Studies are in progress in our laboratory to elucidate this problem[10].

Résumé. L'évolution morphologique des cellules trophoblastiques humaines, ainsi que leur potentiel de sécrétion hormonale, in vitro, ont été examinés pendant plus de 2 mois, à l'aide d'une technique simplifiée de culture de placenta. La signification des cellules dites «géantes», l'aspect fibroblastique des trophoblastes dans les cultures agées, et le développement des vrais fibroblastes dans ces cultures, sont discutés.

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Effect of N^6O^2-Dibutyryl Cyclic 3', 5'-Adenosine Monophosphate on the Pinocytosis of Brain Capillaries of Mice

Pinocytotic activity in brain capillaries, except for those capillaries of brain areas not protected by the 'blood-brain' barrier, is known to operate at very low level under normal conditions[1]. The failure of penetration of certain substances from the blood circulation into brain substance has already been connected with the low rate of pinocytosis revealed in the endothelial cytoplasms of brain capillaries[2]. In contrast, in almost every study of pathological damage of the 'blood-brain' barrier, during which the capillary macromolecular transfer is usually enhanced, a considerable increase in the number of pinocytotic[3] and coated vesicles[4] has recently been reported.

The present investigation was designed to elucidate whether the pinocytosis of capillaries in the 'blood-brain' barrier-protected brain areas could be influenced experimentally by the lipid soluble substituted derivative of cyclic adenosine monophosphate (cAMP).

Mice, weighing about 20–25 g, were each given, by i.p. injection, a single dose (10 mg/kg) of N^6O^2-dibutyryl cyclic 3', 5'-adenosine monophosphate (dibutyryl cAMP, Sigma) dissolved in 0.1 ml saline. Control mice were given 0.1 ml saline solution only. The animals were killed by decapitation 5 or 20 min after injection. Small cubes of the parietal cortex and the cerebellar vermis were fixed in Karnovsky's aldehyde fixative and postfixed in Millonig's buffered osmic acid. Thin sections were examined in a Jeol 100B electron microscope. Plates of the non-nuclear areas of the endothelial cells to be measured were taken from randomly selected capillaries at a magnification of × 30,000. Portions of the endothelial cytoplasm were outlined and planimetrically measured on prints of final magnification × 90,000. The pinocytotic and coated vesicles, both attached to the luminal and abluminal surfaces and lying free in the cytoplasm, were counted for 1 μm^2 of endothelial cytoplasm on prints derived from the experimental and control groups. The counts of vesicles per unit area of endothelial cytoplasm in the experimental groups were compared statistically with those of the controls using the Student's *t*-test.

Evaluation of the results obtained is summarized in the Table. There was a significant increase in the numbers of pinocytotic vesicles lying free and attached to the basement membrane 5 and 20 min after the treatment and also in the counts of coated vesicles 5 min but not 20 min after the dibutyryl cAMP administration. A possible rate of induced pinocytosis, judged by comparing the total counts of vesicles obtained for brain capillaries after dibutyryl-cAMP treatment with those previously reported by BRUNS and PALADE[5] and CASLEY-SMITH[6] for qlood capillaries of diaphragm and endothelium of lymphatics, respectively, can be estimated. It appears that

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Effects of dibutyryl adenosine 3',5'-cyclic monophosphate on the counts of vesicles involved in the pinocytosis of brain capillaries (mean ± S.E.M.)

	pvp	pvi	pva	cp	ci	total count of vesicles
Control	0.03 ± 0.04	7.09 ± 0.10	0.41 ± 0.02	0.05 ± 0.06	0.55 ± 0.21	7·89 ± 0.11
5 min	0.41 ± 0.25	14.84 ± 0.18	4.88 ± 0.1	0.32 ± 0.01	1.51 ± 0.3	22.14 ± 0.24
P value	<0.02	<0.001	<0.001	<0.05	<0.001	<0.001
20 min	0	15.37 ± 0.48	4.15 ± 0.16	0.33 ± 0.03	1.02 ± 0.52	21.15 ± 0.52
P value	–	<0.001	<0.001	<0.05	<0.20	<0.001

Legend: pvp, pinocytotic vesicles pinching off from the luminal membrane; pvi, pinocytotic vesicles inside the endothelial cytoplasm; pva, pinocytotic vesicles attached to the basement membrane; cp, coated vesicles pinching off from the luminal membrane; ci, coated vesicles inside the endothelial cytoplasm; 0, evaluation was not made. *n* in control = 54; *n* in 5 min group = 53; *n* in 20 min group = 25.

the evoked pinocytosis operates with approximately one quarter the efficiency of that existing normally in capillaries having intense macromolecular transport.

