Pharmacology Biochemistry & Behavior, Vol. 26, pp. 235–240. Copyright © Pergamon Journals Ltd., 1987. Printed in the U.S.A. 0091-3057/87 $3.00 + .00

Mechanism of Lithium Action: *In Vivo* and *In Vitro* Effects of Alkali Metals on Brain Superoxide Dismutase

GIRJA S. SHUKLA[1]

Department of Pharmacology, Faculty of Health Sciences, School of Medicine
University of Ottawa, Ottawa, Ontario, Canada K1H 8M5

Received 12 March 1984

SHUKLA, G. S. *Mechanism of lithium action:* In vivo *and* in vitro *effects of alkali metals on brain superoxide dismutase.* PHARMACOL BIOCHEM BEHAV **26**(2) 235–240, 1987.—Intraperitoneal administration of lithium (2 mEq/kg/day) was found to increase the superoxide dismutase (SOD) activity in certain brain regions after 24 hours (2 injections) and 3 days (once a day) of exposure. *In vitro* addition of wide range of lithium (0.1 to 8 mEq) to enzyme preparation as well activated cortical SOD activity; however, at 10 mEq concentrations an inhibition was observed. The increase in SOD activity did not appear to be region specific as under both *in vivo* and *in vitro* conditions lithium enhanced enzyme activity in all the tested brain regions. The effects of intraperitoneal administration of 2 mEq/kg rubidium and cesium for 24 hr (2 injections) and 6 days (once a day) were also studied on central SOD. Both the alkali metals were not found to produce any significant alteration in the cortical enzymic activity. When the *in vitro* effects of these monovalent alkali metals were tested, only 2 mEq rubidium was found to increase cortical SOD; however, cesium and potassium at similar concentration did not produce any appreciable effects. It appears from the data that lithium-induced increase in brain SOD activity is not an unspecific effect of alkali metals. SOD enzyme disposes cytotoxic superoxide radicals which, if not removed, could impair the normal functioning of cellular membrane and produce a variety of psychedelic compounds as well. The activation of central SOD by lithium would enhance the disposal process of superoxide radicals whose pathological concentrations may be present in affective disorders. The mechanism of lithium-induced activation of SOD, at present, is not known.

Lithium Mechanism Rat brain Superoxide dismutase Free radicals Catecholamine oxidation Membrane Rubidium Cesium

LITHIUM salts are the most frequently used drugs in the treatment of mania and in the prophylactic treatment of recurrent, bipolar manic-depressive psychosis [26]. The general biology and pharmacology of lithium have been extensively studied and its mechanism of action as a mood stabilizing agent has been investigated to some extent. Much attention has centered on the interactions of lithium with various neurotransmitter systems that have been implicated in the pathophysiology of mood disorders [1, 10, 12]. Additionally, lithium has been shown to affect membrane related phenomena such as electrolyte transport and balance, uptake and release of cerebral transmitter amines as well as of their precursors [13,14]. Despite much effort, the exact mechanism of action of lithium still remains unknown.

Brain tissue is highly supplied with oxygen and is rich in oxidizable substrates such as catecholamines and unsaturated lipids. The peroxidation of membranal lipids by a highly reactive free radical such as superoxide (O_2^-) has been reported to change the activities of membrane bound enzymes and receptors, which could result in altered membrane permeability and neural transmission [9, 12, 23]. Therefore, the components of external and intracellular membrane need to be protected from oxidative attack for the normal functioning of a nerve cell. Moreover, the oxidation products of catecholamines like adrenochrome have been shown to produce psychedelic response in experimental animals and healthy volunteers [7]. Excessive levels of free radicals that result either from a high rate of their generation or slower disposition through decreased superoxide dismutase (SOD, E.C. 1.15.1.1) activity could have an important role in the etiology of affective disorders. SOD, which plays a prominant role in the inactivation of O_2^-radicals formed during the metabolic reduction of molecular oxygen, thus provides an important protective mechanism against oxidative damage of the tissues [3,4]. The present investigation was conducted to study the *in vivo* and *in vitro* effects of lithium on regional SOD activity in rat brain. The effects of other alkali metals, rubidium and cesium, were also studied for comparison.

