Effect of Lithium Carbonate Administration Singly or in Combination with Some Psychotropic Drugs on the Radiiodide Uptake by Mouse Thyroid

Masumitsu TAKASUGI, Kazuhiko TERAOKA, Kazuo MINAKUCHI, Seiji AKAMATSU, Mikio NISHIDA, Naoyuki KAMATA, Toyoyasu KUWAЕ, Munetsugu KURATA, and Jun KAWADA

Department of Pharmacy, University Hospital, School of Medicine* and Faculty of Pharmaceutical Sciences, The University of Tokushima, Tokushima City, 770, Japan and Faculty of Pharmaceutical Sciences, Josai University, Sakado City, 350-02, Japan

(Received February 6, 1989)

The present study dealt with two objects; the first object was to examine whether or not lithium uptake in the thyroid is modified by the thyroid state and how the intrathyroidal Li affects iodide uptake by the thyroid. Male and female mice were given lithium carbonate (Li) with propylthiouracil (PTU) or thyroxine (T₄). Li was measured by a flameless atomic absorption spectrometry. The total Li content in thyroid was unaffected with PTU or T₄, however, Li concentration per unit mass was reduced by PTU but unaffected by T₄. The thyroid: serum ratio (T/S) of 125I resulted that the T/S became higher when Li concentration per unit mass was lower and vice versa, suggesting that Li uptake is controlled by thyroid states and Li in the gland interferes with the iodide uptake. Serum triiodothyronine (T₃) and T₄ by radioimmunoassay showed that PTU alone and in combination with Li lowered serum T₄, while a high level of T₄ by its supplement was suppressed by co-administration of Li. T₃ level was lowered by Li alone, but not severely affected by other drugs. The results suggest that Li enhances T₄ clearance without T₄-T₃ conversion.

The second object was to examine the effect of combined psychotropic drugs on thyroid function. Carbamazepine (CBZ), haloperidol (HLP) and imipramine (IPA) were given singly or in combination with Li to examine their effects on the T/S of 125I. Only CBZ reduced the T/S but CBZ plus Li had no summative effect. Neither HLP nor IPA affected the T/S, singly or in combination with Li, suggesting that HLP or IPA does not interfere with an iodide pumping machinery. No distinct sex difference was observed in drug effects.

Keywords — psychotropics; neuroleptics; lithium; carbamazepine; haloperidol; imipramine; thyroid; iodide metabolism

Introduction

Li salts have been used for the treatment of mania and prophylaxis against recurrent manic-depressive disorders.1,2) In the use of Li, however, various side effects appeared.3) It is said that 5—10% of patients on a long-term Li treatment develop hypothyroidism and goiter.4) Evidences indicated the reductions in iodide uptake by the thyroid,5,6) the hormone secretion7) and protein bound iodine7,8) due to Li treatment. In contrast, other reports showed a trend of increase or little changes in the iodide uptake9) and iodide metabolism10) with Li treatment. Currently it is more difficult to evaluate the Li effect singly because psychiatric abnormality is often associated with thyroid diseases and antithyroid drugs are combined with various psychotropic drugs. Recently we observed in an animal experiment11) that the total content of Li in the thyroid was merely changed, although a glandular weight and Li concentration per unit mass were affected by various therapeutic drugs. This evidence suggests that intrathyroidal level of Li depends on the mass of the thyroid and not directly on the pharmacological effect of drugs.

The present study dealt with two objects to be examined; the first was to determine how propylthiouracil (PTU)-induced hyperthyrotic and thyroxine (T₄)-induced hypothyrotic conditions modify the effect of Li on the iodide uptake which is the initial step for thyroid hormone synthesis. For this end, Li uptake by the mouse thyroid was examined under the influence of PTU or T₄ which modifies thyroid states. The second was to investigate the com-
bined effect of Li and other psychotropic drugs on the iodide uptake by the thyroid, because psychiatric abnormality is often associated with thyroid diseases. The study was conducted to demonstrate whether each of carbamazepine (CBZ), haloperidol (HLP) and imipramine (IPA) singly or in combination with Li affects the iodide uptake in the thyroid. It has been recognized that the population of female patients with thyrotic and psychotic diseases is much greater than that of male patients. We also studied the effects of drugs in both male and female mice.

