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Effects of triiodothyronine and imipramine on basal 5-HT levels and 5-HT$_1$ autoreceptor activity in rat cortex

Eitan Gur, Tsuri Lifschytz, Bernard Lerer, Michael E. Newman*

Biological Psychiatry Laboratory, Department of Psychiatry, Hadassah-Hebrew University Medical Center, P.O.B. 12000, Jerusalem 91120, Israel

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Abstract

Clinical studies have shown that triiodothyronine (T3) both augments and accelerates the therapeutic response to antidepressant drugs, particularly tricyclics. There is evidence that this effect is mediated by the serotonergic system. We show here that T3 administered daily for 7 days over the range 0.02–0.5 mg/kg increases basal serotonin (5-hydroxytryptamine, 5-HT) levels, as measured by in vivo microdialysis in rat cortex, in a dose-dependent fashion. All the doses of T3 examined reduced 5-HT$_{1A}$ autoreceptor activity, as measured by the effect of 8-hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT, 0.05 mg/kg s.c.) to decrease 5-HT levels in frontal cortex. T3 administered daily for 14 days at 0.02 mg/kg also reduced 5-HT$_{1B}$ autoreceptor activity, as measured by the effect of locally administered 3-(1,2,5,6-tetrahydropyrid-4-yl)pyrrolo[3,2-b]pyrid-5-one (CP 93129, 10 μM) to decrease 5-HT levels. In animals administered imipramine (10 mg/kg/day by osmotic minipump) concurrently with T3 injections, no further changes in either 5-HT$_{1A}$ or 5-HT$_{1B}$ autoreceptor activity were seen. We suggest that the effect of T3 to accelerate the therapeutic actions of antidepressant drugs may be due to a combination of the actions of T3 at autoreceptors and the actions of the drugs at postsynaptic 5-HT$_{1A}$ receptors.

Keywords: 5-HT (5-hydroxytryptamine, serotonin); Triiodothyronine; Antidepressant; 5-HT$_{1A}$ receptor; 5-HT$_{1B}$ receptor

1. Introduction

The delay after onset of administration until a therapeutic effect is observed remains one of the major problems associated with the use of antidepressant drugs in the treatment of depression. A further problem is that many patients do not respond to any of the drugs in clinical use. Several strategies have been proposed to overcome these problems, notably the use of potentiating or accelerating agents, which themselves may not have therapeutic effects, to potentiate or accelerate the effects of established antidepressants. One such agent is pindolol, which has been shown to act by blocking the somatodendritic 5-HT$_{1A}$ autoreceptors on serotonergic neurons which normally decrease serotonin (5-hydroxytryptamine, 5-HT) synthesis and cell firing, thus resulting in an overall increase in serotonergic transmission (Artigas et al., 1996). More recent work (Artigas et al., 2000) has suggested that the mechanism of action of pindolol may involve inhibition of the activity of 5-HT$_{1B}$ autoreceptors, which block 5-HT release at the nerve terminals. Another agent is lithium, which has also been shown to produce its therapeutic effect in treatment-resistant depression by enhancing 5-HT neurotransmission, probably because of its 5-HT releasing properties (Haddjeri et al., 2000).

The thyroid hormone triiodothyronine (T3) was shown in a meta-analysis aggregating eight studies (Aronson et al., 1996) to be highly effective as an augmenting agent in potentiating the effects of drug treatments in treatment-resistant depression, inducing an overall 23.2% improvement in response rates with moderate to large improvements in depression scores. In a more recent meta-analysis aggregating six studies, Altshuler et al. (2001) showed T3 to be highly effective in accelerating clinical response to tricyclic antidepressants in patients with nonrefractory depression. There are several lines of evidence supporting the idea that T3 may also exert its therapeutic effects by an action on serotonergic transmission in the brain (reviewed

* Corresponding author. Tel.: +972-2-6777617; fax: +972-2-6439294.
E-mail address: newman@hadassah.org.il (M.E. Newman).