METHOD

Animals and Treatment

Male Sprague-Dawley rats (150–175 g) were housed in groups of three per cage in an environment of controlled

[1]Present address: Dr. Girja S. Shukla, Industrial Toxicology Research Centre, Post Box NO. 80, Lucknow-226 001, India.

TABLE 1

EFFECT OF SHORT-TERM ADMINISTRATION OF Li_2CO_3 ON SUPEROXIDE DISMUTASE* OF DIFFERENT REGIONS OF RAT BRAIN

Region	Control§	Li_2CO_3-Treated 12 Hr†	Percent of Control	12+12 Hr‡	Percent of Control
Cerebral cortex	1.44 ± 0.101	1.56 ± 0.099	109	1.81 ± 0.110	126¶
Cerebellum	1.73 ± 0.106	1.83 ± 0.121	106	2.02 ± 0.116	117
Corpus striatum	1.56 ± 0.098	1.60 ± 0.101	103	1.90 ± 0.108	122¶
Hippocampus	1.49 ± 0.093	1.46 ± 0.114	98	1.80 ± 0.101	121¶
Hypothalamus	1.91 ± 0.113	2.10 ± 0.123	110	2.33 ± 0.117	122¶
Mid-brain	2.16 ± 0.104	2.33 ± 0.111	108	2.57 ± 0.124	119¶
Pons-medulla	2.23 ± 0.110	2.32 ± 0.121	104	2.59 ± 0.120	112

The values represent arithmatic mean ± S.E. of 6 rats in each group.

*Activity represented as units/mg protein—one unit of SOD activity is defined as the amount of enzyme which inhibited the reaction by 50% [19].

†Li_2CO_3 (2 mEq lithium/kg) injected IP in a volume of 0.5 ml.

‡One injection was followed by another after 12 hr.

§Control animals received an equivalent amount of NaCl.

¶Significantly different ($p<0.05$) compared to control.

TABLE 2

EFFECT OF Li_2CO_3 ADMINISTRATION ON SUPEROXIDE DISMUTASE* OF DIFFERENT REGIONS OF RAT BRAIN

Treatment† Period (Days)	Cerebral Cortex	Cerebellum	Corpus Striatum	Hippocampus	Hypothalamus	Mid-Brain	Pons-Medulla
Control	1.52 ± 0.093	1.69 ± 0.101	1.61 ± 0.110	1.41 ± 0.090	1.79 ± 0.103	2.24 ± 0.110	2.29 ± 0.106
Li_2CO_3							
1	1.66 ± 0.121	1.74 ± 0.111	1.80 ± 0.121	1.73 ± 0.121	1.88 ± 0.111	2.52 ± 0.131	2.61 ± 0.126
3	1.83 ± 0.101 (121‡)	1.82 ± 0.117	1.99 ± 0.101 (124‡)	1.68 ± 0.081 (119‡)	1.83 ± 0.109	3.02 ± 0.121 (120#)	2.84 ± 0.131
6	1.94 ± 0.106 (128§)	2.01 ± 0.102 (119‡)	1.98 ± 0.113 (123‡)	1.75 ± 0.116 (124‡)	2.16 ± 0.114 (121‡)	2.75 ± 0.118 (123§)	2.70 ± 0.108 (118‡)
9	1.99 ± 0.117 (131§)	2.03 ± 0.107 (120‡)	2.08 ± 0.109 (129‡)	1.76 ± 0.109 (125#)	2.15 ± 0.118 (120‡)	2.78 ± 0.121 (124#)	2.80 ± 0.114 (122¶)
12	2.06 ± 0.121 (136¶)	2.06 ± 0.110 (122‡)	2.06 ± 0.113 (128‡)	1.79 ± 0.116 (127‡)	2.22 ± 0.122 (124‡)	2.73 ± 0.118 (122§)	2.77 ± 0.118 (121§)

The values represent arithmatic mean ± S.E. of 6 rats in each group. The data in parentheses are percent compared to control as 100%.

