over a rather large area and direct counting of mast cell number within 10 randomly chosen observation fields, but were unable to find evidence for any seasonal difference in the mast cell population. Equally high numbers of mast cells were apparently present in all preparations, and the granulation and staining properties of the cells were also similar.

Histamine was assayed in interfemoral membranes and lungs of 8 bats, 4 collected in October and 4 in April. A standard spectrofluorometrical assay technique[9] was applied, except that all samples had been dried in air (at about 30 °C) and were kept in the dry state for a few weeks before being subjected to assay. In accordance with the microscopic examination, a comparison of the histamine contents of the samples, calculated as μg of the base per g of fresh tissue (Table), revealed no significant difference between bats collected in October and bats collected in April.

Our observations are in good agreement with those of BALLOWITZ[10], who studied the effect of inanition on the mast cells in various tissues of bats of the species *Vesperugo noctula* and found that bats kept in the laboratory without food for the winter (5–6 months) maintained a mast cell population similar to that of bats killed immediately upon capture in the autumn. Another more recent report[4] directly compares the mast cell number of winter bats (*Myotis lucifugus*) obtained from a cave with that of summer bats of the same species obtained from a farmhouse and also lists data in agreement with ours. However, by stressing that the failure to find any difference in mast cell number in tissues from summer and winter bats might be accounted for by strain differences, the latter report unfortunately leaves the impression – which has subsequently been passed on by reviewers[6,11] – that during hibernation there is probably an increase in the number of tissue mast cells after all. From the results hitherto published it nevertheless seems that as far as bats are concerned, hibernation fails to induce any general changes in the mast cell population.

Histamine levels in tissues of the Indiana bat (*Myotis sodalis*) immediately prior to and at the end of the hibernating period

Time of year	Histamine content (μg/g) Interfemoral membrane	Lung
October	9.9–15.8	1.3–2.8
April	10.2–13.2	1.0–3.3

Zusammenfassung. Untersuchungen über die Zahl der Mastzellen und den Histamingehalt in Gewebe der Indiana-Fledermaus *Myotis sodalis* während des Winterschlafes zeigten keine Unterschiede zwischen den im Herbst, Winter und Frühling getöteten Tieren.

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Evidence for a Cholinergic Mechanism Inducing Histamine Increase in the Rat Brain in vivo

The function of cerebral histamine is not well established as yet[1,2]. Cerebral acetylcholine has been attributed a role in excitatory as well as inhibitory mechanisms[3–6]. Electrical[7] or chemical[8] stimulation of the central nervous system induces depletion of brain acetylcholine presumably through an accelerated release of this neurohumor[9–12]. It is shown here that during central nervous system stimulation, a cholinergic mechanism induces an increase in cerebral histamine.

Material and method. Albino rats of either sex, 150–200 g of body weight, were placed in individual wooden cages provided with a copper-wired grid bottom connected to an electronic stimulator. Electrical stimulation to the paws was applied over a period of 5 min, 4 c/sec, 20 msec duration, and a voltage high enough to cause a discrete jumping of the animal. At the end of the stimulation period the rats were sacrificed by decapitation, the brains were removed as soon as possible (less than 3 min) and the cerebral hemispheres and brain stem homogenized in ice-cold acid ethanol. After extraction, acetylcholine and histamine were separated by descending paper chromatography and the eluates assayed in the rat duodenum and guinea-pig ileum respectively. Antagonists were employed for further testing of specificity. Recoveries obtained were over 72% for amounts added to sample homogenates in the range of concentrations studied. Drugs used for treatments added to sample homogenates (3 μg/g for cholinesterase inhibitors and 3 mg/g of fresh tissue for L-histidine) did not interfere with the procedure.

