Blood samples were taken immediately before surgery and afterward at various times including an immediate post mortem sample.

The observations are summarized in the Table. First stage preparatory surgery was a strong stimulus to CxRP appearance in blood. All animals responded, 7 of 16 reaching a maximum response in 24 h and 9 in 48 h. Eleven had titers $\geqq 1:8$ at 24 h. Four rabbits tested repeatedly during the first 24 h had reached half their maximum response in 14 h, in essential agreement with the temporal sequence of response noted by others[1,3]. Over $^2/_3$ of the animals had remained CxRP positive until the time of total surgical removal of the liver, generally 4–6 weeks after the first stage. The extended response was somewhat unexpected and may be related to continuing vascularization in the abdominal area or to continuing inflammatory processes around the initial midline incision. The moderately high mean titer was caused by 2 rabbits with high titers, 7 of the 11 having titers of 1:4 or less. Titers of 10 of the 11 positive at surgery declined while 1 remained the same (1:16). The extent of the decline in titer was quite variable: titers of 2 rabbits surviving nearly 24 h declined from 1:4 to negative and 1:4 to 1:2 while 2 surviving 9 and 11 h dropped from 1:32 to 1:4 and 1:8 to 1:1 respectively. 4 rabbits were negative at the time of the second stage operation. All of these remained negative for the 9, 12, 18 and 35 h they survived. The mean survival time for all hepatectomized rabbits reported in the Table was 15 h. No relationship between survival time and serum CxRP status was observed.

These results support the view of HURLIMANN et al.[5] that CxRP is produced uniquely by the liver. They show, however, that CxRP can be eliminated from the blood stream without mediation of the liver and that the protein at times can disappear from the blood quite rapidly. It is conceivable, therefore, that a source other than the liver exists but with synthetic capabilities too small to keep up with elimination of the protein from the circulation. We feel this is improbable and what is seen in serum at any particular time in the intact animal represents a balance between the rate of synthesis by the liver and the rate of elimination from the serum, one mechanism of which appears to be binding to damaged tissues[2,3,9].

Zusammenfassung. Synthese und Stoffwechselfähigkeit von Cx-reaktiven Proteinen in hepatektomierten Kaninchen wurden untersucht. Diese Kaninchen konnten das Protein nicht bilden, sie behielten jedoch die Fähigkeit, es aus ihrem Blutkreislauf auszuscheiden.

R. F. RILEY, M. K. COLEMAN and H. D. SNOW

Department of Radiology, UCLA Health Science Center, Los Angeles (California 90024, USA),
16 September 1968.

[9] This investigation was supported by a grant from the U.S. Public Health Service.

Brain Uptake of ^{3}H Noradrenaline in Normal and *Shigella dysenteriae* Exotoxin Treated Mice

Experimental evidence indicates that noradrenaline does not pass in a significant quantity into brain capillaries[1]. On the other hand, molecules of comparable size evidently cross the capillary wall quite easily, rapidly equilibrating with a space of about 1–2 ml/100 g of brain tissue[2–4].

In the present study, an attempt has been made to investigate the transcapillary passage of noradrenaline in the brain of normal and *Shigella dysenteriae* toxin treated mice. *Shigella dysenteriae* toxin has been shown to damage the capillary wall in the mouse brain[5].

We have measured simultaneously the space of ^{3}H noradrenaline, ^{14}C sucrose and ^{131}I albumin. ^{14}C sucrose was found to pass very rapidly out of brain capillaries and its penetration into the brain tissue is probably hindered by some of the pericapillary structures[4]. ^{131}I albumin was used as an indicator of approximate intravascular plasma volume.

Material and methods. DL noradrenaline-7-H^3-hydrochloride, specific activity 1,820 mc/m*M*, sucrose C^{14} (U) specific activity 10 mc/m*M*, ^{131}I-human serum albumin were used.

The experiments were carried out in female mice, weight range 20–22 g. To 1 group of the animals, 36 h before the administration of labelled compounds, 4 LD_{50} of *Shigella dysenteriae* exotoxin was injected i.v. The other group of mice received saline. To both groups of animals, 3 μc of ^{131}I-albumin and 25 min later, 3 μc of ^{3}H noradrenaline and 0.5 μc of ^{14}C sucrose were injected i.v. 30 min after the first injection the animals were killed and blood and brain tissue were handled as reported previously[4]. ^{3}H noradrenaline was isolated by absorbtion on aluminium oxide.

The γ-radioactivity was measured in a well scintillation counter. β-radioactivity of ^{3}H and ^{14}C was counted in a Packard liquid scintillation spectrometer, using the screening method.

The results are expressed in terms of the space of a substance in the brain tissue defined in the case of ^{131}I albumin as: (cpm per 1 g of brain tissue)/(cpm per 1 ml of plasma) × 100 in the case of ^{3}H noradrenaline and ^{14}C sucrose as: (dpm per 1 g of brain tissue)/(dpm per 1 ml of plasma water) × 100; water content of plasma was assumed to be 93%.

Results. The space of ^{3}H noradrenaline, ^{14}C sucrose and ^{131}I albumin in the brain of animals intoxicated with *Shigella dysenteriae* toxin is significantly increased. The values of the space of ^{3}H noradrenaline and ^{14}C sucrose are approximately equal in both, the normal and the toxin treated group (Table).

