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BRIEF COMMUNICATION

A Comparison of the Locomotor Effects of 5-Hydroxytryptamine and 5-Hydroxytryptophan Administered via Two Systemic Routes[1]

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JACOBS, B. L. AND E. E. EUBANKS. *A comparison of the locomotor effects of 5-hydroxytryptamine and 5-hydroxytryptophan administered via two systemic routes.* PHARMAC. BIOCHEM. BEHAV. **2**(1) 137–139, 1974.–This study compared the effects of systemically administered 5-hydroxytryptamine (5-HT) and 5-hydroxytryptophan (5-HTP) on tilt cage locomotor activity in rats. 5-HT was a more potent inhibitor of activity than 5-HTP via both the s.c. and i.p. routes. The effect of 5-HT itself was greater when administered i.p., whereas the effects of 5-HTP were independent of the route of administration. These results indicate that behavioral changes following 5-HTP injection may be attributable to the peripheral effects of 5-HT.

5-hydroxytryptamine 5-hydroxytryptophan Rats Locomotor activity

5-HYDROXYTRYPTOPHAN (5-HTP), the immediate precursor of serotonin (5-hydroxytryptamine, 5-HT), has been used in a large number of neuropharmacological studies instead of 5-HT because of its greater penetrability of the blood-brain barrier. Since 5-HT and 5-HTP decarboxylase are widely localized in areas outside of the central nervous system [10], any behavioral or physiological effects observed following the systemic administration of 5-HTP might be attributable to peripheral effects of serotonin.

For this reason, we compared the effects of 5-HTP and 5-HT in a gross behavioral situation and using a quantifiable activity measure. Because 5-HT is so heavily concentrated in various parts of the gastrointestinal tract [10], we also decided to compare the effects of 5-HT injected via the typical intraperitoneal (i.p.) route with the effects following subcutaneous (s.c.) injection.

METHOD

Adult male Sprague-Dawley rats weighing approximately 400 g were used in these experiments. They were housed individually and maintained on an ad lib food and water schedule. Activity was measured by means of center-balanced wire mesh stabilimeter cages (17.5 cm × 20.0 cm × 37.5 cm) which tilted back and forth in the longitudinal direction as the rat crossed the fulcrum. Crossings were recorded by means of a microswitch at one end of the cage which was connected to an electronic counter in an adjacent room. The cages were housed in a well-ventilated temperature controlled room (22°C) with a 70 db masking noise and lighted by a 200 W incandescent bulb on a 12 hr L–D schedule with lights on at 2100 hr. Animals were habituated to the cages for one day prior to the beginning of all experiments. On the experimental day, hourly activity measures were begun at 1000 hr, 1 hr after lights off and were continued for 3 hr. The activity during this 3 hr period served as the baseline and as a basis for the semi-random assignment of animals to groups of approximately equal mean activity. At 1300 hr the counters were turned off and all injections were given between 1315 and 1330 hr. Following the i.p. injections, activity measures were begun again at 1400 hr and were continued on an hourly basis for the next 3 hr; however, due to the slower onset of action following s.c. injections, the 3 hr post-drug activity period was not begun until 1430 hr.

The DL form of 5-HTP was used, but doses are stated in terms of the L form in order to equate the doses of the non-stereoisomeric serotonin which was injected as the creatinine sulfate complex (doses are expressed as mg/kg of

