Effects of desipramine on regional serotonin synthesis in the rat brain: acute and chronic autoradiographic studies

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Abstract

Various studies have implicated the involvement of noradrenaline (NA) and/or serotonin (5-hydroxytryptamine (5-HT)) in the pathogenesis and treatment of depression. The aim of the present study was to investigate the effects of acute and 7 days of administration of desipramine, a NA re-uptake inhibitor, on the rate of 5-HT synthesis in the rat brain. The study was done by an autoradiographic method using α-[14C]-methyl-l-tryptophan as a tracer. The acute (10 mg/kg, i.p., 2 h before i.v. infusion of the tracer) or 7 days of desipramine (10 mg/kg per day, i.p.) did not affect plasma tryptophan (Trp) concentrations, as compared to control (saline treated) rats. Acute treatment with desipramine decreased the rate of 5-HT synthesis in the brain regions that contain 5-HT cell bodies between 19 and 28%, and increased the rate of 5-HT synthesis in the majority of areas containing 5-HT terminals between 21 and 65%. In contrast to the acute treatment, a 7-day administration increased 5-HT synthesis rates in the dorsal raphe (24%), but decreased it in raphe magnus (35%), superior olive (45%), caudate (31%), superior (38%) and inferior (53%) colliculus, and in the auditory cortex (35%). This suggests that the effect of desipramine on 5-HT synthesis rate is time-dependent and differs in the cell bodies and structures containing 5-HT nerve terminals.

Keywords: α-Methyl-l-tryptophan; Tracer; Antidepressant treatment; Serotonin synthesis; Rat brain

1. Introduction

Neurotransmitter and neuromodulator serotonin (5-hydroxytryptamine (5-HT)) has been implicated in the control of many physiological (cardiovascular, respiratory and thermoregulatory) and behavioural (feeding and sexual behaviour, circadian rhythm, sleep–wake cycle, aggression, learning and pain sensitivity) functions that could be disturbed by depression (Stahl, 1998). There is substantial evidence supporting the roles of 5-HT and noradrenaline (NA), as well as the interaction between serotonergic and noradrenergic systems, in the pathogenesis and treatment of depression (Blier and de Montigny, 1994; Bel and Artigas, 1996; Stahl, 1998). It has been proposed that drugs acting as selective NA or 5-HT re-uptake inhibitors achieve their antidepressant effects by increasing serotonergic or noradrenergic neurotransmission, and consequently enhance their input to the post-synaptic neurons in many regions of the brain to which serotonergic and noradrenergic terminals project (Blier and de Montigny, 1994).

Desipramine is a tricyclic antidepressant and active metabolite of imipramine and lofepramine, with predominantly NA re-uptake inhibiting properties (Sanchez and Hytell, 1999). It inhibits the NA transporter (Hebert et al., 2001). Desipramine is a highly lipophilic drug and has a washout period of 76 h in rats (Benmansour et al., 1999).

The reports on the effect of antidepressant drugs on 5-HT synthesis are scarce and inconsistent, presumably due to a lack of a suitable and specific method for the determination of 5-HT synthesis in the discrete brain regions. Although the understanding of the role of the neurotransmitter synthesis in the overall neurotransmission is still not clear, it has been described as one of the most important steps in it (Nelson, 1993). There are only a few reports showing that desipramine treatment decreased brain NA synthesis (Nimgaonkar et al., 1986; Moret and Briley, 1992), and did not affect (Moret and Briley, 1992) or decreased (Esteban et al., 1999) brain 5-HT synthesis. In these studies, 5-HT synthesis was equated with 5-HT turnover, expressed as a ratio of 5-hydroxyindolacetic acid (5-HIAA) and 5-HT, or...
from the accumulation of 5-hydroxytryptophan (5-HP) following the inhibition of aromatic amino acid decarboxylase by m-hydroxydihydroxyaniline (NSD-1015). In this respect, it should be noted that NSD-1015, by itself, has an effect on 5-HT synthesis (Mück-Seler and Diksic, 1995) and, therefore, in combination with certain other drugs, a confounding effect from drug-drug interaction could be present.