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by Newman et al., 2000; Bauer and Whybrow, 2001; Bauer et al., 2002) In particular, Cleare et al. (1995) showed that the cortisol and prolactin responses to the 5-HT releasing agent fenfluramine were significantly reduced in drug-free hypothyroid patients. The same investigators (Cleare et al., 1996) also showed that replacement thyroxine (T4) therapy to seven hypothyroid patients increased the cortisol response to fenfluramine without altering the prolactin response. The reduced responses to fenfluramine resemble the responses seen in depressed patients, while the increased response after T4 administration resembles the increased responses seen after chronic antidepressant treatment of such patients (for review, see Newman et al., 1998).

In a study from this laboratory (Gur et al., 1999a), we showed that daily s.c. administration of T3 at 0.1 mg/kg to rats for 7 days resulted in an increase in 5-HT levels in frontal cortex as measured by in vivo microdialysis. Administration of the tricyclic drug clomipramine at 10 mg/kg daily for 4 weeks also resulted in elevated 5-HT levels in frontal cortex, while in rats which received T3 simultaneously with the last week of clomipramine treatment, the increase was significantly greater than in rats which received one treatment only. This experimental design was intended to model the clinical use of T3 as an augmenting drug in treatment-resistant depression. Administration to the rats of the 5-HT_{1A} receptor agonist 8-hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT) showed that the increase in 5-HT levels was due to subsensitivity of the 5-HT_{1A} autoreceptors controlling 5-HT release in frontal cortex. Behavioural and some receptor binding data from other laboratories also support the hypothesis that T3 may exert its antidepressant-potentiating and-accelerating effects by increasing serotonergic neurotransmission as a result of inducing subsensitivity of 5-HT_{1A} and possibly also 5-HT_{1B} autoreceptors (reviewed in Newman et al., 2000; Bauer and Whybrow, 2001; Bauer et al., 2002). The present work had two objectives: (a) to determine the dose dependence of the effects of T3 on basal 5-HT levels and 5-HT_{1A} and 5-HT_{1B} autoreceptor activity in rat frontal cortex and (b) to model the accelerating effect of T3 in the clinic using experimental animals. For this purpose, the effects of daily administration of T3 for 2 weeks either alone or together with the tricyclic antidepressant imipramine were compared with those of administration of imipramine for 4 weeks. Imipramine was chosen for this experiment as the majority of the clinical studies comprising the meta-analyses of Aronson et al. (1996) and Altshuler et al. (2001) used this drug.

2. Materials and methods

2.1. Treatment of animals

Male Albino rats (Sabra strain, derived from Sprague–Dawley strain) were used in all experiments. The rats were housed by treatment group in a temperature-controlled environment (24 °C) with a regular 12-h light/dark cycle. Food and water were freely available. For the dose dependence experiment, animals received s.c. injections of 0.9% saline or 0.9% saline containing T3 at concentrations of 0.02, 0.05, 0.2, or 0.5 mg/kg daily for 7 days. The stock solution of T3 was prepared in 1 M NaOH and all solutions for injection were adjusted so that the final concentration of NaOH was 0.01 M NaOH and the injection volume 1 ml/kg. Implantation of guides and probes was performed on the last day of injection, at least 1 h after administration of T3. For the ‘‘acceleration’’ experiment, four groups of rats were studied. Group (a) received 0.9% saline by Alzet minipumps (2ML4) over 4 weeks, group (b) received saline by minipump (2ML2) over 2 weeks simultaneously with T3 by s.c. injection at 0.02 mg/kg daily for 2 weeks, group (c) received imipramine at 10 mg/kg/day by minipump (2ML4) for 4 weeks, and group (d) received T3 by s.c. injection at 0.02 mg/kg daily for 2 weeks simultaneously with 10 mg/kg/day imipramine by minipump (2ML2) for 2 weeks. The minipumps were implanted subcutaneously under anesthesia with a 17:3 mixture of ketamine (100 mg/ml) and xylazine (2%). Implantation of minipumps was performed in a staggered manner so that a 3-day experimental period (1 day for implantation of guides and probes and 2 days for collection of fractions) was available for each pair of animals. Implantation of guides and probes was performed on the 14th or 28th day after insertion for the 2ML2 and 2ML4 minipumps, respectively, so that on the days when fractions were collected, the results would not be complicated by the presence of freshly delivered drug.

