were added to the hybridization buffer (1.5×10^7 cpm/mL, final concentration). The hybridization buffer was composed of 50% formamide, 10% dextran sulfate, 0.6 M NaCl, 10 mM Tris (pH 7.5), 1 mM EDTA (pH 8.0), 2.5× Denhardt's solution, 50 μg/mL yeast tRNA, 150 μg/mL total RNA, 0.1 mg/mL denatured salmon sperm DNA, and 10 mM dithiothreitol. One hundred microliters of hybridization buffer containing the labelled probe was added to each slide (four sections per slide). Then the sections were covered with glass coverslips, and incubated at 55° for 15 hr. After the incubation, the coverslips were floated off in 4× SSC at room temperature so that the slide-mounted sections could be rinsed briefly in 2× SSC at room temperature. The slides were washed for 1 hr in 2× SSC at 50°, later treated with ribonuclease A at 20 μg/mL in a Tris–EDTA buffer for 1 hr at 37°, and then washed again for 1 hr in 2× SSC at 50°. Next, the slides were washed in a high-stringency solution composed of 0.1× SSC, 13 mM 2-mercaptoethanol, and 0.05% sodium pyrophosphate for 2–3 hr at 60°. The slides were then air-dried and dehydrated in a series of ascending concentrations of ethanol. The slide-mounted tissue sections were exposed to Kodak Biomax™ films for 3.5 days at room temperature. The films were developed, and the autoradiograms were analyzed by densitometry, using the program NIH Image 1.58 on a Power Macintosh™ 7100 with a constant source of illumination, and a standard grayscale strip was used to generate a calibration curve for optical densities.

Levels of mRNA for the dopamine transporter were measured in the substantia nigra pars compacta and in the ventral tegmentum area; these regions were each divided into left and right axes. Data were computed separately for each region analyzed, and the levels of dopamine transporter mRNA were expressed as optical densities.

TABLE 1. Regional distribution of the binding of [^{125}I]RT1-121 on cerebral slices from control and Li^+-treated rats (2 days)

Region	[^{125}I]RT1-121 binding (fmol/mg protein)	
	Controls	Treated
Medio-dorsal rostral neostriatum	3.6 ± 0.3	4.4 ± 0.5
Medio-ventral rostral neostriatum	4.3 ± 0.5	4.2 ± 0.4
Latero-dorsal rostral neostriatum	4.1 ± 0.4	5.0 ± 0.6
Latero-ventral rostral neostriatum	4.6 ± 0.6	5.2 ± 0.6
Dorso-caudal neostriatum	3.3 ± 0.6	3.3 ± 0.4
Ventro-caudal neostriatum	4.3 ± 0.7	4.5 ± 0.6
Ventral tegmental area	5.2 ± 0.5	5.3 ± 0.5
Substantia nigra	4.2 ± 0.4	3.8 ± 0.5
Dorsal raphe nucleus	2.1 ± 0.3	2.2 ± 0.4

Values are means ± SEM of 4 animals.

TABLE 2. Regional distribution of the binding of [^{125}I]RT1-121 on cerebral slices from control and Li^+-treated rats (28 days)

Region	[^{125}I]RT1-121 binding (fmol/mg protein)	
	Controls	Treated
Medio-dorsal rostral neostriatum	2.9 ± 0.2	5.1 ± 0.4*
Medio-ventral rostral neostriatum	3.3 ± 0.2	5.8 ± 0.6*
Latero-dorsal rostral neostriatum	3.2 ± 0.3	5.3 ± 0.4*
Latero-ventral rostral neostriatum	3.7 ± 0.4	6.7 ± 0.5*
Dorso-caudal neostriatum	2.7 ± 0.4	5.1 ± 0.2*
Ventro-caudal neostriatum	4.0 ± 0.1	6.5 ± 0.6*
Ventral tegmental area	5.5 ± 1.1	5.7 ± 1.1
Substantia nigra	4.3 ± 0.6	3.9 ± 0.9
Dorsal raphe nucleus	1.7 ± 0.4	1.8 ± 0.4

Values are means ± SEM of 3–4 animals. The significant differences were determined with Student's *t*-test from the program T&ANOVAR [26].
*$P < 0.01$, compared with controls.