Using the autoradiographic method with α-methyl-l-tryptophan (α-MTrp), we have reported that the acute administration of fluoxetine differently affected 5-HT synthesis rates in the cell bodies and terminals (Tsuiki et al., 1990; Nagahiro et al., 1996). In the present study, a determination of 5-HT synthesis rates was made using the original α-MTrp autoradiographic method (Diksic et al., 1995; Nagahiro et al., 1990) that permits a measurement of the rate of 5-HT synthesis in discrete rat brain regions. The aim of this study was to evaluate the effects of the acute and 7-day administration of desipramine, a NA re-uptake inhibitor, on 5-HT synthesis in various rat brain regions using the α-MTrp autoradiographic method. 5-HT synthesis was selected because it is probably one of the most important factors contributing to 5-HT neurotransmission (Nelson, 1993).

2. Methods

2.1. Animals

Sprague-Dawley male rats (Charles River) weighing between 200–220 g were used in the acute study, whereas rats weighing 150–170 g were used in the 7-day study. The latter rats reached body weights of 200–220 g on the 8th experiment day. The rats were housed in the animal facility (room temperature 22 °C and 12 h day–night cycle) for at least 3 days before being used in the experiments. The night before the experiment, the rats fasted and water was supplied ad libitum. The tracer was injected between noon and 2 pm, and all of the rats were sacrificed between 2 and 4 pm. The mean physiological parameters (pH, pCO₂, pO₂, hematocrit, body weight) for the experimental groups were within the range of previously published values in saline treated rats (Nagahiro et al., 1990).

In the acute experiment, the rats were injected i.p. with desipramine (10 mg/kg) or saline (the control group) immediately following the surgical procedure (i.e. 2 h before the infusion of the tracer). In the 7-day protocol, the rats were injected with desipramine (10 mg/kg, i.p.) or saline once a day for 7 days. The tracer was infused 24 h after the last dose of desipramine or saline and 2 h after the surgical procedure. The body weight was recorded every day following the injection of the desipramine or saline. A minimal number of animals were used in our studies. All animal use procedures were in strict accordance with the Canadian Council on Animal Care guidelines, and were approved by the Animal Care Committee of McGill University.

2.2. Drugs

Desipramine hydrochloride (Sigma, St. Louis, MO, USA) was dissolved in saline. The tracer, α-[14C]MTrp (specific activity of about 55 mCi/mmol) was synthesized using the procedure described by Mzeengeza et al. (1993). The tracer was dissolved in saline for i.v. injection.

2.3. Experimental procedure

The femoral artery (for blood sampling) and vein (for tracer injection) were cannulated with plastic catheters under light halothane (0.5–1.0%) anesthesia. The lower body of the rat was placed in a loose-fitting plaster cast and the animals were allowed to awaken. Two hours following the surgical procedure, the tracer, α-[14C]MTrp (30 μCi (1.11 MBq) in 1 ml saline) was injected through a catheter into the femoral vein over 2 min by an injection pump. At the beginning of the tracer injection, arterial blood samples were taken at progressively increased time intervals up to the time of death. A total of 12–15 blood samples were taken. The blood samples were centrifuged for 3 min at 12,500 × g. Twenty microliters of plasma was taken for the radioactive determination by a liquid scintillation counting to measure plasma [14C]radioactivity, which was needed for the input function. Blood gases were determined with a micro-blood gas analyser (Model 178, CIBA-Corning, Canada). The animals were sacrificed using a guillotine 60 or 150 min following the tracer injection. The brains were removed, frozen in isopentane and cut into 30 μM slices in a cryostat at about −20 °C. The brain slices were mounted on glass slides, dried, and contacted to X-ray film along with 14C-polymer standards, calibrated to the brain tissue equivalent, for 3 weeks in order to obtain autoradiograms. The films were developed, and radioactivity concentrations in different structures identified with reference to a rat brain atlas using the Microcomputer Imaging Device (MCID/M4-Image Analysis System; Imaging research, St. Catharines, Ontario, Canada). Optical densities were converted into tissue tracer concentration, utilizing a calibration curve made by plotting the optical density of 14C-standards as a function of their tissue equivalent concentration.