2.2. Implantation and perfusion of microdialysis probes

Animals were anaesthetised with a 17:3 mixture of ketamine (100 mg/ml) and xylazine (2%) and mounted in a stereotaxic apparatus. Guides for dialysis probes (CMA/12) were implanted into frontal cortex at anterior 3.2 mm from bregma, 2.5 mm lateral, and 6.0 mm vertical. Rats were maintained under anesthesia for approximately 1 h, after which they were free-moving and had unlimited access to food and water. Dialysis probes (4 mm) were inserted into the guides towards the end of the period of anesthesia. The inlets of the probe were connected, through plastic tubing with an internal volume of 12 μl/m, to 2.5-ml gas-tight syringes mounted on a microinfusion pump. The inlet and outlet tubing of the probe were mounted to a flexible cable running from the head of the rat to a liquid swivel, allowing the animal to rotate and rear without entangling the fluid tubing. The probes were perfused with Ringer’s solution containing 2.25 mM $CaCl_2$, 4 mM KCl, 147 mM NaCl, and 10 μM citalopram, pH 6.5, at 0.2 μl/min overnight. The following morning, the flow rate was increased to 0.5 μl/min, and 30-min fractions collected. After each experiment, the dialysis probes were removed

under anesthesia, sterilised in alcohol, and if still intact, re-inserted into new animals. The animal procedures outlined above received the approval of the Institutional Animal Care and Use Committee of the Hebrew University Faculty of Medicine and Dental Medicine and Hadassah Medical Organization.

2.3. 5-HT receptor challenges

On the second experimental day for each animal, fractions were injected into the high performance liquid chromatography (HPLC) apparatus immediately after collection for measurement of 5-HT. Once stable baseline 5-HT levels had been obtained, usually after collecting four or five experimental samples, the 5-HT_{1A} receptor agonist 8-OH-DPAT (0.05 mg/kg) was injected s.c. A further six fractions were then collected. On the following day, once stable baseline 5-HT levels had been obtained, the 5-$HT_{1B/1D}$ receptor agonist CP 93129 [3-(1,2,5,6-tetrahydropyrid-4-yl)pyrrolo[3,2-b]pyrid-5-one] at a concentration of 10 μM was infused into the cortex via the microdialysis probe during two fractions, i.e. for 60 min, and a further four or five fractions collected.

2.4. Determination of 5-HT levels

Concentrations of 5-HT were determined by a Bioanalytical systems (BAS) High Performance Liquid Chromatography (HPLC) system. Samples were injected immediately after collection using a Rheodyne 9125 injector with a 5-μl injection loop. The mobile phase was made up of 90 mM sodium dihydrogen phosphate, 10 mM NaCl, 0.5 mM EDTA, 0.15 g/l sodium octyl sulfate, and 10.5% acetonitrile, pH 5, and was delivered by the HPLC pump at 1.0 ml/min. The mobile phase was passed through a flow splitter and pumped through a 10-cm C-18 5-mm reversed phase column at 0.1 ml/min. 5-HT content was analysed with an LC-4C electrochemical detector (BAS) with a glassy carbon working electrode set at 550 mV vs. an Ag/AgCl reference electrode. Concentrations of 5-HT were calculated by comparing peak levels from the microdialysis samples with those of external standards of known concentration of 5-HT. The detection limit was 0.5–1 fmol. The average of the first four or five baseline samples was taken as 100%.

2.5. Materials

T3, 8-OH-DPAT, 5-HT creatinine sulfate complex, and sodium octyl sulfate were obtained from Sigma (St. Louis, MO, USA). CP 93129 was a gift of Pfizer, Groton, CT, USA. Citalopram was a gift of H. Lundbeck, Copenhagen, Denmark. HPLC grade acetonitrile was from Frutarom, Haifa, Israel. All other chemicals were of analytical grade and were obtained from Merck-Darmstadt, Germany.