*Activity represented as units/mg protein—one unit of SOD activity is defined as the amount of enzyme which inhibited the reaction by 50% [19].

†Li_2CO_3 (2 mEq lithium/kg/day) injected IP in a volume of 0.5 ml; control animals received an equivalent amount of NaCl.

Significantly different compared to control, ‡$p<0.05$, §$p<0.02$, ¶$p<0.01$, #$p<0.001$.

temperature (24±1°C) and alternating 12 hours light (0600–1800 hr) and dark cycles. The rats were allowed free access to food and water at all times.

In one set of the experiments, twelve rats were administered with lithium carbonate (Li_2CO_3, adjusted to pH 7.4 with citric acid), lithium chloride (LiCl), rubidium chloride (RbCl) or cesium chloride (CsCl) intraperitoneally (2 mEq alkali metal/kg body weight) in a volume of 0.5 ml. After 12 hr, half of the injected animals were sacrificed by decapitation while the rest of them were given one more injection of 2 mEq/kg of respective alkali metal and sacrificed after another 12 hr.

The other set of animals received daily one intraperitoneal injection of 2 mEq/kg body weight of the above mentioned metals. Six rats from Li_2CO_3 treated groups were randomly selected and sacrificed each after 1, 3, 6, 9 and 12 days of exposure. However, the rats receiving alkali metal halides were sacrificed only after 6 days of treatment period.

The animals of respective control groups were treated with an equivalent amount of sodium chloride (NaCl) in a similar manner.

Sample Preparation and SOD Assay

The brains were rapidly excised, placed on a chilled glass plate and, after removing grossly visible blood vessels, dis-

TABLE 3

IN VITRO EFFECTS OF LITHIUM ON AUTO-OXIDATION OF PYROGALLOL AND SUPEROXIDE DISMUTASE ACTIVITY OF RAT CEREBRAL CORTEX

Conc. of Lithium (mEq)*	Auto-Oxidation of Pyrogallol (Δ Optical Density/min)†	Percent of Control	Superoxide Dismutase (Units/mg Protein)‡	Percent of Control
Control	0.214 ± 0.021	100	1.43 ± 0.090	100
0.10	0.215 ± 0.022	100	1.46 ± 0.092	102
0.50	0.212 ± 0.021	99	1.51 ± 0.093	106
1.00	0.210 ± 0.023	98	1.58 ± 0.072	111
1.50	0.209 ± 0.019	98	1.66 ± 0.083	116
2.00	0.208 ± 0.020	97	1.87 ± 0.132	131
4.00	0.207 ± 0.022	97	1.83 ± 0.120	128
6.00	0.205 ± 0.021	96	1.76 ± 0.108	123
8.00	0.205 ± 0.019	96	1.70 ± 0.081	119
10.00	0.204 ± 0.021	95	1.10 ± 0.079	77

The values represent arithmatic mean ± S.E. of 3 experiments, each done in duplicate.

*Li_2CO_3 was added in incubation mixture.

†Optical density/min was recorded without homogenate; however, volume was made up with distilled water.

‡One unit of SOD activity is defined as the amount of enzyme which inhibited the reaction by 50% [19].

sected into various regions [6]. Each brain region was homogenized in 20 volumes of triple glass distilled water using glass homogenizer with teflon pestle fitted to a motor drive. Following centrifugation of homogenate at 1,000 × g for 10 minutes at 4°C, SOD activity in the supernatant was measured according to the method of Marklund and Marklund [18] with certain modifications [19].