Statistical Evaluation

Group comparisons between control and treated animals were assessed by means of Student's *t*-test with the program T&ANOVAR [26].

RESULTS

Effects of Lithium Treatments on the Regional Distribution of [^{125}I]RT1-121 in the Rat Brain

The use of [^{125}I]RT1-121, considered to be a very selective and specific marker of DA transporters [21, 22], permitted a very detailed mapping of central dopaminergic neurons and terminal fields of innervation (Fig. 1), and the density of labelling values for the different brain regions examined are given in Tables 1–3. As expected, the highest densities were observed in regions rich in DA nerve endings or in DA neurons; thus, we concentrated our study on the rostral and caudal neostriatum (further divided arbitrarily into topological zones), the SN, the VTA, and the dRN. In this latter region, labelling presented the greatest degree of variability, and the optical density readings were very low and close to the limit of quantification, once non-specific labelling had been subtracted from total binding. In this context, we can assume that there could have been an overestimation of the density of [^{125}I]RT1-121 labelling for this region. As illustrated in Table 2, chronic Li^+ treatment (28 days) dramatically increased the binding of [^{125}I]RT1-121 in all divisions of the neostriatum (60–90%), without affecting the DA uptake sites in the SN, VTA, and dRN. Moreover, as can be seen in Tables 1 and 3, this effect was not observed after only 2 days of treatment (Table 1), and was reversed back completely to control conditions when the animals were allowed to recover for 48 hr from the chronic treatment, without Li^+ administration (Table 3).

TABLE 3. Regional distribution of the binding of [^{125}I]RT1-121 on cerebral slices from control and Li^+-treated rats (28 days) that recovered for 48 hr from the treatment

Region	[^{125}I]RT1-121 binding (fmol/mg protein)	
	Controls	Treated
Medio-dorsal rostral neostriatum	3.5 ± 0.7	3.7 ± 0.2
Medio-ventral rostral neostriatum	4.0 ± 0.8	4.1 ± 0.4
Latero-dorsal rostral neostriatum	3.9 ± 0.9	4.2 ± 0.3
Latero-ventral rostral neostriatum	4.7 ± 1.0	4.6 ± 0.5
Dorso-caudal neostriatum	3.3 ± 0.8	3.2 ± 0.3
Ventro-caudal neostriatum	5.7 ± 1.1	4.9 ± 0.5
Ventral tegmental area	4.2 ± 0.2	3.4 ± 0.3
Substantia nigra	3.4 ± 0.3	2.7 ± 0.6
Dorsal raphe nucleus	1.8 ± 0.3	2.0 ± 0.7

Values are means ± SEM of 3–4 animals.

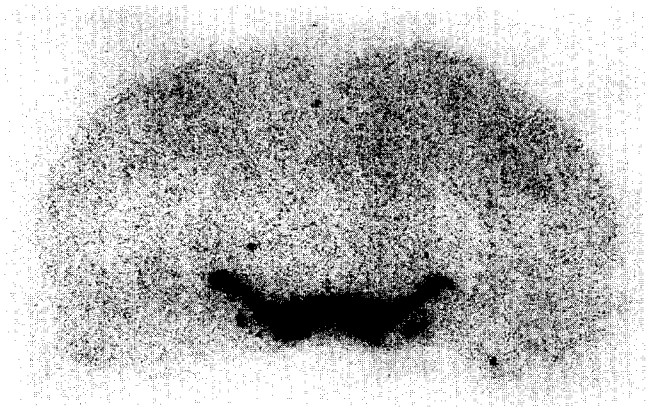

FIG. 2. Autoradiogram of *in situ* hybridization from a control animal which reveals the marked expression of the DA transporter mRNA in the mesencephalic substantia nigra and in the ventral tegmental area; the labelling in the former region is concentrated in the pars compacta, and no labelling could be attributed to the pars reticulata.

Effects of a Chronic Lithium Treatment on the Levels of mRNA for the Dopamine Transporter

The *in situ* hybridization analysis revealed a marked expression of the DA transporter mRNA in the VTA, as well as in the SN; the labelling in the latter region was concentrated in the pars compacta, and no labelling could be attributed to the pars reticulata (Fig. 2). As can be seen in Table 4, a chronic Li^+ treatment did not modify the levels of mRNA encoding for the DA transporter in these two anatomical regions.