2.6. Data analysis

5-HT levels expressed as percentages of the initial levels for each animal were analysed over the time course for each challenge by two-way analysis of variance (ANOVA), with treatment as a "between groups" variable and time (fraction number) as a "within groups" variable, i.e. as a repeated measure. This was followed in the case of the dose dependence experiment by post hoc Newman–Keuls test, and in the case of the "acceleration" experiment by the use of planned comparisons. The appropriate contrasts were used in order to generate a main (overall) effect for T3, a main (overall) effect for imipramine, and an interaction effect. In addition, individual planned comparisons were performed between the various groups.

3. Results

3.1. T3 dose-response experiments

T3 induced a dose-dependent increase in basal 5-HT levels in frontal cortex (Fig. 1). One-way analysis of variance showed a significant effect of dose ($F[4,50]=2.75$, $P=0.038$). Post hoc Newman–Keuls tests showed a significant difference between 5-HT levels in saline-treated rats and in rats treated with 0.2 mg/kg T3 ($P=0.022$).

Administration of 8-OH-DPAT (0.05 mg/kg) induced a decrease in 5-HT levels (Fig. 2). The degree of inhibition was reduced in rats administered T3. There was a significant effect of time after administration of 8-OH-DPAT ($F[5,75]=7.45$, $P=0.00001$) and a significant effect of T3 dose ($F[4,15]=4.87$, $P=0.01$). Post hoc Newman–Keuls test showed significant differences between saline-treated rats

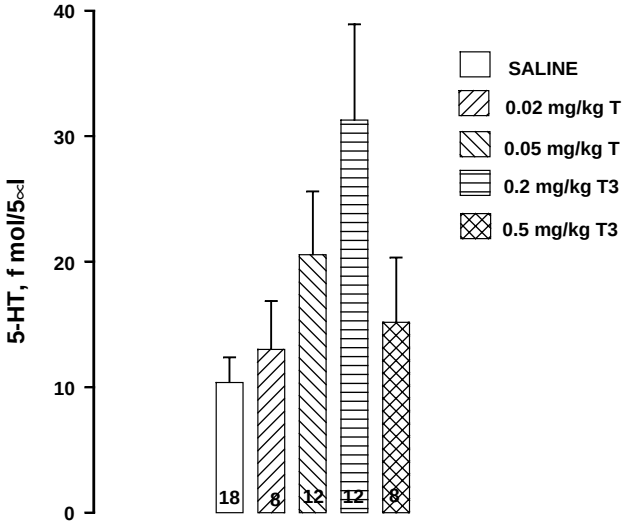

Fig. 1. Dose dependence of the effect of T3 administered daily for 7 days on basal 5-HT levels in frontal cortex. Values are given as mean ± S.E.M. The number of observations at each dose is given at the foot of the columns.

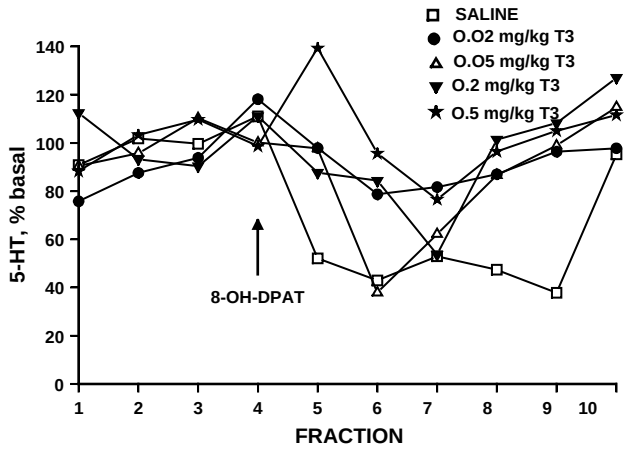

Fig. 2. Effect of 8-OH-DPAT injection (50 μg/kg s.c.) on 5-HT levels in frontal cortex. Results are means of observations from four animals treated with saline, five animals treated with 0.02 mg/kg T3, five animals treated with 0.05 mg/kg T3, three animals treated with 0.2 mg/kg, and three animals treated with 0.5 mg/kg T3. S.E.s are omitted for clarity.