Materials and Methods

Chemicals — CBZ and IPA were purchased from Wako Chemical Co. (Osaka, Japan). HLP (Serenase Injection) was a product of Dainippon Pharm. Co., Ltd. (Osaka, Japan). Polyoxyethylene-hydrogenated castor oil (HCO-60) was purchased from Nikko Chemical Co., Ltd. (Tokyo, Japan). Radioactive iodide (\(^{125}\text{I}\)) in the form of NaI was from New England Nuclear (Boston, U.S.A.). All other chemicals used were in the special reagent grade and obtained from local commercial sources.

Animal Treatment — ddY-Mice of both sexes, weighing 20—25 g, were fed a regular pellet diet (Type MF, Oriental Yeast Co., Ltd., Japan) and tap water ad libitum. Li was added to a drinking water at a concentration of either 0.1 or 0.1% as Li\(_2\)CO\(_3\) for 4 weeks. Mice thus pretreated with 0.01% Li were divided into three groups. To induce an experimentally hyperthyrotic or hypothyrotic states, mice in the first group received 0.5 mg/ml of PTU in a drinking water for 10 d before sacrifice (Li plus PTU treated group). Mice in the second group were intraperitoneally injected with 0.5 \(\mu\)g/d of T\(_4\) for last 10 days before sacrifice (Li plus T\(_4\) treated group). Mice in the third group were Li-treated. Another set of three groups of mice were prepared without Li; one group served for only PTU-administration, the second group for T\(_4\)-administration and the third was as an intact control.

For psychotropic drugs, a dosage and the way of administration of each drug were determined by a preliminary test. A 125 \(\mu\)g/g body weight (bw) of CBZ in a 10% HCO-60 suspension was administered by intubation. A 11.7 \(\mu\)g/g bw of IPA and 4.3 \(\mu\)g/bw of HLP were intraperitoneally injected, respectively. At 2 h before sacrifice, mice pretreated with 0.01% Li for 4 weeks received the above-mentioned dose of CBZ, IPA and HLP, respectively. To the control group, either 10% HCO-60 as a vehicle of CBZ or 0.9% saline solution in case of HLP and IPA was given.

Measurement of Li Concentration in Thyroid and Serum — Thyroids and sera were obtained at autopsy. The thyroid was rapidly weighed on a semi-micro balance and homogenized in 500 \(\mu\)l of re-distilled water with a motor-driven glass homogenizer. The homogenate was centrifuged at 3000 rpm for 10 min at room temperature. The supernatant fluid was diluted with re-distilled water and used for the determination of Li concentration by a flameless atomic absorption spectrometry (Type AA-8220 Nippon Jarrel-Ash Co., Ltd., Kyoto) using a solution of Li\(_2\)CO\(_3\) as standard. The assay condition was in the following; the wave length at 670.8 nm; drying at 20 A for 15 s; ashing at 70 A for 20 s; atomizing at 230 A for 10 s.

Determination of Thyroid: Serum Concentration Ratio of \(^{125}\text{I}\) — The thyroid: serum concentration ratio (T/S) of \(^{125}\text{I}\) was measured by the method of Halmi et al. To examine the effect of a psychotropic drug on the T/S, each drug replaced PTU at 2 h before sacrifice. A protocol of the determination was indicated below.

```
Li ➔ drug ➔ ▶ 1 h ➔ ▶ 125I ➔ ▶ autopsy
```

The radioactivity was measured by an Aloka auto well gamma scintillation counter (Type ARC-360). T/S was defined as a ratio of the radioactivity in 100 mg thyroid tissue (T) to the radioactivity in 100 \(\mu\)l of the serum (S).

Measurement of Triiodothyronine (T\(_3\)) and T\(_4\) Concentrations in Serum — Serum T\(_3\) and T\(_4\) levels were determined by radioimmunoassay (RIA). Double antibody RIA systems for T\(_3\) and T\(_4\) were developed in our laboratory. The
Fig. 1. Lithium Effect on Growth Curves of Male (a) and Female (b) Mice

Each point represents the mean ± S.D. of 5 mice. Arrow indicates the initiation of Li administration.

○, control; ●, 0.01% Li; △, 0.1% Li.

RIA systems have the following features; the T₃ RIA system shows a reasonably straight line in a range of 1.0 - 30.0 ng/ml, showing less cross-reactivity than a few % with other thyroid hormones (T₄, reverse T₃). The T₄ RIA system is in a range of 6.0 - 150.0 ng/ml, showing no cross-reactivity with T₃ or reverse T₃.

Statistical Analysis — Statistical analysis of the result was carried out by Student's t-test.

