HT ($K_D = 2 \times 10^{-9}$M) and a second, saturable and reversible site of less affinity with a corresponding K_D close to 2 to 3×10^{-8}M. Various plotting systems of the binding curve indicate for the second site a positive cooperativity, the Hill coefficient being 3.7. Dissociation rates are quite high for both ligands, however it is much higher for 5-HT than for LSD; both are temperature-dependent with a respective Q_{10} close to 2.5 and 3. Regional distributions of binding capacities for LSD and 5-HT are very similar. They are not homogenous within the brain but vary according to the studied region, e.g. in decreasing order: striatum, hippocampus, cortex, raphé, cerebellum.

Specific lesions of the tryptaminergic system have been performed by stereotaxic injections of 5–6 dihydroxytryptamine within raphe and anterior ventricles; their efficacy has been controlled by inhibition of the uptake of 5-HT. In these conditions, lesions do not modify significantly binding of one or the other ligand; this might indicate a postsynaptic location of the corresponding site.

Comparative assays of the specificity of the high affinity sites for 5-HT and LSD indicate they are related to the tryptaminergic structure but some differences are observed, i.e. bromlysergamide and cyproheptadine are more efficient in displacing LSD than 5-HT (respective ID_{50}s are 3×10^{-8} M and 6×10^{-7}M for LSD, 1.5×10^{-6} and 6×10^{-6} for 5-HT).

Detailed studies of interactions between 5-HT and LSD on these high affinity sites and preliminary assays involving various pretreatments of the membranes might indicate that the different sites observed correspond to an agonist and an antagonist conformation of the same 5-HT receptor-site.

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Catecholamine-stimulated prostaglandin synthesis in rat brain synaptosomes

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Although prostaglandins (PGs) have been shown to be synthesized in brain homogenates from a number of species, little information is available concerning the subcellular distribution and activity of the relevant enzymes (Raffel, Clarenbach, Peskar & Hertting, 1976). It has been suggested that PGE_2 may function as a modulator of synaptic transmission in the periphery, where prostaglandin inhibits noradrenaline (NA) release from sympathetic structures (Hedqvist, 1976); it was therefore of interest to find a facilitation of [^{3}H]-NA release from rat brain synaptosomes in the presence of low concentrations of PGE_2 (Roberts & Hillier, 1976).

Since the signal for PG release in the peripheral sympathetic system is considered to be associated with the postsynaptic actions of noradrenaline, in this study we have investigated the effects of several

Table 1 Stimulation of PGE synthesis in rat brain synaptosomes

Treatment		*Control (ng PGE)*	*Treated (ng PGE)*	*% stimulation*
Noradrenaline	100 μM (6)	2.41 ± 0.37	6.3 ± 0.84*	161
Dopamine	100 μM (6)	2.52 ± 0.37	5.14 ± 0.82†	103
Adrenaline	100 μM (3)	1.93 ± 0.14	4.7 ± 0.44*	143
Acetylcholine +physostigmine	100 μM 10 μM (2)	2.1	2.2	—
5-HT	100 μM (2)	2.1	1.65	—
K^+	50 mM (5)	1.94 ± 0.28	1.72 ± 0.18	—

Results are expressed as means ± s.e. with the number of experiments in parentheses. Levels of significance refer to differences from the results of the control group. * $P < 0.01$; † $P < 0.02$.

neurotransmitter candidates on the PG synthesizing ability of synaptosomes.

Twenty per cent (w/v) homogenates of rat brain tissue (excluding cerebellum) were prepared in 0.32 M sucrose, and crude synaptosomes (P_2 fraction) prepared as described by Whittaker (1969). An aliquot (200 µl) of the synaptosome suspension (containing approx. 4 mg protein) was added to 1.3 ml Krebs bicarbonate medium (pH 7.4) containing ascorbic acid (1 mg/ml), isoniazid (10 µM) and, where appropriate, the substance under investigation (Table 1). Synaptosomes were incubated for 5 min at 37°C under 95% O_2 : 5% CO_2 followed by rapid centrifugation. To each supernatant was added marker amounts (2–5 pg) of [^{3}H]-PGE_2 or [^{3}H]-PGF_2 and the PG's released into the incubation media assayed, using a modification of the method of Hillier & Dilley (1974). Briefly, this involves selective extraction, column chromatography separation of PGE and PGF groups and radioimmunoassay using antibodies with relatively good selectivity for the PG's under investigation.

The results show that 100 µM amounts of NA, dopamine and adrenaline signficantly stimulate the generation of PGE by synaptosomal tissue. In another experiment NA also significantly stimulated PGF synthesis.

The effect is selective as acetylcholine, 5-HT and K^+ depolarization were without a stimulatory effect in this system.

We conclude that (a) contrary to the findings of Raffel *et al.* (1976) noradrenaline and dopamine do stimulate prostaglandin formation in rat brain synaptosomes, (b) this strengthens the link for PGEs exerting a positive feedback on NA release on central nerve terminals as described by Roberts & Hillier (1976).

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Ontogenesis of muscarinic receptor sites in rat brain

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Cholinergic neurotransmission in the central nervous system does not appear to be fully functional until the second week of life. Thus rats do not respond to injections of scopolamine with changes in locomotor activity until after the 15th day of life (Campbell, Lytle & Fibiger, 1969) and whilst scopolamine has a synergistic effect on amphetamine-induced gnawing in 30-day-old rats, this synergism could not be seen in 10-day-old rats (McGeer, Fibiger & Wickson, 1971). Both acetylcholinesterase and choline acetyltransferase (ChAc) are present in the rat neostriatum 4 days after birth although ChAc activity is very low (McGeer *et al.*, 1971; Guyenet, Beaujouan & Glowinski, 1975). However, it is possible that the lack of response to scopolamine indicates the absence of the muscarinic cholinoceptor on the postsynaptic neuron rather than the immaturity of the presynaptic pathway. This possibility has been examined by studying the binding of [^{3}H]-atropine to homogenates of neonatal rat cerebral cortex and caudate nucleus.

Homogenates of cerebral cortex and caudate nucleus in Krebs-Henseleit solution were preincubated for 15 min at 37°C before the addition of [^{3}H]-atropine (Radiochemical Centre, Amersham; 245 mCi mmol^{-1}). After a further 15 min the incubation was terminated by centrifugation at 14,000 *g* for 30 seconds. The surface of the pellet was rinsed twice with Krebs-Henseleit solution and radioactivity in the pellet was determined by liquid scintillation counting. Two series of samples were incubated together and one of these series contained 10 µM propylbenzilylcholine, a potent antimuscarinic compound, throughout the experiment. Receptor-specific binding of [^{3}H]-atropine was taken to be that fraction of binding which was abolished by the presence of the propylbenzilylcholine.