2.4. Determination of plasma tryptophan (Trp) concentrations

At the beginning, mid-point, and at the end of each experiment, further 50 μl of plasma were deproteinized with 25 μl of 20% TCA, and a supernatant was used for the total Trp determination. Plasma ultrafiltrate (Ultrafree-MC filter with 10,000 MW cut-off span at 12,000 rpm for 10 min) was used for the determination of the non-albumin bound (free) Trp in the plasma. Trp concentrations were measured by the HPLC using a fluorescence detector (Diksic et al., 1990).
trapping constant (K) which converts the regional trapping constant of Tagliamonte et al., 1973). The LC is a conversion constant been demonstrated by many investigators to best relate to (pmol/ml) is the plasma concentration of free Trp, which has rates of 5-HT synthesis (Diksic et al., 1990, 1999; Nagahiro et al., 1990; on which the method is based are detailed in previous pub-
lications (Diksic et al., 1999), if the calculation is done in some instances (Gharib et al., 1999), if the calculation is done in a way that is equivalent to our calculation (Diksic et al., 2000), it seems that the basis of criticism is neither well founded, nor supported by the experimental data. In some expressions reservations to our approach (Gharib et al., 1999; Shou et al., 1999, 2000). As discussed in our recent publica-
tions (Bough and Faull, 1999; Diksic, 2000; Diksic et al., 2000), it seems that the basis of criticism is neither well founded, nor supported by the experimental data. In some instances (Gharib et al., 1999), if the calculation is done in a way that is equivalent to our calculation (Diksic et al., 1999), the final results accord rather well with those reported by us.

2.5 Calculation of 5-HT synthesis rate

The theoretical basis of the method and the assumptions on which the method is based are detailed in previous publi-
lications (Diksic et al., 1990, 1999; Nagahiro et al., 1990; Diksic, 2000, 2001; Diksic and Young, 2001). In brief, the rates of 5-HT synthesis (R; pmol/g min) in the brain using the α-[14C]MTrp method are calculated from the brain trapping constant (K; ml/g min−1), and a conversion factor (LC) of 0.42 (Vanier et al., 1995); R = CpK/LC. This conversion is analogous to the procedure used with labelled Trp and the separation of different metabolites in dissected brain tissues (Lin et al., 1969). In the present study, Cp (pmol/ml) is the plasma concentration of free Trp, which has been demonstrated by many investigators to best relate to brain Trp and brain 5-HT concentrations (Salter et al., 1989; Tagliamonte et al., 1973). The LC is a conversion constant which converts the regional trapping constant of α-MTrp (K) to the regional constant for metabolic conversion of Trp to 5-HT (K; ml/g min−1); K = K/LC. By definition, the LC = K/LC. This conversion constant was determined in vivo in the rat brain and it was found to be constant throughout the brain (Vanier et al., 1995). Some researchers have expressed reservations to our approach (Gharib et al., 1999; Shou et al., 1999, 2000). As discussed in our recent publications (Bough and Faull, 1999; Diksic, 2000; Diksic et al., 2000), it seems that the basis of criticism is neither well founded, nor supported by the experimental data. In some instances (Gharib et al., 1999), if the calculation is done in a way that is equivalent to our calculation (Diksic et al., 1999), the final results accord rather well with those reported by us.

2.6 Statistical analysis

The data are expressed as mean ± S.D. The differences between the groups (body weights, free or total Trp) were assessed by one-way ANOVA, followed by a Tukey’s multiple comparison test. The statistical evaluation of the data was performed using a one-group two-tailed t-test on the ra-
tios of the synthesis rates in the two groups. The main effects between the control and respective treatment groups were compared using the two-tailed Student’s t-test, in which the observed ratios between the control and treated groups of 5-HT synthesis were compared with the null hypoth-
esis of the ratios being equal to 1 ± 0 (i.e. no difference between the control and treated groups) with a S.D. of 0. The criterion for significance was P > 0.05. As dis-
cussed in our previous publication (Yamane et al., 2001), the measurements in the individual brain structures are not independent variables and have a high correlation between the rates in different brain structures and, as such, a cor-
rection for the Type I error is not possible (Tabachnick and Fidell, 1996). The statistical analyses were done using SYSTAT-9 (SPSS Inc., 2000) and SigmaStat-2.03 (SPSS Inc., 1999).

3. Results

Plasma free or total Trp concentration did not differ significantly (P > 0.05; one-way ANOVA) following the adminis-
tration of a single dose of desipramine, or follow-
ing a 7-day administration of desipramine as compared to the corresponding control groups. In the acute experiments, plasma free and total Trp concentrations were 10.3 ± 1.7 and 90 ± 22 nmol/ml in the treatment groups, and 10.8 ± 3.1 and 104 ± 12 nmol/ml in the control groups. In the 7-day treatment protocol, plasma free and total Trp concentrations were 8.8 ± 2.2 and 78 ± 25 nmol/ml in the treatment groups, and 9.1 ± 1.6 and 75 ± 25 nmol/ml in the control groups. During the 7-day treatment of desipramine, a significant increase in the body weight was observed, both in the control and desipramine treated rats, but no significant difference in body weight between the control and desipramine treated rats was found.