and rats administered with each of the T3 doses (saline vs. 0.02 mg/kg, $P=0.014$; saline vs. 0.05 mg/kg, $P=0.02$; saline vs. 0.2 mg/kg, $P=0.032$; saline vs. 0.5 mg/kg, $P=0.0087$).

Administration of the 5-HT_{1B} receptor agonist CP 93129 (10 μM) via the dialysis probe for 60 min, i.e. during two fractions, reduced 5-HT levels in frontal cortex (Fig. 3). There was a significant effect of time after administration of CP 93129 ($F[5,95]=5.52$, $P=0.00016$) but no significant effect of T3 dose or interaction between time and T3 dose.

3.2. "Acceleration" paradigm: experiments involving simultaneous administration of T3 and imipramine

Basal 5-HT levels in rats administered with (a) saline by minipump over 4 weeks, (b) saline by minipump over 2 weeks and T3 by s.c. injection at 0.02 mg/kg daily for 2 weeks, (c) imipramine by minipump for 4 weeks, and (d) T3 by s.c. injection at 0.02 mg/kg daily for 2 weeks together with imipramine by minipump for 2 weeks were not different (Table 1).

Table 1
Basal 5-HT levels in frontal cortex of rats administered T3 and imipramine

Group	5-HT (fmol)/5 μl dialysate
(a) Saline 4 weeks	26.84 ± 4.12 (21)
(b) 0.02 mg/kg T3 14 days	33.83 ± 4.74 (21)
(c) 10 mg/kg/day imipramine 4 weeks	25.26 ± 11.14 (13)
(d) 10 mg/kg/day imipramine + 0.02 mg/kg T3 2 weeks	22.94 ± 3.24 (10)

Values in each case are mean ± S.E.M. of the number of observations in parenthesis.

Fig. 4 shows the effects of administration of 8-OH-DPAT (0.05 mg/kg) to the four groups of rats. An overall ANOVA of the data for the effect of challenge with 8-OH-DPAT gave a significant main effect of treatment ($F[3,19]=3.26$, $P=0.043$) and a significant interaction between time and treatment ($F[18,114]=1.72$, $P=0.045$). Planned comparison tests showed a significant main effect of T3 ($F[1,19]=4.62$, $P=0.044$) but no main effect of imipramine or interaction. Individual planned comparisons showed a significant effect of T3 vs. saline ($F[1,19]=6.66$, $P=0.018$) only.

Fig. 5 shows the effects of administration of CP 93129 to the four groups of rats. An overall ANOVA of the data for the effect of challenge with CP 93129 gave a significant main effect of treatment ($F[3,21]=6.36$, $P=0.003$) and a significant interaction between time and treatment ($F[21,147]=1.9$, $P=0.015$). Planned comparison tests showed a highly significant main effect of T3 ($F[1,21]=19.07$, $P=0.0003$) but

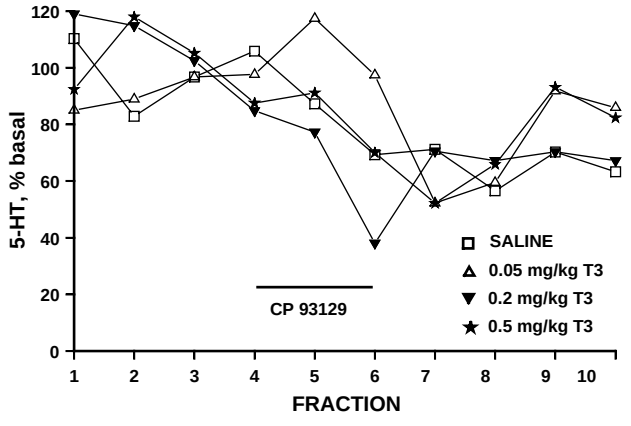

Fig. 3. Effect of CP 93129 (10 μM via the probe) on 5-HT levels in frontal cortex. Results are means of observations from six animals treated with saline, five animals treated with 0.05 mg/kg T3, six animals treated with 0.2 mg/kg T3, and six animals treated with 0.5 mg/kg T3. S.E.s are omitted for clarity.