Briefly, the assay mixture in a total volume of 1.0 ml consisted of Tris-cacodylate buffer, pH 8.2 (50 mM), nitroblue tetrazolium (0.1 mM) Triton X-100 (0.001%), pyrogallol (0.2 mM), enzyme preparation and where needed, appropriate amounts of alkali metal. In the blank, enzyme was substituted by an equal amount of water. The activity was measured at 24°C at the wavelength of 540 nm, using a Beckman DU-7 spectrophotometer. The cuvettes were washed after every reading with concentrated nitric acid followed by several rinses of distilled water to remove the traces of alkali metal sticking to the glass wall. One unit of SOD activity was defined as the amount of enzyme which inhibited the auto-oxidation of pyrogallol by 50% under the given experimental conditions [19].

Protein was estimated using the Folin phenol reagent method [16] with bovine serum albumin as the standard.

Statistical Analysis

The significance of difference between mean values was calculated by using Student's t-test. p-Values less than 0.05 were considered to be significant.

Materials

Rubidium chloride, cesium chloride and nitroblue terazolium were purchased from Sigma Chemical Co., St. Louis, MO. Pyrogallol was obtained from J. T. Baker Chemical Co., Phillipsburg, NJ. All other reagents used in this study were from Fisher Scientific Co., Fairlawn, NJ or of the purest grade available in the laboratory.

TABLE 4

IN VITRO EFFECTS OF Li_2CO_3 ON SUPEROXIDE DISMUTASE ACTIVITY IN VARIOUS BRAIN REGIONS

Brain Regions	Superoxide Dismutase (Units/mg Protein)*		Percent of Control
	Control	Li_2CO_3†	
Cerebral cortex	1.30 ± 0.10	1.74 ± 0.11	134
Striatum	1.41 ± 0.09	1.76 ± 0.12	125
Mid-brain	2.01 ± 0.12	2.64 ± 0.14	122
Hippocampus	1.42 ± 0.08	1.65 ± 0.06	116
Pons-medulla	2.23 ± 0.14	2.59 ± 0.12	116
Hypothalamus	2.11 ± 0.13	2.41 ± 0.16	114
Cerebellum	1.60 ± 0.11	1.76 ± 0.08	110

The values represent arithmatic mean ± S.E. of 3 experiments, each done in duplicate.

*One unit of SOD activity is defined as the amount of enzyme which inhibited the reaction by 50% [19].

†Two mEq lithium was added in incubation mixture.

RESULTS

The effects of acute treatment of Li_2CO_3 on SOD activity of different brain regions are presented in Table 1. The administration of Li_2CO_3 (2 mEq lithium/kg) did not change SOD activity of any of the studied regions compared to saline control after 12 hr. However, in the other groups where second injection was given after 12 hr and animals were sacrificed after 24 hr of the first injection, activity of this enzyme increased in cerebral cortex (26%), corpus striatum (22%), hippocampus (21%), hypothalamus (22%) and mid-brain (19%) regions. Furthermore, the SOD activity in cerebellum and pons-medulla regions remained unchanged compared to control.

TABLE 5

EFFECT OF LiCl, RbCl AND CsCl ADMINISTRATION ON SUPEROXIDE DISMUTASE* OF RAT CEREBRAL CORTEX

Treatment†	Control¶	12 + 12 Hr‡	6 Days§
LiCl	1.42 ± 0.110	1.82 ± 0.114 (128#)	1.92 ± 0.121 (135**)
RbCl	1.37 ± 0.108	1.66 ± 0.126	1.55 ± 0.133
CsCl	1.49 ± 0.120	1.58 ± 0.131	1.74 ± 0.119

The values represent arithmatic mean ± S.E. of 6 rats in each group. The data in parentheses are percent compared to control as 100%.

*Activity represented as units/mg protein—one unit of SOD activity is defined as the amount of enzyme which inhibited the reaction by 50% [19].

†LiCl, RbCl and CsCl were injected IP in a volume of 0.5 ml.

‡Two mEq of an alkali metal/kg, one injection followed by another after 12 hr.

§Two mEq of an alkali metal/kg, daily one injection.

¶Control animals received an equivalent amount of NaCl.

Significantly different compared to control, $\#p<0.05$, $^{**}p<0.02$.

