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REGIONAL DISTRIBUTION AND ELIMINATION KINETICS
OF IMIPRAMINE IN RAT BRAIN

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The elimination of imipramine (IMP) from the brain regions was investigated in rats after a single intraperitoneal administration. The levels of IMP (ng/g wet tissue) in the brain regions reached concentrations approximately 30-fold higher than the plasma level 0.5 h after the administration. Brain regions could be divided into two classes as to IMP elimination: IMP in the cerebellum, mid-brain and medulla, striatum, posterior cortex and frontal cortex was eliminated in a mono-exponential manner the similar to the plasma level while the disappearance in the hypothalamus, thalamus, hippocampus and nucleus accumbens followed a biexponential profile.

KEYWORDS — imipramine; rat brain region; elimination; distribution; pharmacokinetics; intraperitoneal dose

INTRODUCTION

Imipramine (IMP), a tricyclic antidepressant, is widely used in the management of endogenous depression. Although the pharmacokinetics of IMP and distribution of IMP in the brain have been investigated extensively,¹⁻⁶⁾ there is little information available on its elimination from the brain regions. The brain is a target organ of IMP so its disappearance pattern is pharmacodynamically important. The presence of specific high-affinity binding sites for IMP has been demonstrated in the brain by in vitro experiments.⁷⁻¹³⁾ Biochemical studies suggest that serotonergic presynaptic terminals are primary targets for IMP binding.¹⁰⁻¹³⁾ However, the role of the IMP binding sites in its clinical action is not yet clear. In this study, we investigated the disappearance of IMP from certain brain regions after a single intraperitoneal dose in rats and it was shown that the regions can be divided into two classes regarding IMP removal.

MATERIALS AND METHODS

IMP hydrochloride and clomipramine hydrochloride were obtained from Ciba-Geigy (Japan) Limited (Tokyo, Japan). All other chemicals were analytical grade products available commercially.

Male Wistar rats, weighing 200-230g, were dosed i.p. with IMP (5mg/kg).

At various intervals after the administration (0.25, 0.5, 1, 2, 4, 6, 8 and 12 h), the animals were sacrificed. Blood samples were collected in heparinized tubes and the plasma was stored at -30°C . The brain was quickly removed and was dissected into 9 regions according to the technique of Glowinski and Iversen.¹⁴⁾ These regions were cerebellum, mid-brain and medulla, hypothalamus, thalamus, striatum, hippocampus, posterior cortex, nucleus accumbens and frontal cortex. After being weighed the brain regions were also stored at -30°C until the analysis.

The IMP levels in the plasma and brain regions were assayed according to the method of Elik et al.¹⁵⁾ with slight modification. For extraction, to 1 ml of plasma were added 1 ml of 0.5N NaOH, 100 μl of methanol containing 1 $\mu\text{g}/\text{ml}$ of clomipramine hydrochloride as an internal standard and 5 ml of hexane-isopropyl alcohol (98.3:1.7, v/v). The mixture was shaken for 10 min, centrifuged, and the organic layer was transferred to another centrifuge tube. After the addition of 2 ml of 0.1N HCl, the mixture was shaken and centrifuged again. The organic phase was discarded and the aqueous phase was made alkaline with 1 ml of 1M $\text{Na}_2\text{CO}_3\text{-NaHCO}_3$ (pH 9.8), then reextracted with 1 ml of hexane-isopropyl alcohol. The extracts were evaporated to dryness. The residue was redissolved in 20 μl of methanol and a 2 μl aliquot was injected into the gas chromatograph (GC) under conditions described below. Each brain region was homogenized in 1.5 ml of saline solution using the sonicator 5202-PZT (Ohtake, Tokyo, Japan), and then processed in the same way as the plasma.

