ments, the obesity index was greatly elevated throughout the experiment.

2. Female rats. Table II indicates that ether anesthesia did not significantly alter the colonic temperature in female VMNL rats. Similar to the male VMNL rat, however, were the normophagia and the greatly elevated obesity index.

The date indicate an age difference in the hyperthermic response in female VMNL rats and a sex difference in weanling VMNL animals.

The age difference is probably related to the interference by the lesion at an early age with the 'setting' of the central temperature control mechanism(s); weanling VMNL rats would then be unable to respond with hyperthermia as do mature female VMNL rats[5]. It might also be related to the lack of hyperphagia in the weanling VMNL rat, since MAYER and GREENBERG[5] were unable to find hyperthermia in normophagic mature female VMNL rats. Weanling VMNL animals, however, are consistently normophagic[7-10,16].

The sex difference in the hyperthermic response in the weanling rat could possibly be related to a different sensitivity of the central regulatory mechanism(s) to the lesioning procedure. This appears in keeping with the report by THOMPSON and STEVENSON[17] that the exercized mature female rat can tolerate a higher temperature before commencing vasodilatation and is able to regulate core temperature at a higher level than the mature male rat as exersize continues. The data show finally that the hyperthermic response (male VMNL rats) or the lack thereof (female VMNL rats) is not related to food intake and the obesity status of the animals[5].

Zusammenfassung. Entwöhnte junge weibliche Ratten mit ventromedialen hypothalamischen Läsionen zeigten keine Hyperthermie, wie sie bei geschlechtsreifen weiblichen Ratten bei Äthernarkose beobachtet wird. In männlichen entwöhnten, jungen Ratten mit Läsionen war jedoch eine signifikante Hyperthermie nachweisbar, welche in beiden Geschlechtern von der Futtermenge und dem Ausmass der hypothalamischen Fettsucht unabhängig war. Läsionen im ventromedialen Hypothalamus der weiblichen jungen Ratte scheinen einen zentralen Mechanismus zu beeinflussen, welcher es ihnen nicht ermöglicht, auf Äthernarkose mit Hyperthermie zu reagieren.

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Calcium Evoked Release of 5-Hydroxytryptamine from the Brain of the Unanesthetized Cat[1]

The ionic milieu of neurones in the nervous system is an important determinant of transmitter activity at synapses[2]. It is known, for instance, that Ca ions can affect the release from nerve tissue of certain substances such as acetylcholine[3], noradrenaline[4] or γ-amino-butyric acid[5]. However, little is known about the role of Ca ions in the release of 5-hydroxytryptamine (5-HT) particularly within the intact brain. We now present evidence that the level of Ca ions in extracellular fluid can influence markedly the release of 5-HT from sites scattered throughout the hypothalamus of the unanesthetized cat, and that the neural activity of this amine could depend upon the levels of extracellular Ca within the brain-stem.

Materials and method. In cats of either sex, modified push-pull cannulae[6] were implanted into the rostral, caudal and lateral areas of the hypothalamus according to methods described earlier[7]. 5–7 days later, a 1 mm sphere of tissue at the tips of the cannulae was perfused with Krebs-Henseleit solution prepared with glass-distilled, ion-exchange water. Pyrogen-free glassware, syringes and tubing were used in all experiments, and the rate of perfusion was 50 µl per min during a 30 min interval. Samples were accepted for assay only if the volume of the effluent matched that of the inflow, and if the perfusate was clear and devoid of tissue fragments. During the course of a perfusion, the animal was held gently and showed no untoward signs of disturbance or discomfort. The effluents were collected on ice and if not tested on the same day, were kept at $-10\,^{\circ}C$ until assayed. At the conclusion of each series of experiments, the perfusion site was verified following standard histological procedures.

The content of 5-HT in each 30 min sample was determined by the sensitive assay method of VANE[8], the isolated rat stomach fundus strip. The contractile activity of a perfusate was considered to be due to 5-HT only if: a) the contraction produced by a sample was of a similar shape and magnitude to that caused by 5-HT; and b) it was abolished by either methysergide or brom-lysergic acid added to the bath in doses of 10 to 20 µg. Values for 5-HT were calculated in terms of the creatinine phosphate salt.

Results and discussion. At 19 sites in the hypothalamus extending from the anterior region, between the anterior commissue and optic chiasm, to the posterior area dorsal to the corpora mammillaria, the resting release of 5-HT per 30 min interval varied in the control perfusions from 0.1 to 2.0 ng. However, when Ca was present in the Krebs-Henseleit perfusate in 2.6 or 10.4 mM in excess of the normal mM value of extracellular fluid, the level of 5-HT in the perfusate varied between 0.5 and 3.6 ng per 30 min interval. The average resting output was 0.56 ± 0.11 ng per 30 min in those 19 sites at which 5-HT was released.

Three distinct types of loci could be identified. The Table presents the mean values of 5-HT released from: 1. those sites at which Ca evoked a significant increase in the liberation of 5-HT from the hypothalamus; 2. those at which little change in 5-HT output was observed; and 3. those sites at which the release of 5-HT declined. Of

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special importance is the finding that 2.6 mM excess Ca produced an increase in the output of 5-HT to a level of 1.03 ± 0.29 ng per 30 min interval, whereas 10.4 mM excess Ca caused an even greater release of this amine to a level of 1.65 ± 0.35 ng per 30 min. Within those sites at which Ca evoked an increase in the release of 5-HT, physiological and behavioral changes in some instances accompanied the elevated activity of 5-HT. For instance, the cats often became drowsy, occasionally slept, or their body temperature declined and vasodilatation occurred.