TABLE 6

IN VITRO EFFECTS OF LiCl, RbCl, CsCl AND KCl ON SUPEROXIDE DISMUTASE OF RAT CEREBRAL CORTEX

Alkali Metal* (2.0 mEq)	Superoxide Dismutase (Units/mg Protein)†	Percent of Control
None (control)	1.51 ± 0.121	100
LiCl	1.95 ± 0.133	129
RbCl	1.99 ± 0.129	132
CsCl	1.58 ± 0.127	105
KCl	1.54 ± 0.124	102

The values are arithmatic mean ± S.E. of 6 data obtained from 3 experiments each done in duplicate.

*Two mEq lithium, rubidium, cesium and potassium were added in incubation mixture.

†One unit of SOD activity is defined as the amount of enzyme which inhibited the reaction by 50% [19].

The data presented in Table 2 show the effects of Li_2CO_3 treatment on regional SOD activity at different time intervals for 12 days of exposure. The single injection of 2 mEq lithium/kg did not produce any effect on regional SOD activity after one day. However, daily treatment for 3 successive days enhanced SOD enzyme activity in cerebral cortex (21%), corpus striatum (24%), hippocampus (19%) and midbrain (20%) regions compared to their respective control. The administration of Li_2CO_3 for 6, 9 or 12 days was found to elevate the enzyme activity in all the studied brain regions. Furthermore, the magnitude of increase was more in cerebral cortex compared with other regions.

The data of Table 3 demonstrate the effects of *in vitro* addition of different concentrations of Li_2CO_3 on pyrogallol auto-oxidation and SOD enzyme activity in cerebral cortex region of normal rats. The addition of 0.1 to 10 mEq lithium did not produce any appreciable effect on the capacity of superoxide radical generation in an incubation mixture without the homogenate. However, the presence of a wide range of lithium concentration (0.1 to 8 mEq) increased cerebral SOD activity followed by a decrease (23%) at 10 mEq. The maximum activation of SOD was seen at 2 mEq concentration of lithium in the assay system.

When the *in vitro* effect of lithium (2 mEq) was tested on SOD activity in different brain regions, an increase was observed in all of the regions examined with maximum change in cerebral cortex (34%) followed by striatum (25%) and midbrain (22%) (Table 4). The distribution of this enzyme was found to be uniform in different regions of normal brain with comparatively little higher values in pons-medulla, hypothalamus and mid-brain regions.

Table 5 depicts the effects of LiCl, RbCl and CsCl (2 mEq alkali metal/kg) administration on SOD of cerebral cortex region after 24 hr (2 injections) and 6 days (once a day). LiCl produced an increase in the cortical SOD activity after 24 hr (28%) and 6 days (35%) of exposure, however, RbCl and CsCl did not show any significant change at both the time intervals compared to control. Furthermore, the administration of an equivalent amount of NaCl (control) did not show any change on this enzyme compared to untreated normal rats (data not given).

The *in vitro* effects of LiCl, RbCl, CsCl or KCl on SOD activity of cerebral cortex are given in Table 6. While the addition of 2 mEq of lithium and rubidium produced an increase in cortical SOD activity respectively by 29% and 32% cesium and potassium at the same concentration did not show any appreciable change.

DISCUSSION

Over the recent past, a number of hypotheses based on the balance of electrolytes, hormones and neurotransmitters have been proposed to explain the therapeutic and prophylactic action of lithium [10,25]. However, the initial neurochemical events involved behind these hypotheses are still unknown. In the present investigation, we studied the effects of lithium on brain SOD activity. Our results demonstrated that the acute administration of Li_2CO_3 (2 mEq/kg, two injections in 24 hr) intraperiteneally elevated the SOD level in most of the brain regions. However, there was no significant change in the enzyme activity after 12 hr of single injection. Furthermore daily administration of Li_2CO_3 produced an increase in the SOD activity of certain regions after 3 days and in all the regions after 6 days. The increased levels remained stationary even after 12 days, if the treatment continued. Under the *in vitro* condition, lithium enhanced SOD activity in the cerebral cortex region of normal rat brain at all concentrations tested, except 10 mEq when an inhibition was observed. Since lithium was not found to change the pyrogallol auto-oxidation, the observed results were due to the action of lithium on the enzyme itself rather than on the free radical production. These findings suggest that the activation of cerebral SOD after lithium treatment could be the result of direct action of alkali metal on this enzyme. Furthermore, the observed activation of SOD did not appear to be region specific as the *in vitro* addition of 2 mEq lithium increased enzyme activity in all brain regions,