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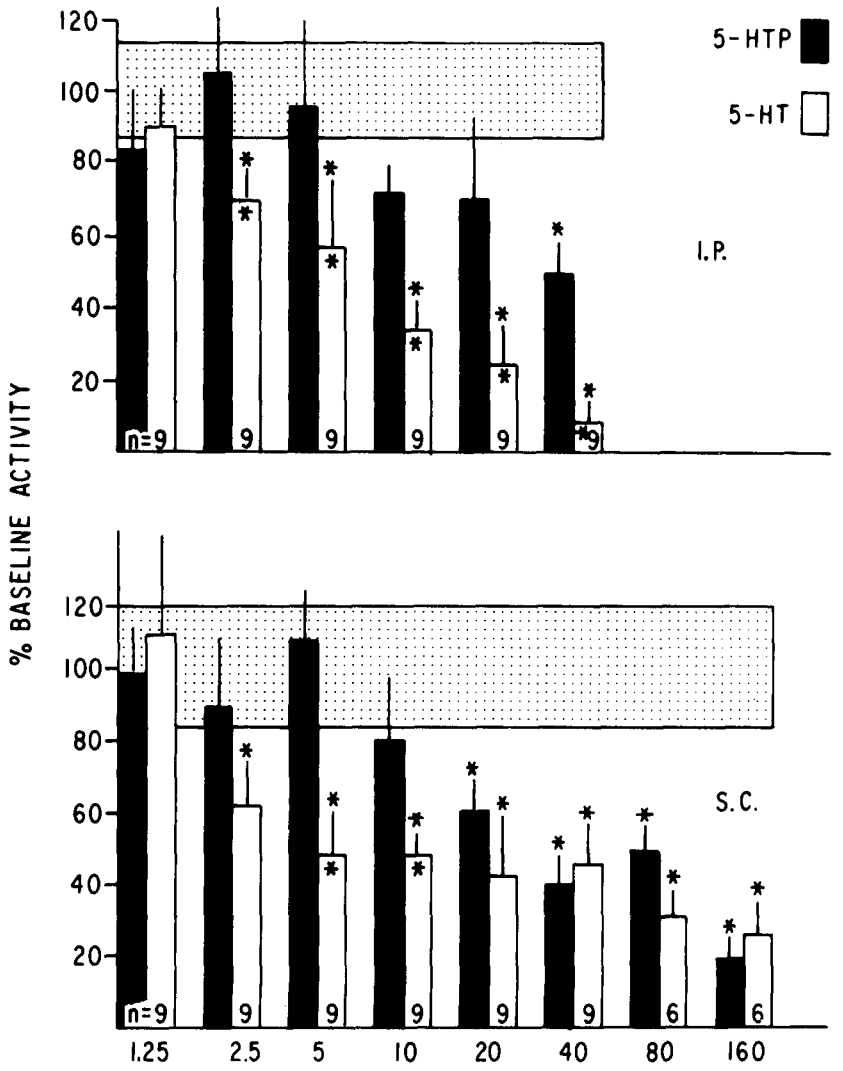

FIG. 1. Effect of serotonin or 5-HTP (i.p., top; s.c., bottom) on locomotor activity in the rat. Data are plotted as a % of the 3 hr baseline activity. The horizontal shaded bars at the top of each panel represents the mean activity ± S.E.M. for the vehicle injected control group (n = 36 & 48 for top and bottom respectively). The means for both were normalized to equal 100%. The vertical lines at the top of each bar represent S.E.M.'s. An asterisk above a bar indicates that the activity in this group was significantly different from control, while an asterisk within a serotonin bar indicates a significantly lower activity than the corresponding 5-HTP group ($p<0.05$, Newman-Keuls test). None of the 5-HTP groups were significantly less active than the corresponding serotonin group. Number of subejcts used at each dose is indicated by n = at the bottom of the bars. Doses refer to the L form of 5-HTP, although the DL form was injected, and the dose of serotonin is expressed as mg/kg of the base, although it was injected as serotonin creatinine sulfate.

the base). 5-HTP was dissolved in saline plus a few drops off 5N HCl, and the same vehicle was used for 5-HT. The volume of all injections was 1.6 ml. The s.c. injections were given on the loose skin of the back.

RESULTS

Several things are obvious from the data presented in Fig. 1. Serotonin was a significantly more potent inhibitor of locomotor activity than 5-HTP at most doses tested via the s.c. route ($F = 4.76$; 1,116 df; $p<0.05$) and especially via the i.p. route ($F = 16.83$; 1,96 df; $p<0.01$). 5-HTP never produced a significantly greater inhibition of locomotor activity than serotonin. The dose of serotonin required to inhibit locomotor behavior to a specific degree was approximately 1/8 the dose of 5-HTP required. Serotonin itself was more effective when given i.p., whereas the effect of 5-HTP was independent of the route of administration. Comparisons of serotonin and 5-HTP i.p. were discontinued at 40 mg/kg because this dose constituted the LD 50 for serotonin, whereas the LD 50 for serotonin s.c. was not

reached until doses of 160 mg/kg were given. None of the doses were lethal for 5-HTP i.p. or s.c.

Behaviorally, serotonin was much more potent than 5-HTP via both routes of administration. The ED 50 for producing ataxia with serotonin was 10 mg/kg i.p. and 40 mg/kg s.c. These doses also slowed the righting reflex. Maximum doses of 5-HTP never produced ataxia or affected the righting reflex, but did produce marked sedation.

DISCUSSION

These data indicate that peripherally administered serotonin has a potent effect on behavior even at low doses and that the effects of serotonin were greater than those of equivalent doses of 5-HTP. Furthermore, serotonin administered i.p. had a larger effect than when given s.c., whereas the effects of 5-HTP were independent of the route of administration. If these effects of serotonin are due to peripheral actions, then we must similarly assume that the effects of 5-HTP may be attributable to its peripheral actions. These results should therefore caution us against concluding that behavioral changes following administration of peripherally applied 5-HTP are due to its direct central action. There is also considerable evidence that 5-HTP may act indirectly by displacing brain catecholamines [6,9].