A set of representative autoradiograms obtained in the control (inserts A, B, E and F) and treatment (inserts C, D, G and H) groups in the acute (A–D) and 7-day (E–H) exper-
iments are provided in Fig. 1, showing that 5-HT synthesis rates are not uniform throughout the rat brain. The 5-HT synthesis rate in the raphe nuclei is substantially greater than in other brain areas which receive projections from the raphe nuclei. The 5-HT synthesis in some structures like the cortex and caudate is not homogenous throughout these structures: the layer VI in the cortex and the medial part of the caudate have higher 5-HT synthesis rates than other parts of these structures.

In the acute experiments, the mean ration between synthe-
sis in control to those in the treatment group was 1.00 ± 0.23 (N = 26; P < 0.05 for ratios being >1). The main reason for the ration not indicating significant global influence on 5-HT synthesis is the fact that the synthesis in some structures is >1 while is smaller than 1 in other structures. However, acute administration of desipramine significantly decreased the rate of 5-HT synthesis in all raphe nuclei (Table 1). In the 7-day treatment experiments the mean ration between syn-
thesis in the control and that in treated rats was 1.21 ± 0.31 (N = 26; P < 0.001) suggesting a significant global influ-
ence of desipramine on 5-HT synthesis. Repeated admin-
istration of desipramine elicited a significant increase and decrease in the dorsal and magnus raphe, and there was no rate change in the median raphe (Table 2). A single dose of desipramine significantly increased 5-HT synthesis rates in all cortices except the visual cortex (Table 1), while repeated desipramine administration decreased the rate solely in the auditory cortex (Table 2). In the basal ganglia, acute admin-
istration (Table 1) significantly increased the 5-HT synthe-
sis rates in the globus pallidus and lateral caudate, while repeated desipramine administration significantly decreased the rate of 5-HT synthesis in the medial part of the caudate only (Table 2).

Neither the acute (Table 1) nor 7-day (Table 2) adminis-
tration of desipramine significantly affected 5-HT synthesis
Fig. 1. A set of representative autoradiograms obtained in rats used in the acute and 7-day treatments: control rats (A, B, E and F) and rats treated with desipramine (10 mg/kg, i.p.; C, D, G and H). The autoradiograms exemplify the regional 5-HT synthesis rates calculated from the trapping constant of \( \text{H}^{[14]} \text{C} \)-methyl-\( \text{L} \)-tryptophan. The pixel \( \times \) pixel synthesis rates for this pictorial presentation were calculated using the brain average value for the precursor pool. However, the data provided in Tables 1 and 2 were calculated by using the precursor pool as a variable (Diksic et al., 1995). No special attempt was made to get the slices from the same levels of the brain. The structure abbreviations are: AC, nucleus accumbens; CM, caudate-medial; CL, caudate-lateral; DR, dorsal raphe; MR, median raphe; P, pineal body; VCx, visual cortex; SMCx, sensory-motor cortex.

Rates in the dorsal and ventral thalamus, dorsal and ventral hippocampus, and the substantia nigra reticulata. Acute desipramine (Table 1) administration induced a significant increase in 5-HT synthesis rates in the medial geniculate body, and there was no change in the lateral geniculate body. Repeated desipramine administration (Table 2) did not significantly affect 5-HT synthesis rates in both parts of the geniculate body.

The significant decrease in 5-HT synthesis rates in the ventral tegmental area (VTA) following acute administration (Table 1) was not found after repeated desipramine administration (Table 2). Acute (Table 1) or repeated desipramine administration (Table 2) did not significantly affect 5-HT synthesis rates in the medial forebrain bundle or in the hypothalamus. Acute desipramine administration (Table 1) significantly
increased 5-HT synthesis rates in the superior colliculus and significantly decreased it in the superior olive. Repeated desipramine treatment significantly decreased 5-HT synthesis rates in the superior and inferior colliculus and in the superior olive (Table 2).

4. Discussion

In the present study, we found region-specific and time-dependent effects of desipramine (acute and 7-day treatment) on 5-HT synthesis rates in the rat brain, using the autoradiographic method with α-MTrp as a tracer (Diksic et al., 1990; Nagahiro et al., 1990; Diksic, 2001) This method permits in vivo measurements of the rate of 5-HT synthesis in many brain regions with a good anatomical resolution and without administration of any auxiliary drugs, such as NSD 1015, that could interfere with the results (Mück-Šeler and Diksic, 1996).