Of the microvesicles evaluated, the pinocytotic vesicles are in general presumed to be of non-selective transporting elements[7], whereas the coated vesicles are thought to absorb selectively and carry proteins[8] into the blood capillaries. Recent studies on the permeability of toad urinary bladder to water and certain other small molecules, in relation to the pinocytosis of mucosal cells, have clearly indicated that the well-known permeability-modifying effect of neurohypophyseal hormones is firmly associated with cAMP mediated induction of pinocytosis[9].

Increase in pinocytosis of brain capillaries has long been observed in several cases of pathological damage to the 'blood-brain' barrier[10,11]. No hypothesis has, however, as yet been put forward to explain the altered cellular activity resulting from any pathological circumstance. From our results, the fact that cAMP plays a key role in the regulation of pinocytosis in the brain capillaries can be readily established. For this reason, it might be assumed that an increase in level of cAMP may also occur during the evoked breakdown of the 'blood-brain' barrier, regardless whether it is of experimental or pathological origin.

Whether the described facilitation of pinocytosis, by means of which certain macromolecules can be readily transported through the endothelium, is effected directly by cAMP generated in the tissue or is a secondary effect, still remains to be elucidated[12].

Zusammenfassung. Die Anzahl der pinocytotischen und stachligen Vesicula wurde unter normalen Bedingungen mit Bezug auf einheitliche Gebiete der pinocytotischen Aktivität der Kapillaren von Mäusegehirnen quantitativ charakterisiert. Die Pinocytose der Gehirnkapillaren war durch das substitutielle cyclische N^6O^2-Dibutyrylderivat des 3′, 5′-Adenosinmonophosphats signifikant gesteigert.

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Effect of Serum from Depressed and Manic Patients on Maze Behavior of Rats

In 1961 Poliakova[1] reported that serum from patients with manic-depressive illness had differential effects on the behavior of dogs in a maze. Dogs injected intravenously with serum from depressed patients ran the maze more slowly than controls. Those injected with serum from manic patients ran slightly, but consistently, faster and made more errors. This study has been cited in reviews of affective illness[2,3], but no further work in this area has been reported. The present study was designed to further investigate the effect of serum from manic-depressive patients on animal behavior.

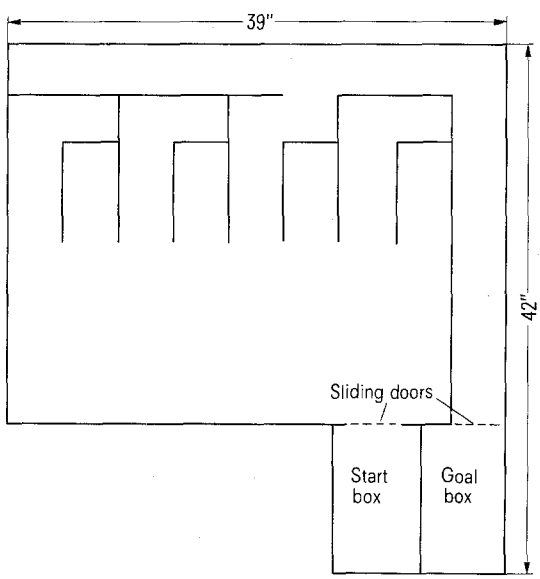

Fig. 1.

Methods. Male Sprague-Dawley rats were trained to run the maze illustrated in Figure 1 (adopted from Poliakova). This maze requires a choice of 9 alleys and a left-right discrimination for reward of moist food. Animals were food deprived daily from 07.00 h. Trials were conducted between 14.00 h and 15.00 h following which food was given ad libitum. Time from leaving start box to reaching goal box and number of errors were recorded for each trial. All rats were trained to a criterion of 5 consecutive trials, each under 10 sec with less than 2 errors. On experimental days, rats were run immediately before and at 0, 10, 20 and 30 min after tail-vein injection of 0.4 ml[4] of serum from manic and depressed patients or normal controls. Serum was injected within 1 h of being drawn and the investigator running the animals was blind to its source.

Seven depressed patients donated serum a total of 14 times for trials in 20 different rats. Six manic patients' serum was used 12 times in 12 different rats. All patients were drug free for at least 1 week prior to the experiment. Control serum from 5 euthymic investigators was used in 24 trials.

Patients were hospitalized on 2 psychiatric research units at the NIMH designed for intensive longitudinal study of manic-depressive illness. All patients were diagnosed by 3 staff psychiatrists and a social worker, and patients with other than primary affective illness were excluded from the study. Depression and mania were

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[4] This amount of serum exceeds by 50% that used by Poliakova in dogs (10 ml) on a ml/kg basis.