of course parallel to *in vivo;* the maximum increase being in cerebral cortex.

It appears that lithium enhances the activity by directly interacting with SOD enzyme. The administration of lithium for 3 successive days has been reported to produce metal concentration in the range of 0.7–2.7 μEq/g after 1 mEq/kg dose, 1.5–5.7 μEq/g after 3 mEq/kg and 2.2–8.8 μEq/g after 6 mEq/kg, depending on the region [28]. These data are also supported by several other groups of researchers [11,20]. In the present investigation, we injected lithium 2 mEq/kg and after 3 successive injections it is supposed to produce a level of lithium around 1.4–5 μEq/kg in different brain regions. It is, therefore, reasonable to presume that lithium reaches the brain in a concentration (1.4–5 mEq) which on the basis of *in vitro* experiments, is sufficient to produce an increase in the SOD activity. Furthermore, there is a possibility that a single injection of lithium after 12 hr or 24 hr did not elevate regional lithium to a level that could activate SOD.

The mechanism of lithium-induced activation of cerebral SOD, at present, is not known. One can suggest that SOD is metallic enzyme containing Zn^{++} and Cu^{++} [4] and monovalent ions in general are known to activate intracellular enzymes containing divalent ions [30]. However, the administration of 2 mEq/kg rubidium or cesium twice in 24 hr or once daily for 6 days did not produce any significant change in the SOD activity. Furthermore, *in vitro* addition of 2 mEq rubidium, cesium or potassium showed that only rubidium increased enzyme activity. The use of LiCl salt in parallel to RbCl, CsCl and KCl produced effects almost similar, but a little more pronounced to those observed from Li_2CO_3. This could be due to the faster rate of distribution of LiCl compared to Li_2CO_3 in the tissue.

The data presented here thus showed that amongst alkali metals studied only lithium under *in vivo* as well as *in vitro* conditions increased the SOD activity, while rubidium enhanced the activity only under *in vitro* condition. There is a possibility that under present experimental conditions rubidium does not reach to the brain in a concentration significant to increase SOD level. If it is not the case and both lithium and rubidium, activators of SOD *in vitro,* are reaching to the brain with equal facility; then there is another possibility of involvement of certain factors, other than simple metal-SOD interaction, under *in vivo* condition in producing lithium-induced increase in enzymic activity.