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Changes in the levels of acetylcholine and histamine in the rat brain under various conditions

Treatment	Acetylcholine Cerebral hemispheres	Brain stem (μg/g of fresh tissue[a])	Histamine Cerebral hemispheres	Brain stem
Controls	2.88 ± 0.07	3.23 ± 0.07	1.77 ± 0.07	1.89 ± 0.07
Electrical stimulation[b]	1.71 ± 0.06 (−37)	2.05 ± 0.04 (−39)	2.18 ± 0.03 (+23)	2.40 ± 0.02 (+27)
Pysostigmine[b]	3.39 ± 0.03 (+18)	3.91 ± 0.03 (+21)	1.90 ± 0.07[c] (+7)	2.36 ± 0.03 (+29)
Electrical stimulation + Physostigmine	2.20 ± 0.05 (−24)	2.70 ± 0.07 (−17)	3.02 ± 0.04 (+70)	3.50 ± 0.06 (+85)
L-Histidine	2.81 ± 0.08[c] (−3)	3.12 ± 0.05[c] (−4)	2.05 ± 0.03[d] (+16)	2.58 ± 0.03 (+37)
Parathion	4.50 ± 0.08 (+56)	5.92 ± 0.08 (+52)	2.43 ± 0.04[e] (+32)	2.54 ± 0.05[e] (+35)
L-Histidine + Parathion	4.48 ± 0.16 (+55)	5.82 ± 0.18 (+51)	4.05 ± 0.03 (+128)	4.81 ± 0.10 (+153)

[a] Mean ± S.E. for 8 experiments. All values for treated groups are different from controls, $p < 0.001$, except where indicated. In brackets percent change of controls. [b] Different from electrical stimulation + Physostigmine, $p < 0.001$. [c] Not different from controls, $p > 0.05$. [d] Different from controls, $p < 0.01$. [e] Different from L-Histidine + Parathion, $p < 0.001$.

Further details will be published elsewhere as part of a multi-assay technique[13].

Results and discussion. Electrical stimulation induces a depletion of brain acetylcholine, which is accompanied by a concomitant increase of brain histamine in comparison with controls (Table). Acetylcholine depletion, presumably occurring through an accelerated release of the neurohumor from store sites[9–12], might bear a relationship with the increase of histamine. Treatment with cholinesterase inhibitors (physostigmine salicylate, 0.2 mg/kg i.p., 30 min before sacrifice, or an equimolar dose of parathion) induced a significant increase in the levels of both acetylcholine and histamine in the cerebral hemispheres and brain stem of the rat, exception made for histamine in the cerebral hemispheres when physostigmine was employed, where the change was not significant. The latter finding is possibly related to a slower rate of synthesis of histamine in the hemispheres as compared with the brain stem in the various experimental conditions. Free acetylcholine seems to be promoting an increase in the levels of histamine in the brain. Treatment with parathion showed more marked increments of both neurohumors, which is in agreement with its more potent cholinesterase inhibition[14].

Physostigmine plus electrical stimulation (the drug given 25 min before the stimulation) induced a considerably higher rise of histamine than the one found with either treatment alone. The levels of acetylcholine went below those found with physostigmine alone or in the control group, but remained higher than those found with electrical stimulation alone, as if the acetylcholine released during central nervous system stimulation, and accumulated because of cholinesterase inhibition, enhanced in some way to a larger extent the synthesis and/or the accumulation of histamine.

Administration of L-histidine (1000 mg/kg, i.p., 1 h before sacrifice) induced an increase in the histamine levels with no change in the acetylcholine levels. When parathion was administered half an hour after the administration of L-histidine, the combined treatment induced a similar increase in the acetylcholine content to that found with parathion alone; however, the increase in the histamine content was considerably higher than that found with either treatment alone, as if the increased concentration of free acetylcholine were enhancing the synthesis of histamine from L-histidine.

All these findings strongly suggest that the brain acetylcholine released during central nervous system stimulation is triggering a mechanism to accelerate the synthesis of brain histamine in vivo, since it is not likely that histamine might be mobilized from somewhere else in the body[15].

Résumé. Pendant l'excitation du système nerveux central chez le rat, un mécanisme cholinergique augmente l'histamine cérébrale.

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Behavioural Aspects of the Extraocular Light Sense of *Urodacus*, a Scorpion

An extraocular light sense has been demonstrated electrophysiologically in the Australian scorpion, *Urodacus*[1]. An indication was also given that the eyes are not necessarily involved in some forms of light dependent behaviour; the present paper continues this line of enquiry.

Urodacus is a nocturnal animal. During the day it withdraws under stones or in its burrow to emerge after sunset and remain active during the first few hours of the night. These two aspects of its behaviour, negative phototaxis and nocturnal activity, were chosen for an assessment of the relative importance of eyes and extraocular light sense.

Withdrawal. Animals in half-darkened Petri dishes withdraw from the exposed to the darkened half (Table).