IMP was determined by a GC (Shimadzu GC-7A, Shimadzu, Japan) equipped with an alkali-flame ionization detector (Shimadzu FTD-8, Shimadzu). The 2.1 m x 3 mm i.d. glass column was packed with 3% OV-17 on Chromosorb W (AW-DMCS), 80-100 mesh. The analysis was carried out isothermally with the oven temperature at 280°C and the injector and detector at 300°C , and with the carrier gas (He) flow at 50 ml/min. The hydrogen and air flows were optimized for the selected oven temperature and carrier gas flow. The chromatograph was recorded and the peak heights were computed with a Shimadzu Chromatopack C-R 2AX (Shimadzu).

The pharmacokinetic data were calculated using a weighted non-linear least squares fitting program on a Sharp MZ-80B computer.

RESULTS AND DISCUSSION

After the injection of 5 mg/kg of IMP in rats, the time course of IMP concentrations in the plasma and in the 9 brain regions, cerebellum, mid-brain and medulla, striatum, posterior cortex, frontal cortex, hypothalamus, thalamus, hippocampus and nucleus accumbens, were examined. The results for the plasma, the first 5 brain regions (termed class A) and the last 4 regions (termed class B) are shown in Figs. 1, 2 and 3, respectively. Each point represents the mean of 5 animals. Under these experimental conditions, the IMP plasma level (ng/ml) was reduced rapidly following a monoexponential profile with an elimination rate constant of 0.52 h^{-1} .

The IMP levels in the brain regions (ng/g wet tissue) reached concentrations approximately 30-fold higher than the plasma level 0.5 h after the administration, indicating marked accumulation in the brain. IMP levels in the class A brain regions showed monoexponential elimination in a manner similar to the plasma level and the elimination rate constant was 0.47 h^{-1} (Fig. 2). On the other hand, the disappearance curve in the class B brain regions followed a biexponential profile

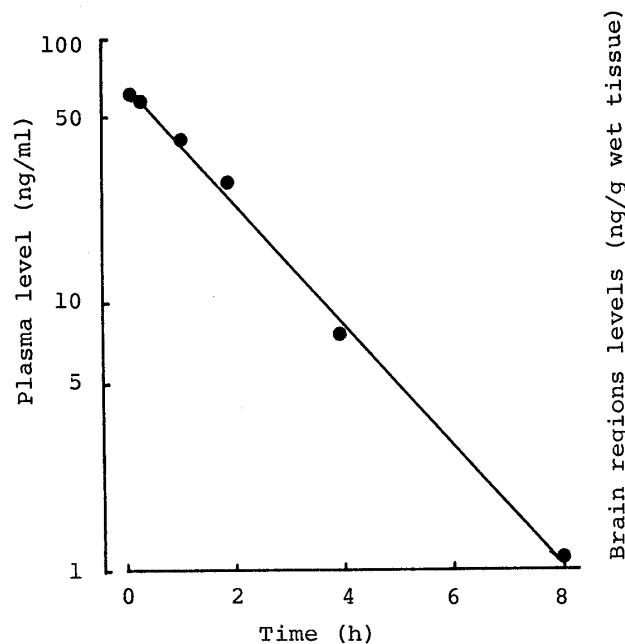


Fig. 1. Time Course of Imipramine Plasma Level after i.p. Administration of 5 mg/kg of Imipramine