Our results obtained in the control animals show that 5-HT synthesis was greater in the cell bodies than in the structures containing 5-HT terminals, which is in line with our previous data, as well as with the report that measured 5-HT turnover in the rat brain (Neckers and Meek, 1976). The acute administration of desipramine affected 5-HT synthesis in many brain regions with a good anatomical resolution (Mück-Šeler and Diksic, 1996). Such effects were observed in the structures with high 5-HT turnover like the hippocampus, cerebral cortex, substantia nigra, and raphe, as well as regions with moderate turnover like the substantia nigra, the subthalamic nuclei, and the ventral tegmental area (VTA).

Table 1

<table>
<thead>
<tr>
<th>Structures</th>
<th>Control</th>
<th>Desipramine</th>
<th>Differences*</th>
<th>P-values</th>
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<tr>
<td>Raphe Dorsal</td>
<td>147 ± 2 ± 22</td>
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Table 2

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<tr>
<td>Inferior colliculus</td>
<td>40 ± 15</td>
<td>19 ± 8</td>
<td>−53</td>
<td>&lt;0.002</td>
</tr>
<tr>
<td>Superior olive</td>
<td>39 ± 14</td>
<td>22 ± 10</td>
<td>−45</td>
<td>&lt;0.005</td>
</tr>
</tbody>
</table>

* Differences are given as the percent (%) of the synthesis rate in the corresponding control (Student’s two-tailed t-test). NS, non-significant, N, number of animals, VTA, ventral tegmental area; MFB, medial forebrain bundle.

Data are given as mean ± S.D.

Table 1: Chronic effects of desipramine (10 mg/kg per day, i.p., for 7 days, N = 11) or saline (control, N = 11) on the 5-HT synthesis rate (pmol/g min) in the rat brain regions given as mean ± S.D.
synthesis in the soma (nuclei raphe) and terminals (the cortex, basal ganglia, geniculate body, VTA, and superior colliculus) of 5-HT neurons in a dual and regional-dependent manner. A single administration of desipramine decreased 5-HT synthesis rates in the dorsal, median and magnus raphe. This is similar to the acute effects of the 5-HT re-uptake inhibitors fluoxetine (Mück-Seler et al., 1996) and paroxetine (Yamane et al., 2001), the 5-HT1A agonist buspirone (Okazawa et al., 1999), the simulator of the release and the inhibitor of the 5-HT re-uptake d-fenfluramine (Mück-Seler and Döskis, 1996; Yamane et al., 1999), and the partial 5-HT1A receptor agonist, WAY100135 (Tohyama et al., 2001) on 5-HT synthesis rates. This decrease in the synthesis in the raphe nuclei may be related to the small affinity of desipramine to 5-HT re-uptake sites (Frazier, 2001), and the possible increase in the extraneuronal 5-HT which would, in turn, decrease 5-HT synthesis through 5-HT1A autoreceptors present in the cell bodies and dendrites. Similarly, as mentioned earlier, this could be the case for other drugs known to elevate extraneuronal 5-HT levels (Mück-Seler et al., 1996; Mück-Seler and Döskis, 1996; Yamane et al., 1999, 2001; Okazawa et al., 1999; Tohyama et al., 2001). Of course, a reduction in 5-HT synthesis could also be the result of direct noradrenergic projection to these cell bodies from the locus coeruleus (Baraban and Aghajanian, 1981). In contrast to the desipramine-induced reduction of 5-HT synthesis in the cell bodies, desipramine increased the rate of 5-HT synthesis in the cell bodies, desipramine increased the rate of 5-HT synthesis (between 21 and 65%) in the majority of rat brain areas containing 5-HT terminals. Increased 5-HT synthesis rates in the cortical areas following desipramine discords with the lack of effect of acute desipramine on 5-HT synthesis in the dissected rat cortex (Moret and Briley, 1992; Esteban et al., 1999). The discrepancies between the studies may be explained by the different methods used for the determination of 5-HT synthesis rates, i.e. the accumulation of 5-HTP following (NSD-1015 administration (Moret and Briley, 1992; Esteban et al., 1999), and differently designed studies (oral administration of desipramine 90 min before decapitation; Moret and Briley, 1992). Desipramine increased 5-HT synthesis rates in the cortical structures and parts of the basal ganglia, suggesting that a blockade of NA re-uptake triggers the increase of 5-HT synthesis. A similar increase in the cortical structures was observed in rats treated with fluoxetine (Mück-Seler et al., 1996), zimelidine, 8-OHDPAT, buspirone, and fibanserin (Brambilla et al., 1999; Esteban et al., 1999), indicating that acute desipramine treatment activates the inhibitory α2C-autoreceptors and α1A-heteroreceptors which could then stimulate 5-HT synthesis. The elevation of the synthesis in the terminal areas may also be a consequence of desipramine preventing sufficient amounts of 5-HT to be recycled, that then need to be replenished by a de novo synthesis of 5-HT.