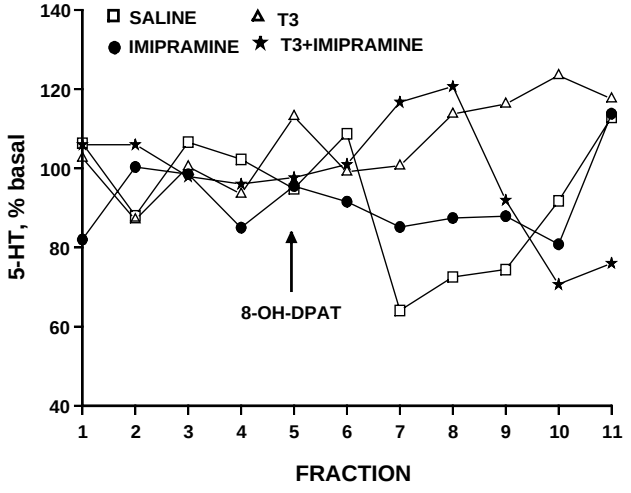

Fig. 4. Effect of 8-OH-DPAT injection (50 μg/kg s.c.) on 5-HT levels in frontal cortex. Results are means of observations from four animals treated with saline for 4 weeks, nine animals treated with 0.02 mg/kg T3 for 2 weeks, five animals treated with 10 mg/kg imipramine for 4 weeks, and five animals treated with 0.02 mg/kg T3 and 10 mg/kg imipramine for 2 weeks. S.E.s are omitted for clarity.

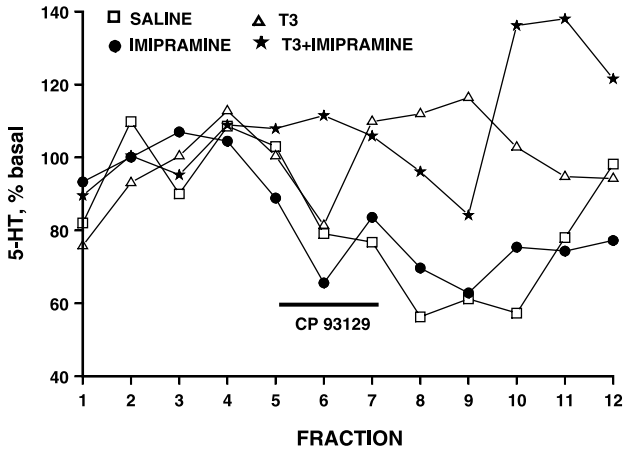

Fig. 5. Effect of CP 93129 (10 μM via the probe) on 5-HT levels in frontal cortex. Results are means of observations from seven animals treated with saline for 4 weeks, eight animals treated with 0.02 mg/kg T3 for 2 weeks, six animals treated with 10 mg/kg imipramine for 4 weeks, and five animals treated with 0.02 mg/kg T3 and 10 mg/kg imipramine for 2 weeks. S.E.s are omitted for clarity.

no main effect of imipramine or interaction. Individual planned comparisons showed a significant effect of T3 vs. saline ($F[1,21]=7.66$, $P=0.011$), a significant effect of the combination of imipramine and T3 vs. saline ($F[1,21]=11.16$, $P=0.003$), and a significant effect of the combination of imipramine and T3 vs. imipramine alone ($F[1,21]=11.42$, $P=0.003$).