The occurrence of SOD enzyme has been demonstrated in the central nervous system of several species including human beings [5,15]. Of particular interest are the studies of Thomas *et al.* [29] who showed the presence of SOD in synaptosomes of rat brain. The physiological function of synaptosomal SOD could be the prevention of free radical induced oxidation of catecholamines stored in certain nerve terminals. It is known that catecholamines get immediately oxidized in the presence of superoxide radicals and produce psychoactive compounds [22,24]. The formation of adrenochrome, adrenolutin and leuco-adrenochrome, suggested to be responsible for mental illness, could be the result of excessive levels of superoxide present in the vicinity of neurotransmitters in the brain. Pink epinephrine (adrenochrome), formed due to prolonged storage, when given to asthmatics or used as premedication for anesthesia produces psychotic reactions in patients [7]. Furthermore, Taborsky [27] studied the effects of adrenochrome and adrenolutin on the behavioural performance in trained rats and found both of them exerting a strong psychotropic action. It was suggested that the formation of such oxidized products under certain pathological conditions could result in mental illness. It has been hypothesized that the therapeutic actions of lithium are mediated through its effects on cellular membrane [8, 13, 14, 17], as this alkali metal has been found to affect the electrolyte balance, uptake and release of neurotransmitters, enzymes like Na^+, K^+-ATPase and adenylate cyclase, receptors, etc. The increased concentration of cytotoxic superoxides may also attack neuronal membrane and initiate a vicious cycle of free radical generation by the process of lipid peroxidation. Such peroxidation of membranal lipids can affect membrane associated enzymes and receptors leading to altered cation balance, neurotransmitter functions and efficiency of adenyl cyclase/cyclic AMP system. Brain Na^+, K^+-ATPase, uptake and release of neurotransmitters and dopamine receptors have already been reported to be sensitive to the lipid peroxidation [9, 12, 21, 23]. It is possible that the lithium-induced enhanced regional SOD activity may increase the removal of superoxide radicals and thus help in the stabilization of membrane. It is further interesting to mention here that a number of the antipsychotic drugs have been demonstrated to have antioxidant activity in inhibiting lipid peroxidation and to recover peroxidation-induced inhibited Na^+, K^+-ATPase in the brain tissue [23].

Because of genetic, chemical or biochemical insult, pathological levels of superoxide radical could result either due to its excessive generation or slower disposition by superoxide dismutase. There is sufficient evidence to believe that an excess of superoxide radicals in the central nervous system under certain abnormal conditions may produce a variety of nervous disorders. The demonstrated lithium-induced increase in SOD enzyme activity may facilitate the process of disposal of these cytotoxic superoxide radicals. However, further studies are needed before one can correlate the therapeutic efficacy of lithium in affective disorders with its activating effects on brain SOD.

ACKNOWLEDGEMENT

This work was supported by the Ontario Mental Health Foundation.

REFERENCES

1. Ahluwalia, P. and R. L. Singhal. Lithium and central monoamine neurotransmitter systems. *Drug Dev Res* **3:** 111–122, 1983.
2. Colburn, R. W., F. K. Goodwin, W. E. Bunney, Jr., and J. M. Davis. Effect of lithium on the uptake of noradrenaline by synaptosomes. *Nature* **215:** 1395–1397, 1967.
3. Fridovich, I. Superoxide dismutase. *Adv Enzymol* **41:** 35–47, 1974.
4. Fried, R. Superoxide dismutase activity in the nervous system. *J Neurosci Res* **4:** 435–441, 1979.
5. Fried, R. and P. Mandel. Superoxide dismutase of mammalian nervous system. *J Neurochem* **24:** 433–438, 1975.
6. Glowinski, J. and L. L. Iversen. Regional studies of catecholamines in the rat brain. The disposition of (^{3}H) norepinephrine, (^{3}H) dopamine and (^{3}H) DOPA in various regions of the brain. *J Neurochem* **13:** 655–669, 1966.
7. Grof, S., M. Vojtechovsky, V. Vitek and S. Prankova. Clinical and experimental study of central effects of adrenochrome. *J Neuropsychiatry* **5:** 33–50, 1963.