TABLE 4. Effects of chronic Li^+ treatment (28 days) on the mRNA tissue levels encoding for the dopamine transporter in substantia nigra (SN) and ventral tegmental area (VTA)

Treatment	Optical density	
	SN	VTA
Controls	0.48 ± 0.03	0.40 ± 0.02
Treated	0.44 ± 0.04	0.39 ± 0.03

Values are means ± SEM of 5 animals.

DISCUSSION

The aim of this study was to further examine the effects of a chronic Li^+ treatment on central DA neurotransmission, as well as the possible reversibility of eventual changes. First, we studied the effects of chronic Li^+ on the density of labelling to DA uptake sites with quantitative autoradiography, using the highly specific ligand [^{125}I]RT1-121. We focused our survey on the rostral and caudal neostriatum, subdivided into six topological zones, since this structure comprises the majority of nerve terminals arising from central DA projections, as well as on SN, the VTA, and the dRN; these latter regions enclose cell bodies of DA neurons. The chronic administration of Li^+ (28 days) was shown to increase dramatically the labelling to DA transporters in all divisions of caudal and rostral neostriatum (60–90%), but no alterations were observed in the regions containing the majority of cell bodies of central DA neurons, namely in SN, VTA, and dRN (Table 2). Increases in DA uptake following a chronic Li^+ treatment have been suggested previously [27], and it has been shown recently that Li^+ chloride could stimulate *in vitro* the DA uptake in PC12 cells [28]. However, important discrepancies in methodology between these studies and ours prevent us from establishing tighter correlations. These findings are in accord with previous reports suggesting that the central DA activity is increased in mania [1–7]. Moreover, the concentration of HVA, the major metabolite of DA, has been shown previously to be increased in the CSF of patients with mania [29], which was expected in light of an increased DA activity in mania. We have also shown that a chronic Li^+ treatment decreases the concentration of HVA in the neostriatum of treated animals [16]. Since HVA is a metabolite produced in the extracellular space, the observed increase in DA uptake could explain the decrease in HVA levels induced by chronic Li^+, as well as a possibly deficient DA uptake in manic patients leading to increased HVA levels in the CSF. Taken together, these results suggest that the role of Li^+ in mania would be to bring the hyperactive DA activity back to normal levels.

It was essential at this point to determine if the effects of Li^+ on central DA uptake sites were already present after a very short duration of Li^+ administration, since its therapeutic effects have been shown to appear only after a relatively long period of time (14–21 days). Interestingly, no alterations in the labelling of DA neurotransporters were observed after only 2 days of Li^+ treatment (Table 1), further supporting the temporal course of effects attributed to Li^+, and outlining the therapeutic relevance of the results reported in this study.

Another interesting aspect that we examined was the possible reversibility of the observed effects. Indeed, it is well established that patients suffering from bipolar disorder who are treated with Li^+ must take this medication constantly; the withdrawal of Li^+ leads to a recurrence of severe manic symptoms [30]. In this context, if the DA uptake sites are implicated in the pathophysiology and/or treatment of the disorder, it would be logical to expect that the observed modifications on the density of labelling to DA transporters will be reversed following the end of the treatment. Consequently, we treated rats for 28 days with Li^+, followed by a withdrawal period of 48 hr without Li^+ administration. As can be observed in Table 3, the dramatic increase in the striatal density of labelling to DA uptake sites observed following a chronic Li^+ treatment was no longer present after the withdrawal period. It was reported previously that Li^+ is cleared very rapidly, and its plasma levels are very low 1 day after its administration is stopped [24]; thus, the rapid reversibility of the observed effects suggests that they require the constant presence of this ion, which is in accord with clinical observations. This rapid reversibility further supports the possible implication of the central DA transmission in affective disorders.