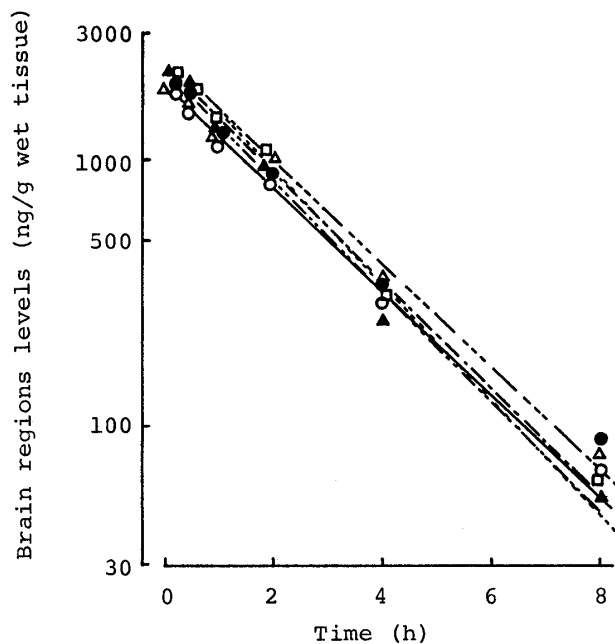


Fig. 2. Time Course of Imipramine Levels in Class A Brain Regions after i.p. Administration of 5 mg/kg of Imipramine

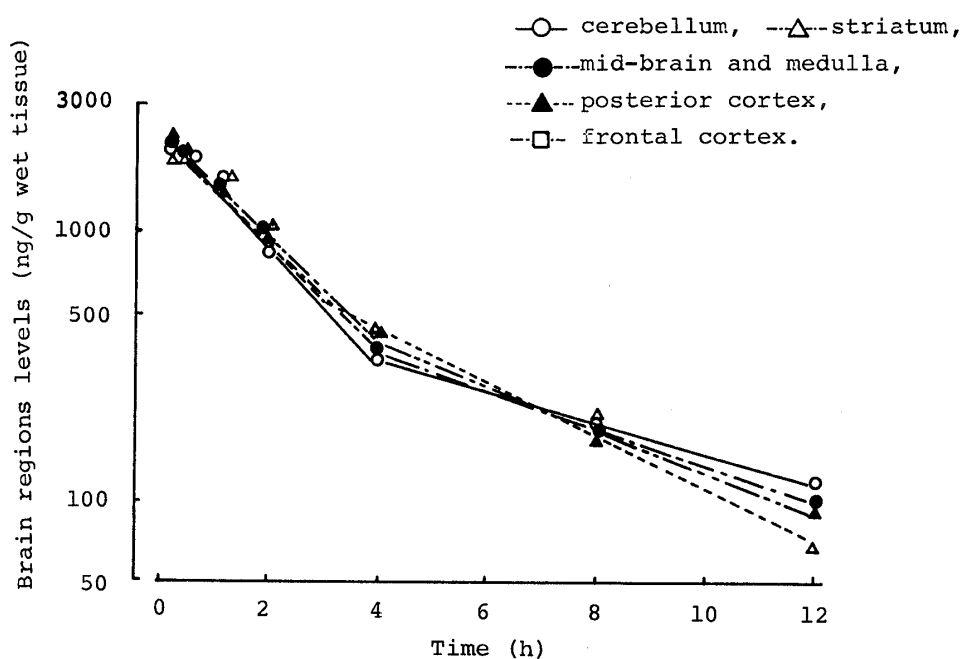


Fig. 3. Time Course of Imipramine Levels in Class B Brain Regions after i.p. Administration of 5 mg/kg of Imipramine

—○— hypothalamus, —●— thalamus,
 ---△--- hippocampus, ---▲--- nucleus accumbens.

and could be resolved into two major components (Fig. 3). The refraction points appeared 3-4 h after the administration. The initial phase (α -phase) of the curves in the class B regions was similar to those in the class A regions. In contrast, regarding the later phase (β -phase), the IMP was cleared more slowly from the class B regions with an elimination rate constant of 0.14 h^{-1} and was maintained at around 100 ng/g wet tissue even at 12 h after the administration. The initial concentration of β -phase ($C_{0\beta}$) was $0.60 \text{ }\mu\text{g/g}$ wet tissue. It is interesting that the brain regions can be divided into two classes regarding IMP disposal in vivo. The class B brain regions may be different from the class A brain regions regarding the binding affinity for IMP. The specific high-affinity sites for IMP shown by in vitro experiments⁷⁻¹³⁾ are considered to act as effective sites. Additional studies are in progress to examine the relationship between the binding of IMP in the class B brain regions and the high-affinity binding sites previously demonstrated.

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