In line with the effects of acute and repeated administration of various classes of serotonergic drugs on 5-HT synthesis rates, including buspirone (Okazawa et al., 1999), WAY100135 (Tohyama et al., 2001), fluoxetine (Mück-Seler et al., 1996), or d-fenfluramine (Yamane et al., 1999), desipramine had an opposite effect on 5-HT synthesis rates when given acutely or for 7 days. Our results, which accord with those of Esteban et al. (1999), suggest that 7 days of desipramine administration induces a somewhat less apparent but different change in 5-HT synthesis rates in the 5-HT regions of the cell bodies and nerve terminals. Repeated desipramine administration in the majority of the brain structures investigated normalizes the 5-HT synthesis rates. This is probably mediated by adaptive changes such as the desensitization of the α2-adrenoceptors on NA neurons in the locus coeruleus, as a result of a direct blockade of the NA transporter (Tanda et al., 1996), and/or the down-regulation of the cortical 5-HT1A (Strinsv et al., 2001), 5-HT2 or the α1-adrenoceptors (Goodnough and Baker, 1994). In contrast to the effects of paroxetine (Yamane et al., 2001), fluoxetine (Mück-Seler et al., 1996), d-fenfluramine (Yamane et al., 1999), and buspirone (Okazawa et al., 1999), which all decreased 5-HT synthesis in the raphe region, the 7-day administration of desipramine increased 5-HT synthesis rates in the dorsal raphe, but decreased it in the raphe magnus. The was no significant difference between the control and desipramine treated rats in the median raphe. Hence, 5-HT synthesis, in the dorsal raphe, which is directly innervated by the NA neurons (Baraban and Aghajanian, 1981), is probably modulated differently by an increase of NA concentration after a repeated administration of the NA re-uptake inhibitor, then following repeated administration of drugs known to increase extraneuronal 5-HT (Mück-Seler et al., 1996; Okazawa et al., 1999; Yamane et al., 1999, 2001). The finding that desipramine increased 5-HT synthesis in the dorsal raphe may be a rather interesting observation, however, unexpected which could be related to the antidepressant properties of desipramine. The increase in 5-HT synthesis in the dorsal raphe would likely be contributing to the increased neurotransmission as a result of greater amounts of 5-HT released to the neuronal cleft.

The finding that repeated desipramine administration did not affect 5-HT synthesis rates in the hypothalamus, nucleus accumbens, hippocampus and prefrontal cortex is consistent with the unaltered tissue levels of 5-HT and 5-HIAA (Zangen et al., 1997), and the unchanged density of 5-HT transporters (Hebert et al., 2001) in these regions following chronic desipramine treatment. The loss of the ability of repeated desipramine treatment to alter 5-HT synthesis in the hippocampus is supported by the lack of its effects on basal 5-HT levels in dialysates collected from the rat hippocampus (Hagos-Korcok et al., 2000). However, the same effect was not found by Yoshioka et al. (1995), or Esteban et al. (1999) who found a decreased 5-HTP synthesis in the hippocampus during 1 and 2 weeks of treatment. The same authors found that there is no effect of desipramine on 5-HT synthesis after 3 weeks of treatment.

Serotonin synthesis in the brain depends on different factors such as Trp availability, the activity of the rate-limiting
reported that desipramine administration in 5-HT synthesis rates is not related to the alteration of MAO activity. In addition, the changes in 5-HT synthesis rates following a single or short term desipramine administration could not be ascribed to the alterations in Trp availability, as the plasma free or total Trp concentration between desipramine and saline treated rats did not differ significantly. On the other hand, the direct inhibitory effect of the increased NA levels on tryptophan hydroxylase activity (Martinez et al., 2001) and consequently on 5-HT synthesis could not be excluded, particularly in short term desipramine administration. It has been reported that desipramine affects 5-HT synthesis in the cerebral cortex and hippocampus (Esteban et al., 1999). However, it seems that desipramine treatment has different influence on the desensitization of \( \alpha_2 \)-adrenoceptors in these two structures, with desensitization in the cortex but not in the hippocampus of rat.