It is interesting to compare these results to previous data from our laboratory [12] which indicate that doses of L-tryptophan, which presumably elevate central and peripheral levels of serotonin [14], have no effect on locomotor activity. It has been hypothesized that L-tryptophan does not affect behavior because it only elevates intraneuronal serotonin which is then catabolized by monoamine oxidase [11,14]. We have no clear explanation of why our results with 5-HT, 5-HTP, and L-tryptophan differ from the results of Brown [4], who found L-tryptophan a more potent inhibitor of mouse locomotor activity than either 5-HT or 5-HTP.

There are other plausible explanations of the present results. Even though 5-HT less effectively crosses the blood-brain barrier, it may be more effective than 5-HTP in activating central serotonergic receptor sites. This is supported by evidence indicating that systemic injections of 5-HTP may produce large increases in brain serotonin content, but that much of it may be in non-serotonergic sites such as endothelial cells and catecholamine neurons [8, 9, 13]. This is also supported by reports that unlike iontophoretically applied 5-HT and systemically applied L-tryptophan, which inhibit the activity of raphe neurons [1,3], systemically administered 5-HTP had no effect on raphe neuronal activity [2].

Finally, if as some investigators have reported, 5-HT does cross the blood-brain barrier in significant quantities [5,7], this might account for the behavioral effects of 5-HT. We are currently testing this notion directly by administering 5-HT peripherally while recording the activity of raphe neurons. If 5-HT is acting on central serotonin receptors, we would expect it to inhibit raphe unit activity.

REFERENCES

1. Aghajanian, G. K. Influence of drugs on the firing of serotonin-containing neurons in brain. *Fedn. Proc.* **31:** 91–96, 1972.
2. Aghajanian, G. K., A. W. Graham and M. H. Sheard. Serotonin-containing neurons in brain: depression of firing by monoamine oxidase inhibitors. *Science* **169:** 1100–1102, 1970.
3. Aghajanian, G. K., H. J. Haigler and F. E. Bloom. Lysergic acid diethylamide and serotonin: direct actions on serotonin containing neurons. *Life Sci.* **11:** 615–622, 1972.
4. Brown, B. B. CNS drug action and interaction in mice. *Archs. int. Pharmacodyn.* **128:** 391–414, 1960.
5. Bulat, M. and Z. Supek. The penetration of 5-hydroxytryptamine through the blood-brain barrier. *J. Neurochem.* **14:** 265–271, 1967.
6. Butcher, L. L., J. Engel and K. Fuxe. Behavioral, biochemical and histochemical analyses of the central effects of monoamine precursors after peripheral decarboxylase inhibition. *Brain Res.* **41:** 387–411, 1972.
7. Costa, E. and M. H. Aprison. Distribution of intracarotidly injected serotonin in the brain. *Am. J. Physiol.* **192:** 95–100, 1958.
8. Dahlstrom, A. and K. Fuxe. Evidence for the existence of monoamine-containing neurons in the central nervous system. *Acta physiol. scand.* Suppl. 232, **62:** 1–55, 1964.
9. Fuxe, K., L. L. Butcher and J. Engel. DL-5-Hydroxytryptophan-induced changes in central monoamine neurons after peripheral decarboxylase inhibition. *J. Pharm. Pharmac.* **23:** 420–424, 1971.
10. Garattini, S. and L. Valzelli. *Serotonin.* New York: Elsevier, 1965.
11. Grahame-Smith, D. G. Studies *in vivo* on the relationship between brain tryptophan, brain 5-HT synthesis and hyperactivity in rats treated with a monoamine oxidase inhibitor and L-tryptophan. *J. Neurochem* **18:** 1053–1066, 1971.
12. Jacobs, B. L., E. E. Eubank, and W. D. Wise. Effect of indolealkylamine manipulations on locomotor activity in rats. *Neuropharmacology* (in press).
13. Lichtensteiger, W., U. Muntzner and H. Langemann. Uptake of 5-hydroxytryptamine and 5-hydroxytryptophan by neurons of the central nervous system normally containing catecholamines. *J. Neurochem.* **14:** 489-497, 1967.
14. Moir, A. T. B. and D. Eccleston. The effects of precursor loading in the cerebral metabolism of 5-hydroxyindoles. *J. Neurochem.* **15:** 1093–1108, 1968.