Fig. 2. Single and Combined Effects of Li with PTU or T₄ on Thyroid Weight

Open column and shaded column represent the thyroid weight for male and female mice, respectively. Each value represents the mean ± S.D. for 6 mice. a, b) mean p < 0.05 and p < 0.001 against the control. Inserted p value indicates a comparison between corresponding columns.

Results

Figures 1a and 1b show the body weight change in both male and female mice by Li administration. When 0.01% Li₂CO₃ was administered, no clear changes in the body weight were observed. When 0.1% Li₂CO₃ was given, a transient decrease in the body weight was observed at the initial period of Li administration. The decrease in body weight was seemingly due to a toxic effect of Li and a refusal of drinking the solution. Therefore, the Li concentration was fixed as 0.01% in subsequent studies.

Figure 2 illustrates that the administration of 0.01% Li in drinking water for 4 weeks did not change the thyroid weight of both sexes. PTU significantly increased the thyroid weight but Li plus PTU increased the thyroid weight more than PTU alone. T₄ administration resulted in a slight reduction in the thyroid weight compared with the weight in the control. This reduction in thyroid weight was statistically significant for female mice (p < 0.05) but insignificant in male mice. Li plus T₄ resulted in normal thyroid weight in both sexes.

The Li effect on iodide uptake by the thyroid was examined by measurement of the T/S of
Fig. 3. Single and Combined Effects of Li with PTU or T4 on the T/S of $^{125}$I

Figures (a) and (b) show the results with male and female mice, respectively. Open column and shaded column represent the T/S of $^{125}$I and Li concentration in thyroid, respectively. Each column presents the mean ± S.D. of at least 5 mice. a, b, c) mean $p < 0.05$, $p < 0.01$ and $p < 0.001$ against the control. Inserted $p$ value is for corresponding columns.

$^{125}$I. Figure 3a for male and 3b for female mice depict the relationship between the effect of Li on T/S and the glandular Li concentration in the presence of PTU or T4. The T/S was reduced when Li was accumulated in the thyroid but the T/S was raised by PTU alone and reduced to a normal range by Li plus PTU. The T/S was severely suppressed by T4. However, the suppression became less by Li plus T4. This effect of Li plus T4 on T/S was much more pronounced in female mice than in male mice, even statistical analyses gave $p < 0.01$ for both sexes. Total Li contents in thyroid from Li plus PTU and Li plus T4 groups were not changed from that of Li alone treated group; in male mice, 8.1 ± 2.6 for Li alone, 9.3 ± 0.1 for Li plus PTU and 9.0 ± 0.8 ng/iodin for Li plus T4 treated group; in female mice, 5.6 ± 0.9 for Li alone, 8.0 ± 2.7 for Li plus PTU and 8.0 ± 1.1 ng/iodin for Li plus T4 group ($n = 5$ for each group). Concentrations of serum Li in PTU or T4 co-administered group unaltered from that in the Li alone (data not presented).

As shown in Fig. 4a, serum T4 level was not affected by 0.01% Li in drinking water for 4
weeks. PTU significantly reduced $T_4$ level but the combination of Li plus PTU did not cause further reduction nor elevation than PTU alone. The exogenous $T_4$ administration reflected to the high level of serum $T_4$ but Li plus $T_4$ reduced the hormone level to the normal range. In contrast to $T_4$, $T_3$ levels in both sexes were reduced by Li (Fig. 4b). Furthermore, $T_3$ levels were not elevated by the addition of $T_4$, although the level in female mice became slightly higher. This heightened $T_3$ level in female was normalized by the addition of Li. PTU alone or in combination with Li affects $T_3$ levels less significantly than $T_4$ levels.

The effect of CBZ and other psychotropic drugs on T/S was examined, using mice pre-treated with 0.01% Li for 4 weeks. Figure 5 shows the administration of CBZ significantly reduced the T/S but the combination of Li and CBZ did not show clear additive effect on T/S. Under the conditions applied, neither HLP nor IPA affected the T/S singly or in combination with Li.

Discussion

Although Li is known to be goitrogenic, the dosage of 0.01% Li for 4 weeks did not induce goiter in the mouse thyroid. This dosage is approximately equivalent to 15 mg/kg/d and five to ten times higher than a therapeutic dosage in human. The concentration of Li was increased to 0.1%. However, some toxic effects such as transient delay of the growth or a refusal of drinking Li-containing water was observed. Thus, the induction of Li-dependent goiter in mice is rather difficult but the addition of PTU to Li resulted in an increase in thyroid weight over that of PTU alone (Fig. 2). Such a synergistic potency of antithyroid drugs in Li-dependent goiter formation has been reported in the rat. The present observation strongly indicates a latent potency of goitrogenic property of Li in the mouse which may appear with other antithyroid drugs.