Somatodendritic 5-HT1A autoreceptors, located in the raphe nuclei, play an important role in the regulation of 5-HT synthesis (Brambilla et al., 1999), release and firing rate (Blair and de Montigny, 1994). This accords with the decrease in the 5-HT synthesis rates in almost all of the rat brain regions following the acute administration of the selective 5-HT1A agonist buspirone (Okazawa et al., 1999), and partial 5-HT1A receptor agonist WAY100135 (Tohyama et al., 2001). Although desipramine has a relatively low affinity for 5-HT receptors and binding sites as compared to the NA re-uptake sites (Sanchez and Hytell, 1999), its effects may be mediated, in part, through the changes associated with 5-HT receptors and re-uptake sites. In line with that, the chronic administration of desipramine down-regulates cortical 5-HT2 receptors (Goodnough and Baker, 1994), decreases the number and affinity of \([1^H]5\)-HT binding sites in the cerebral cortex and the hippocampus, and reduces the cortical 5-HT1A receptors and their second messenger system (adenyl cyclase activity), suggesting a receptor-mediated action (Srinivas et al., 2001). This would suggest that desipramine may have a direct influence on serotoninergic neurotransmission and likely on 5-HT synthesis through some of these receptors.

Despite the fact that the noradrenergic system is the main site of desipramine action (Sanchez and Hytell, 1999), the data of the present study suggests that the effects of desipramine on the serotoninergic system and 5-HT synthesis cannot be neglected. Further, the central serotoninergic and noradrenergic systems are closely interconnected (Hadjerti et al., 1997) and 5-HT cell bodies in the raphe nuclei and NA cell bodies in the locus coeruleus are innervated by NA (Abarbanell and Aghajanian, 1981; Gravel and de Montigny, 1987) and 5-HT (Hadjerti et al., 1997) nerve terminals, respectively. Desipramine-induced changes in 5-HT synthesis rates may be explained by the interactions (cross-talk) between the brain monoamine systems (Reith et al., 1997) that include alterations in the pre-synaptic auto- and hetero-receptor reactivity elicited by increased extracellular concentration of NA (Yoshikawa et al., 1995; Tanda et al., 1996; Perry and Fuller, 1997; Reith et al., 1997). One cannot exclude the possibility that the effect of desipramine on 5-HT synthesis occurs through action on the 5-HT re-uptake sites, despite considerable greater affinity (selectivity of about 240) for the NE re-uptake sites (Frazier, 1997; 2001). However, Hebert et al. (2001) reported that chronic desipramine administration does not affect the density of 5-HT transporters in rat cortical areas and the raphe region. Hebert et al. (2001) concluded that desipramine affects 5-HT synthesis rates through a blockade of the NA transporter. On the basis of the present data, we cannot definitively state the mode of desipramine action, but we can say that it does have an affect on regional 5-HT synthesis, which could be achieved through noradrenergic and/or serotoninergic receptors sites, known to influence 5-HT synthesis.

In conclusion, the results of our study show region-specific and time-dependent effects of single and repeated (7-day) administration of desipramine on the rate of 5-HT synthesis in discrete rat brain regions. The acute effects of desipramine on 5-HT synthesis rates could be the consequence of a direct increase of NA and possibly an increase of 5-HT concentration in the synaptic cleft that induces the activation of pre-synaptic adrenoceptors and 5-HT1A-autoreceptors, which then increase the synthesis of 5-HT in terminals and decrease it in the soma of 5-HT neurons. The repeated desipramine administration effects are likely achieved by a desensitization of the \( \alpha_2 \)-adrenergic heteroreceptors on 5-HT terminals that decrease NA inhibitory input to 5-HT terminals, or by the desensitization of somatodendritic 5-HT1A autoreceptors, eliciting a decrease in 5-HT synthesis in the terminals and an increase in the dorsal raphe region. The data suggests that the central 5-HT system can be modulated, at least as far as 5-HT synthesis is concerned, by the NA system.

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References