8. Guerri, C. Effect of lithium on synaptosomal brain enzymes. *Biochem Pharmacol* **31:** 449–453, 1982.
9. Heikkila, R. E., F. S. Cabbat and L. Manzino. Inhibitory effects of ascorbic acid on the binding of (^{3}H) dopamine antagonists to neostriatal membrane preparations. Relationship to lipid peroxidation. *J Neurochem* **38:** 1000–1006, 1982.
10. Hendler, N. H. Lithium pharmacology and physiology. In: *Handbook of Psychopharmacology,* vol 14, edited by L. L. Iversen, S. D. Iversen and S. H. Snyder. New York: Plenum Press, 1978, pp. 233–273.
11. Heurteaux, C., J. C. Wissocq, T. Stelz and M. Thellier. Microlocalisation quantitative du lithium dans le cerveau de la souris. *Biol Cell* **35:** 251–258, 1979.
12. Hexum, T. D. and R. Fried. Effects of superoxide radicals on transport (Na^+, K^+) adenosine triphosphatase and protection by superoxide dismutase. *Neurochem Res* **4:** 73–82, 1979.
13. Katz, R. I., T. N. Chase and I. J. Kopin. Evoked release of norepinephrine and serotonin from brain slices: inhibition by lithium. *Science* **162:** 466–467, 1968.
14. Knapp, S. and A. J. Mandell. Short- and long-term lithium administration: effects on the brain's serotonergic biosynthetic systems. *Science* **180:** 645–647, 1973.
15. Loomis, T. C., G. Yee and W. L. Stahl. Regional distribution of superoxide dismutase in brain. *Experientia* **32:** 1374–1376, 1976.
16. Lowry, O. H., N. J. Rosenbrough, A. L. Farr and R. J. Randall. Protein measurement with Folin phenol reagent. *J Biol Chem* **193:** 265–275, 1951.
17. Maggi, A. and S. J. Enna. Regional alterations in rat brain neurotransmitter system following chronic lithium treatment. *J Neurochem* **34:** 888–892, 1980.
18. Marklund, S. and G. Marklund. Involvement of superoxide anion radical in the autooxidation of pyrogallol and a convenient assay for superoxide dismutase. *Eur J Biochem* **47:** 469–474, 1974.
19. Minami, M. and H. Yoshikawa. Simplified assay method of superoxide dismutase activity for clinical use. *Clin Chim Acta* **92:** 337–342, 1979.
20. Mukherjee, B. P., P. T. Bailey and S. N. Pradham. Temporal and regional differences in brain concentrations of lithium in rats. *Psychopharmacology (Berlin)* **48:** 119–121, 1976.
21. Pastuszko, A., W. Gordon-Majszake and Z. Dabrowiecki. Dopamine uptake in striatal synaptosomes exposed to peroxidation "in vitro." *Biochem Pharmacol* **32:** 141–146, 1983.
22. Powis, G. Hepatic microsomal metabolism of epinephrine and norepinephrine by superoxide-dependent and independent pathways. *Biochem Pharmacol* **28:** 83–89, 1979.
23. Schaefer, A., M. Komlos and A. Seragi. Lipid peroxidation as the cause of the ascorbic acid induced decrease of adenosine triphosphatase activities of rat brain microsomes and its inhibition by biogenic amines and psychotropic drugs. *Biochem Pharmacol* **24:** 1781–1786, 1975.
24. Schenkman, J. B., I. Jansson, G. Powis and H. Kappus. Active oxygen in liver microsomes: mechanism of epinephrine oxidation. *Mol Pharmacol* **15:** 428–438, 1978.
25. Schou, M. Pharmacology and toxicology of lithium. In: *Annual Review of Pharmacology and Toxicology,* Vol 16, edited by H. W. Elliot, R. George and R. Okun. Palo Alto, CA: Annual Review Inc., 1976, pp. 231–243.
26. Schou, M. The range of clinical uses of lithium. In: *Lithium in Medical Practice,* edited by F. N. Johnson and S. Johnson. Baltimore: University Park Press, 1978, pp. 21–39.
27. Taborsky, R. G. Indoxyl derivatives: potential psychotropic metabolites. *Int J Neuropharmacol* **7:** 483–486, 1968.
28. Thellier, M., C. Heurteaux and J. C. Wissocq. Quantitative study of the distribution of lithium in the mouse brain for various doses of lithium given to the animal. *Brain Res* **199:** 175–196, 1980.
29. Thomas, T. N., D. G. Priest and J. W. Zemp. Distribution of superoxide dismutase in rat brain. *J Neurochem* **27:** 309–310, 1976.
30. Williams, R. J. P. The chemistry and biochemistry of lithium. In: *Lithium, Its Role in Psychiatric Research and Treatment,* edited by S. Gershon and B. Shopsin. New York: Plenum Press, 1973, pp. 15–31.