Serum Li concentration was unaltered by the co-administration of PTU or $T_4$, but Li concentration in the thyroid (neq/mg) was decreased in the presence of PTU, while the total content of Li in the thyroid treated with PTU was not different from that of the Li treated group, indicating that Li concentration per unit of weight was reduced due to PTU-induced goiter. On the other hand, $T_4$ alone or the combination of Li and $T_4$ gave no significant changes in thyroid weight. In contrast to the case of PTU administration, neither Li concentration in neq/mg nor the total Li content in the thyroid were affected by the treatment with $T_4$. This must be owing to unaltered mass of the thyroid under the conditions applied. From this information, the intrathyroidal Li concentration is likely to be controlled by the thyroid state which is modified by drugs. The importance of evaluation of thyroid state for Li therapy was emphasized by Lazarus and Bennie. The evidence in the present study may provide an experimental support for the finding by Lazarus and Bennie.

Results in Figs. 3a and 3b depict that Li interferes with the iodide uptake by the mouse thyroid but the real mechanism of Li action has not been clearly elucidated. Evidences appeared in reference are not always accordant regarding Li effects on the intrathyroidal iodide metabolism; some reports showed that Li interfered with hormone synthesis and its secretion, whereas others showed that the iodide metabolism was hardly affected by Li. Although the data was
not presented, our preliminary study with 0.01\% Li administration for 4 weeks did not cause any striking alterations in radioiodide metabolism in the thyroid.

Since thyroid stimulating hormone (TSH) in serum is elevated by PTU and is suppressed by high levels of serum T\(_4\), the effect of PTU or T\(_4\) in combination with Li on iodide uptake can be explained by the change of serum TSH level. We tried to measure serum TSH by using rat anti-TSH serum with a double antibody RIA but the cross reactivity between the rat anti-TSH serum and mouse serum TSH was not sufficient. Without serum TSH information, serum hormone levels may reflect to an overall physiological condition of the thyroid. When the T\(_4\) level is lowered by PTU, Li does not overcome the PTU effect, but when the T\(_4\) level is extremely heightened, Li may increase a clearance of excess T\(_4\). Since the peripheral thyroxine 5'-monodeiodinase activity is unaffected by Li, the T\(_4\) to T\(_3\) conversion may not be a major metabolic pathway responsible for reducing the serum T\(_4\) by Li. Serum T\(_3\) level is also decreased by Li but not by PTU. It is likely that these drugs affect T\(_4\) and T\(_3\) metabolism differently.

For clinical purposes, a combined regimen of CBZ and Li was used in the hope of alleviating polyurea and polydipsia due to Li. A combined regimen of HLP and Li has been advocated for the acute manic state, and the combination of Li plus IPA has been tested for recurrence of depression. In the present study, the experiment was carried out to examine the effect on the iodide uptake by the mouse thyroid of CBZ, HLP or IPA singly or in combination with Li. CBZ reduced the iodide uptake but the effect was not potentiated by Li. The T/S was not affected with HLP nor Li plus HLP. IPA with or without Li did not affect the T/S, either. CBZ and IPA structurally resemble but their effects on the T/S are different. Thus, further study is necessary to elucidate the structure-activity relationship for tricyclic compounds. Furthermore, whether or not sites of action for Li and CBZ are the same should be clarified. It should be stated, however, that the inability of HLP or IPA to inhibit the T/S does not exclude a possible interaction of HLP or IPA with thyroid hormone at peripheral sites because T\(_3\) enhances the anti-depressant effect of IPA.

Thyroid disease with psychoses is reportedly more common in women than in men. We have been interested in effects of psychotropic drugs on thyroid function in different sexes. No clear difference between male and female mice was found, although several minor differences in drug effects were observed as stated before. We speculate that there is no distinctive sex difference in effects of these psychotropic drugs on mouse thyroid.

**Acknowledgement** The research was supported in part by a Grant-in-Aid for Scientific Research from the Ministry of Welfare of Japan.

**References**

iodide uptake in mouse thyroid treated with propyl thiouracil or thyroxine, *Naturwissenschaften*, submitted.