4. Discussion

The present results confirm and extend our previous findings (Gur et al., 1999a) of an increase in basal cortical 5-HT levels and a decrease in sensitivity of the 5-HT_{1A} autoreceptors which control 5-HT release in frontal cortex after short-term administration of T3. Although it is now established that activation of postsynaptic 5-HT_{1A} receptors on cortical, probably glutamatergic neurons may also inhibit firing of serotonergic neurons and thus exert a negative effect on 5-HT release in nerve terminal areas (Casanovas et al., 1999; Hajos et al., 1999), the term "5-HT_{1A} autoreceptors" will be used in this discussion to denote both the 5-HT_{1A} autoreceptors located somatodendritically on serotonergic neurons in the raphe nuclei and the subset of postsynaptic 5-HT_{1A} receptors in the cortex which influence 5-HT release. While we previously showed an increase in T3 levels in frontal cortex and subsensitivity of the 5-HT_{1A} receptors which control 5-HT release in frontal cortex after administration of T3 at 0.1 mg/kg daily for 7 days by s.c. injection, the present results show that the latter effect can be obtained already at a dose of 0.02 mg/kg T3 given daily for the same period.

It remains unclear why the doses of 0.02 and 0.05 mg/kg/day of T3, which induced subsensitivity of 5-HT_{1A} autoreceptors as shown by the decrease in the ability of 8-OH-DPAT to reduce 5-HT levels in frontal cortex, did not increase basal levels of 5-HT. However, many microdialysis studies both in our laboratory and others (Invernizzi et al., 1994; Sayer et al., 1999; Cremers et al., 2000; Dawson et al., 2000; Dremencov et al., 2000; Gur et al., 1999b, 2002) have failed to show increases in basal 5-HT levels after chronic administration of antidepressant drugs, despite the fact that these treatments induced subsensitivity of 5-HT_{1A} or of 5-HT_{1B} autoreceptors.

In our experiments, local administration of the 5-HT_{1B} receptor agonist CP 93129 at a concentration of 10 μM induced an approximately 40% reduction in 5-HT levels in frontal cortex. The reduction was maintained for 2 h after cessation of the 1-h period during which CP 93129 was infused. These results confirm the earlier findings of Auerbach and Hjorth (1995), who obtained a 50% decrease in 5-HT levels in both cortex and hippocampus after infusion of 10 μM CP 93129 for 40 min. There was no effect of 7 days of T3 administration at any of the doses studied on 5-HT_{1B} receptor activity in frontal cortex, in keeping with our previous results (Gur et al., 1999a) in which a dose of 0.1 mg/kg/day was used and activity was measured with the antagonist GR 127935 rather than the agonist CP 93129.

For the experiments involving simultaneous administration of T3 and imipramine, a dose of 0.02 mg/kg T3 was chosen as this was the minimum dose effective in inducing subsensitivity of 5-HT_{1A} autoreceptors in the dose-response experiments. This dose of T3 administered daily for 2 weeks resulted in subsensitivity of both 5-HT_{1A} and 5-HT_{1B} autoreceptors. The effect on 5-HT_{1B} autoreceptor activity contrasts both with our previous results (Gur et al., 1999a) and the results of the dose-response experiments performed in the present study. In both these cases, T3 was administered for 7 days only. Heal and Smith (1988) observed a reduction in the locomotor response induced by the 5-HT_{1B} receptor agonist RU 24969 after administration of T3 at 0.1 mg/kg for 10 days, and it is thus possible that the duration of administration of T3 has a critical effect on development of 5-HT_{1B} receptor subsensitivity.

In the present experiments, chronic administration of imipramine (10 mg/kg/day for 4 weeks) did not affect basal 5-HT levels in frontal cortex. This contrasts with the increase observed after chronic clomipramine (Gur et al., 1999a), which could be explained by the relatively higher affinity of clomipramine for the 5-HT transporter compared to that of imipramine, and also with the results of Bel and Artigas (1996) who observed elevated 5-HT levels in frontal cortex after administration of 4 mg/kg imipramine daily for 2 weeks, delivered as in our experiments by osmotic minipump. However, these latter investigators carried out the microdialysis experiments with the minipumps still in place and thus delivering imipramine, while in our experiments, the minipumps were removed before measurement so as to observe long-term effects of the drug only.

