tryptaminergic nerves, prevented the accumulation of [^{3}H]-5-HT in supra-ependymal nerve terminals. However, desipramine, a known catecholamine-uptake blocker, had no effect on this accumulation.

The demonstration of a specific uptake mechanism for 5-HT in supra-ependymal nerve terminals confirms the tryptaminergic nature of these nerves (Richards, Lorez & Tranzer, 1973; Lorez & Richards, 1973; Richards & Tranzer, 1974; Lorez & Richards, 1975). The physiological significance of this uptake mechanism is likely to be the removal of the amine from the vicinity of the effector organ (periventricular target cell) in order to terminate the possible neurotransmitter action of 5-HT. The presence of 5-HT in CSF (Holman & Vogt, 1972), probably secreted in part by supra-ependymal nerve terminals, suggests that there may be specific receptors and a physiological role for this amine in periventricular brain regions. This may also be true of the human brain since supra-ependymal nerve terminals have recently been observed in the lateral and fourth ventricles of human postmortem tissue by electron microscopy; it might be expected that these nerves also store and accumulate 5-HT although this has to be demonstrated.

Since the hallucinogenic drug DLSD is known to have a high affinity for 5-HT (Bennett & Snyder, 1975) and DA (Creese, Burt & Snyder, 1976) receptors, the localization of binding sites for [^{3}H]-DLSD was studied by autoradiography in order to identify 5-HT receptors in periventricular brain regions (presuming the absence of DA receptors in these regions). The results indicate that, 1 h after intraventricular administration of 350 μM [^{3}H]-DLSD, a significant binding of LSD could be observed in the ependymal and subependymal zone of several periventricular brain regions. However, the label did not seem to be selectively bound to any particular structure. Experiments are now in progress to test the ability of DLSD and 2-bromo-LSD, but not LLSD, to displace the [^{3}H]-DLSD visualized by autoradiography and thereby identify specifically bound label and possibly the target sites for the 5-HT.

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5-HT and LSD high affinity binding sites to brain synaptosomal membranes

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5-HT and LSD binding have been studied, using purified synaptosomal membranes isolated from different regions of bovine brain by a density gradient centrifugation technique or in some experiments a lysed P_2 fraction isolated either from bovine or from rat brains.

Membranes were incubated at various temperatures (generally 22°C or for different purposes 0°C) in Tris-HCl buffer 50 mM pH 7.4 using tritiated ligands ([^{3}H]-5-HT 17 Ci/mM, [^{3}H]-LSD 22 Ci/mM). Separation of bound and free radioactivity was performed using an ultra-filtration technique (Whatman GFB glass fibre filter) under vacuum. The filter was rinsed with 10 ml of Tris-HCl buffer 0°C and the radioactivity trapped onto the filter was counted by liquid scintillation using Triton X 100 with a Toluene PPO-POPOP mixture.

Previously we described a saturable, reversible, high affinity binding for 5-HT, specific for tryptamines and related structures (Fillion, Fillion, Spinakis, Bahers & Jacob, 1976). Here, parallel studies for 5-HT and LSD bindings show that LSD binding corresponds to a saturable reversible high affinity site with a corresponding dissociation constant similar to that of 5-

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. * $P < 0.01$; † $P < 0.02$.