The technique of quantitative autoradiography has certain limitations. Indeed, since the density of transporters has been evaluated with a fixed concentration of radioligand, it is not possible to determine if this increase in binding is a consequence of a modification in the affinity of the uptake sites for [^{125}I]RT1-121, or of a change in the number of transporters present in the preparation. Indeed, the concentration used in the present study was 20 pM, which is lower than the reported K_D for this ligand in the rat striatum [21]. Therefore, we evaluated with *in situ* hybridization experiments the effects of a chronic Li^+ treatment on the levels of the mRNA encoding for the DA neurotransporter. As can be seen in Table 4, the chronic treatment did not influence the mRNA levels measured in the SN and in the VTA, thus ruling out the possibility of an increased synthesis to account for the elevation in DA transporter labelling. This could mean that the affinity of transporters has been modified by the chronic treatment; however, a change in transporter number could not be ruled out.

The mechanistic explanation of the observed effects remains to be clarified. However, as previously discussed [16, 31], it has been shown that a chronic Li^+ treatment leads to a disorganization of the neuronal plasma membranes [32]. In fact, it was demonstrated that such a treatment alters the lipid composition as well as the fluidity of the terminal neuronal membranes. Since the maintenance of the membrane environment is essential for the proper functioning of a number of cellular processes, it can be speculated that a chronic Li^+ treatment modifies the lipid-protein moiety required for the optimal activity of central DA transporters, leading to structural alterations of these proteins, rendering them less efficient in the accomplishment of their physiological function. This hypothesis is in line with our proposal that the elevated density of labelling to DA transporters is a consequence of an increase in transporter affinity. Indeed, we can also suggest that the modifications in membrane environment lead to a conformational change in the DA uptake sites, thus inducing an alteration in affinity. However, if the number of transport-

ers has been modified by the treatment, which could not be excluded, we can only postulate that [^{125}I]RT1-121 binds to some sites that were not available previously. This could suggest that a pool of transporters exists in the vicinity of the synapse, therefore providing the basis for a rapid regulation of these proteins without modifications of their synthesis.

Interestingly, this effect of chronic Li^+ on neuronal membranes appears to be observable only at the nerve terminals. Indeed, the results showing that the up-regulation of DA uptake sites is located in the neostriatum only suggest that the membranes of the dendrites and of cell bodies of DA neurons, located primarily in the SN, VTA, and dRN, are resistant to such a membrane disruption.

Another element giving further support to the stated hypothesis, namely that Li^+ achieves its therapeutic effects through a disruption of the terminal membranes, comes from a previous report from our laboratory [16]. In that study, we demonstrated that a chronic Li^+ treatment leads to a decrease in the function of the DA D_1 and D_2 receptors, by affecting directly the G proteins associated with these receptors. This observation is in accord with the hypothesis of Li^+ decreasing the central DA activity back to normal conditions in manic patients. However, since the DA neurotransporter has been demonstrated to be the most crucial component in regulating the synaptic DA activity [23], it would be expected, following classic theories of receptor regulation, that the function of DA receptors will be increased to compensate for the lower levels of DA available in the synapse after the observed up-regulation of the DA uptake sites. Since this expected situation does not occur after chronic Li^+ administration, we can speculate that Li^+ is able to circumvent the expected compensation by indirectly affecting the transmembrane proteins implicated in the DA circuitry via a disruption of the terminal membranes. Therefore, Li^+ could be able to produce a dramatic and long-lasting decrease of central DA transmission, and consequently exert its antimanic effect in accord with the clinical observations. Moreover, since a hypothesis of the mechanism of the switch from depression to mania relies on the capacity of the post-synaptic DA receptors to be normally regulated [33], we can speculate that Li^+ prevents this regulation, permits the maintenance of DA activity in a normal state and, consequently, prevents the switch to a depressive state.

In conclusion, a chronic lithium treatment altered striatal dopaminergic activity, mainly through a direct action on DA transporters. Indeed, this study suggests a dramatic increase in the density of labelling to DA uptake sites, which is in accord with the clinical observations in patients with mania. However, this up-regulation is not correlated with an increase in transporter synthesis, suggesting that the increase is probably a consequence of an elevation in the affinity of the uptake sites. Moreover, the rapid reversibility of the observed effects further supports the hypothesis of the implication of central DA neurotransmission in the treatment and pathophysiology of the human affective disorders.

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