The lack of effect of imipramine on 5-HT_{1A} and 5-HT_{1B} autoreceptor activity is consistent with the hypothesis of

antidepressant action put forward by Blier and de Montigny (1994, 1998) according to which tricyclic drugs and also electroconvulsive therapy do not alter the sensitivity of presynaptic autoreceptors, but instead increase the sensitivity of postsynaptic 5-HT_{1A} receptors, especially in the hippocampus. Our experiments showed that in animals which received imipramine treatment simultaneously with T3 for 2 weeks, 5-HT_{1B} autoreceptor activity was reduced to the same extent as that found in animals which had received T3 alone. 5-HT_{1A} autoreceptor activity in these animals, however, was no different to that in control animals which had received saline. The administration of imipramine for 2 weeks thus appeared to prevent the subsensitivity of 5-HT_{1A} autoreceptors induced by T3. At present, we have no explanation for the discrepancy between the effects of the combination treatment on 5-HT_{1A} and on 5-HT_{1B} autoreceptors.

In the study of Moreau et al. (2001), a dose of 4 μg/kg/day T3 administered for 21 days decreased [^{3}H]-citalopram binding to 5-HT transporter in rat midbrain, while a similar effect was given by 10 mg/kg/day imipramine administered for the same time period. Twenty-one days of 4 μg/kg/day T3 also prevented the up-regulation of postsynaptic 5-HT_{1A} receptors in hippocampus induced by 10 mg/kg/day imipramine, while in frontal cortex, the same dose of T3 given for either 7 or 21 days potentiated the decrease in 5-HT_2 receptors induced by imipramine. These investigators chose this dose of T3 as being more in the physiological range than the higher doses used by most investigators. Our present results show that the effects observed by Moreau et al. (2001) are not due to increased 5-HT levels induced by the low dose of T3, since T3 at 0.02 mg/kg/day administered either for 7 days (Fig. 1) or for 14 days (Table 1) did not significantly increase 5-HT levels in frontal cortex. The minimum dose required for an increase in basal levels thus appears to be 0.1 mg/kg/day for 7 days, as observed in our previous study (Gur et al., 1999a).

Our results provide further support for the proposal that T3 acts to accelerate and augment the effects of antidepressants by an action on the serotonergic system in the brain (Newman et al., 2000; Bauer and Whybrow, 2001; Bauer et al., 2002). There are two main lines of evidence suggesting that T3 acts in this manner rather than by increasing thyroid hormone levels and thus reversing any abnormalities in thyroid status found in depressed patients. Firstly, most depressed patients are euthyroid, and secondly, T3 is generally recognised to be far more effective than T4 both as an accelerating and as an augmenting agent, although T4 more effectively restores thyroid status and maintains T3 levels in hypothyroid patients than does T3. Furthermore, as shown by Moreau et al. (2001), T3 administration does not affect serum-free T3 levels and indeed decreases serum-free T4 levels, due to the negative feedback effect of exogenous T3 on the hypothalamo-pituitary-thyroid axis. Another possible mechanism of action of T3, in which it affects the metabolism or phramacokinetics of antidepressant drugs, was considered by Prange (1996) to be very unlikely on the basis of both animal and human studies. Furthermore, the side-effect profile of antidepressants, which is dependent on their blood levels, is not affected by T3 administration.

In summary, we have shown that doses of T3 lower than 0.1 mg/kg/day administered for 7 days are capable of inducing subsensitivity of 5-HT_{1A} autoreceptors in frontal cortex, although they did not lead to an increase in basal 5-HT levels. In addition, administration of T3 at 0.02 mg/kg/day for 14 days resulted in subsensitivity of 5-HT_{1B} autoreceptors. The action of combined treatment with T3 and a tricyclic antidepressant to accelerate the therapeutic effect of the drug may thus be due to a combination of the effects of T3 on autoreceptor sensitivity and of the drug on postsynaptic 5-HT_{1A} receptor sensitivity.

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