It is suggested that when drive level is defined in terms of hours of deprivation, the animals' prior history of maintenance schedules must be taken into account. Experiments on the effects of deprivation experiences occurring in infancy are now in progress.

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References and Notes

1. J. B. Matter, Ph.D. dissertation, Radcliffe College, Cambridge, Mass., 1956. The advice of Jerome S. Bruner is gratefully acknowledged.
2. A further replication of this experiment was attempted, with 3 days allowed between blocks of five trials instead of 2 days. Under these circumstances the animals were unable to learn the task.
3. For an abstract of these studies see J. Matter, J. S. Bruner, and D. D. O'Dowd, *Am. Psychol.* 9, 427 (1954); J. S. Bruner *et al.*, *Psychol. Monographs*, in press.
4. It can be seen from the standard deviations that the distributions, especially for the latency measure, tended to be skewed. Scores were transformed for purposes of statistical analysis.

1 July 1957

Release of 5-Hydroxytryptamine by Benzoquinolizine Derivatives with Sedative Action

Previous investigations have shown that reserpine causes release of 5-hydroxytryptamine (5-HT) from various body depots (brain, intestine, and blood platelets). After a single injection of a large dose of reserpine, the 5-HT content of these organs decreased to values between one-fifth and one-tenth of the normal levels and remained low for several days. Among the *Rauwolfia* alkaloids, only those with tranquilizing action showed this effect. A series of centrally acting drugs belonging to other chemical groups did not influence the 5-HT content of the brain (*1*).

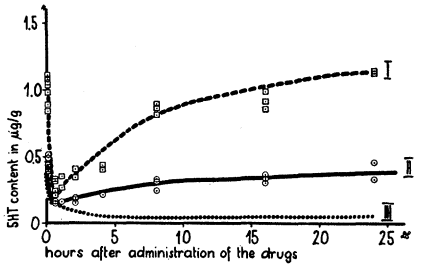
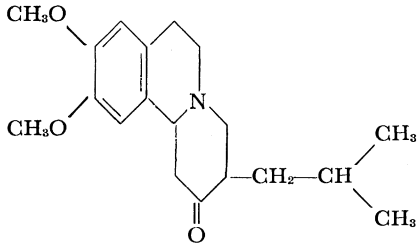


Fig. 1. Effect of compound I and reserpine on the 5-hydroxytryptamine (5-HT) content of brain. The drugs were given at zero time. Broken curve, intraperitoneal injection of 40 mg/kg of compound I to mice; each point represents the 5-HT concentration of five pooled brains. Solid curve, intravenous injection of 40 mg/kg of compound I to rabbits; each point represents the 5-HT concentration of one whole brain. Dotted curve, intravenous injection of 5 mg/kg of reserpine to rabbits (*3*).

It has now been found that, besides reserpine, various synthetic derivatives of 1,2,3,4,6,7-hexahydrobenzo[a]quinolizines (*2*) also release 5-HT. In mice and rabbits, these compounds produce sedation without hypnosis. Among the derivatives examined, compound I (2-oxo-3-isobutyl-9,10-dimethoxy-1,2,3,4,6,7-hexahydro-11bH-benzo[a]quinolizine)



I

showed the most marked sedative and 5-HT-releasing activity (Fig. 1).

After injection of 40 mg of compound I per kilogram, there was an immediate decrease of the brain 5-HT, measured fluorimetrically (*3*), the minimum value being reached within half an hour. As the dose was reduced, the 5-HT decline became gradually smaller, but was still evident with as little as 5 mg of compound I per kilogram. The absolute decrease in 5-HT per gram of tissue was greater in the brain stem than it was in the rest of the brain. During a 4-hour period after injection of 40 mg/kg in rabbits, the colorimetrically determined excretion of 5-hydroxyindoleacetic acid (*4*), a major metabolite of 5-HT, showed an average significant increase of 200 percent as compared with a similar control period before injection ($p < 0.01$). In rabbits, pretreatment with isopropyl isonicotinic acid hydrazide had the same influence on the effect of compound I as on that of reserpine (*5*): compound I no longer caused sedation, but excitation, mydriasis, and piloerection; the brain 5-HT showed only a very slight decline.

In addition to these similarities between the action of compound I and reserpine on the brain 5-HT, there were, however, some differences. (i) To reach maximum depression of 5-HT in the brain, mice required 4 times and rabbits 10 times as much compound I as was required of reserpine. In mice, the LD_{50} of compound I was about 10 times higher than the LD_{50} of reserpine. (ii) After administration of reserpine, the 5-HT in the brain decreased to a minimum of 10 percent, whereas after administration of compound I, the 5-HT concentration was never less than 25 to 35 percent of the original value. Even with doses exceeding 40 mg of compound I per kilogram, no greater decrease of 5-HT could be produced. (iii) Within 10 to 24 hours after injection of compound I, the 5-HT content of the brain had returned to normal values, whereas, after administration of reserpine, complete 5-HT recovery took several days. The sedative action of compound I lasted 4 to 8 hours, that of reserpine 1 to 3 days.

The benzo-quinolizine derivatives are thus a second group of substances which, like the centrally acting *Rauwolfia* alkaloids, cause both sedation and 5-HT depression in the brain. Closer investigations with these compounds may lead to further explanation of the role of 5-HT in brain function and in the central action of certain drugs.

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2. A report on the synthesis of the benzo-quinolizines is in preparation by A. Brossi and O. Schnider. The generic name for compound I is tetrabenazine.
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13 May 1957

Nature of the Glucuronide in Direct-Reacting Bilirubin

Evidence from several laboratories (*1–3*) has established that direct-reacting bilirubin is a diglucuronide. Billing, Cole, and Lathe (*1*) have suggested that bilirubin may be conjugated with glucuronic acid through its carboxyl groups, since the glucuronide is readily hydrolyzed by dilute alkali. Schmid (*3*) has assumed that the glucuronidic linkages occur with the α,α′-hydroxy groups of bilirubin.

Carboxyl (acyl) glucuronides can be differentiated from other glucuronides by the capacity of the former to react with hydroxylamine to yield hydroxamic acids and glucuronic acid (*4*). Treatment of bilirubin diglucuronide with hydroxylamine, therefore, provides a means of determining the nature of the glucuronidic bonds (*5*).

Urines obtained from three patients with obstructive jaundice were the source of the direct-reacting bilirubin (*3*). In each instance, treatment of an aliquot of the urine with hydroxylamine (*p*H 7, room temperature, 30 minutes) resulted in the formation of an appreciable quantity of hydroxamate. When the urine