In contrast to the results of experiments with brain slices[9], our results obtained with unanesthetized cats show that a direct relationship exists between the level of extracellular ions and the release of 5-HT in the central nervous system. This corresponds to the relationship found between the concentration of Ca and the release of noradrenaline and acetylcholine in the peripheral nervous system. Moreover, the output of 5-HT in the hypothalamus is dependent upon Ca in much the same way as the release of acetylcholine is dependent upon this ion within the cerebral cortex[10]. Taken together, these results suggest that an ionic mechanism is required for the synaptic release of 5-HT in the brain-stem and they support further the possible transmitter role for this biogenic amine.

Release of 5-HT expressed in ng per 30 min interval

Control (19 sites)	Excess Ca (2.6 mM to 10.4 mM)		
	Increased 5-HT Release (12 sites)	Unchanged 5-HT Release (4 sites)	Decreased 5-HT Release (3 sites)
0.56 ± 0.11	1.39 ± 0.25[a]	0.75 ± 0.23	0.30 ± 0.09[b]

Values are given as averages $\pm$ the standard error for the sites at which 5-HT output increased above the control level, remained unchanged, or decreased during the perfusion with a Krebs-Henseleit solution containing excess Ca. [a] Significantly different from control at 0.01 level. [b] Significantly different from control at 0.05 level.

Zusammenfassung. Kalzium-Ionen wurden mittels lokaler Perfusion im Hypothalamus von Katzen vermehrt. Der 5-HT-Gehalt der Durchströmungsflüssigkeit nahm im Verhältnis zur Konzentration der Kalzium-Ionen zu.

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Totale Plasmaclearance von Na[^{51}Cr-ÄDTA] und Inulin-^{14}C bei Ratten

Die Bestimmung der glomerulären Filtration mittels des sogenannten «steady state-Clearance»-Verfahrens stösst bei Ratten auf technische Schwierigkeiten. In vorliegender Arbeit wird geprüft, ob die sogenannte «slope»-Methode und das robuste Cr-Chelat des Äthylendiamintetraacetats (Na[^{51}Cr-ÄDTA]), das bei Menschen zur Messung der glomerulären Filtration benutzt wird[1–5], hierfür in Frage kommen. Zum Vergleich wurde Inulin herangezogen, das nur glomerulär ausgeschieden und dessen totale Plasmaclearance mit seiner renalen Clearance gleichgesetzt wird.

Weibliche Albinoratten mit einem Körpergewicht von 185–200 g wurden i.v. mit Inulin-^{14}C-Carboxylsäure (1,1 mg, 3 µCi) bzw. ^{51}Cr-ÄDTA (5–15 µg, 3–10 µCi) injiziert und zu verschiedenen Zeitpunkten seziert. Den ^{51}Cr-Gehalt des Plasmas, der Organe und des Gesamtkörpers bestimmten wir mittels eines γ-Spektrometers, die ^{14}C-Aktivität des Plasmas im Flüssigkeitsszintillator[6]. Aus der Figur ist der Verlauf der Plasmakonzentration und Ganzkörperaktivität der Substanzen zu ersehen. Die Daten, die zur Berechnung der totalen Plasmaclearance führten, sind in der Tabelle angegeben.

Die Zeitabhängigkeit der Plasmakonzentration von ^{14}C und ^{51}Cr lässt sich durch die Summe von zwei exponentiellen Terms wiedergeben. Da der zweite Term nur ca. 0,1% der schnell ausgeschiedenen Fraktion ausmacht, kann er vernachlässigt und der Verdünnungsraum sowie die totale Plasmaclearance aus den Werten für ^{51}Cr-ÄDTA bis zu 120 min und für Inulin-^{14}C bis zu 90 min berechnet werden (Tabelle). Der geringfügige Unterschied zwischen der biologischen Halbwertszeit ($T_{1/2}$) von Inulin-^{14}C und ^{51}Cr-ÄDTA hat eine Signifikanzwahrscheinlichkeit von 0,03; Verdünnungsraum und totale Plasmaclearance dagegen sind nicht signifikant verschieden. Was die Ausscheidung von ^{51}Cr-ÄDTA aus dem gesamten Körper betrifft, so ist die $T_{1/2}$ der ersten Exponentiellen mit der Plasmaclearance praktisch identisch (Figur). Nach 24 h ist der ^{51}Cr-Gehalt der Organe gering; er beträgt in Leber und Niere 0,35 bzw. 0,15% der ^{51}Cr-Dosis. Der an die Blutzellen gebundene ^{51}Cr-Anteil ist in den ersten Stunden zu vernachlässigen, beträgt nach 24 h jedoch rund 50% der Gesamtblutaktivität. Die Frage, was dem langsamen Term der Plasmaclearance ursächlich zugrunde liegt, lassen wir offen. Radiochemische Verunreinigungen und – im Falle von ÄDTA – nicht cheliertes ^{51}Cr könnten dabei eine Rolle spielen.

Da die totale Plasmaclearance von ^{51}Cr-ÄDTA mit der des Inulins übereinstimmt und da weiterhin unsere mit der «slope»-Methode erhaltenen Werte für die totale Plasmaclearance mit den unter «steady state» Bedingungen gewonnenen Daten (0,7–1,1 ml $\times$ min^{-1} $\times$ 100 g^{-1}) über-

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