[1] H. WEIL-MALHERBE, L. G. WHITBY and J. AXELROD, J. Neurochem. *8*, 55 (1961).
[2] L. Z. BITO, M. W. B. BRADBURY and H. DAVSON, J. Physiol. *185*, 323 (1966).
[3] J. ŠTULC, Life Sci. *6*, 1837 (1967a).
[4] J. ŠTULC, Am. J. Physiol. *213*, 1053 (1967b).
[5] J. B. CAVANAGH, J. G. HOWARD and J. L. WHITBY, Br. J. exp. Path. *37*, 272 (1956).

Discussion. As already pointed out, if the entry of ^{3}H noradrenaline into the brain tissue is prevented by a capillary wall, the space of ^{3}H noradrenaline in the brain should be approximately equal to the space of ^{131}I albumin. In the intoxicated animals, the space of ^{3}H noradrenaline and ^{131}I albumin would probably change in a parallel way.

On the other hand, if the passage of noradrenaline into the brain is hindered by pericapillary structures, the space of ^{3}H noradrenaline in normal and probably also in the intoxicated animals would be close to the values of ^{14}C sucrose.

Our present experiments seem to speak for the latter possibility, and therefore we suspect that the barrier preventing the entry of noradrenaline from blood into the brain is probably located outside the capillary wall.

Our hypothesis does not correlate well with the observations of BERTLER et al.[6] Using fluorescence microscopy, these authors found that the vascular endothelium in the brain constitutes a barrier for some monoamines closely related to noradrenaline.

Our approach to the investigated problem is based on the hypothesis of the existence of a rapidly equilibrating pericapillary space in the brain[2-4]. However, the existence of the pericapillary space in the brain tissue has not been confirmed by a direct histological method[7].

It seems that the question of the existence of the pericapillary space in the brain and its relation to the rapidly exchanging compartment has to be elucidated before a final evaluation of the present experiment.

	Space ml/100 g of tissue		
	^{3}H noradrenaline	^{14}C sucrose	^{131}I albumin
Controls	2.20 (1.49–2.91)	2.17 (1.92–2.42)	1.21 (0.70–1.72)
Intoxicated animals	8.10 (5.27–10.93)	8.37 (6.31–10.43)	2.57 (2.07–3.07)

The values represent the mean value from 10 animals with their fiducial limits.

Zusammenfassung. Die Autoren untersuchen den transkapillaren Durchtritt von ^{3}H Noradrenalin in das Gehirn der Mäuse. Auf Grund der Versuche scheint es wahrscheinlich, dass die Barriere, die den Eintritt von Noradrenalin in das Gehirn verhindert, sich nicht in der Kapillarwand befindet.

J. ŠTULC, K. MAŠEK and R. FRIEDRICH

Department of Pharmacology, Faculty of Pediatrics, Charles University, and Institute of Pharmacology, Czechoslovak Academy of Sciences, Praha (Czechoslovakia), 17 October 1968.

[6] A. BERTLER, B. FALCK and E. ROSENGREN, Acta pharmac. *20*, 317 (1963).
[7] S. W. KUFFLER and J. G. NICHOLLS, Ergebn. Physiol. *57*, 1 (1966)

Effects of Alcohol Ingestion on the Intercalated Disc in the Mouse Heart

Heart muscle is divided into distinct cellular units by the intercalated disc at the intercellular boundaries[1]. Physiological studies indicate that the electrotonic coupling between the adjacent heart muscle cells is facilitated by the low resistance of the intercalated disc[2-4]. Recent studies of SOHAL et al.[5] and KAWAMURA and KONISHI[6] suggest that the intercalated disc undergoes morphologic alterations in certain physiologic and pathologic conditions. There is some evidence of a direct causal relationship between alcohol consumption and heart disease in man[7]. This study reports the ultrastructural changes of the intercalated disc in the myocardium of mice following prolonged ethanol ingestion.

Hearts of 20 HaM/ICR mice (3 weeks old) that drank water containing 15% ethanol by volume for a period of 3 months were studied. The mice were allowed access to ethanol-free water for a period of 10 min on each alternate day. They were fed on balanced commercial Purina Chow ad libitum. Ten animals from the same stock were kept as controls and given no ethanol. At the end of 3 months the mice were killed by spinal dislocation. Daily alcohol consumption at the time of sacrifice was approximately 5.2 ml/100 g body weight.

Small pieces of left ventricular myocardium were fixed in 3% phosphate buffered glutaraldehyde for 2 h followed by 1% osmium tetroxide for 75 min. Tissues were embedded in Maraglas. Thin sections were cut with an LKB 'Ultrotome' microtome and stained with uranyl acetate and lead citrate. A Siemens 'Elmiskop I' electron microscope was used for observations.

The fine structure of the intercalated disc in control mice was similar to that described by FAWCETT[1]. The disc is structurally differentiated into 4 types of regions without any particular sequential order of location. These regions are designated as (1) macula adherens or desmosome, (2) macula occludens or nexus, (3) fascia adherens or area of myofibrillar insertion and (4) nonspecialized region or intercellular gap region (Figure 1).

In the alcoholic mice the intercellular space in the fascia adherens region was strikingly widened (Figure 2). In the control animals the maximal width of the intercellular space in this region was approximately 250 Å, whereas in the experimental mice the maximal width increased to about 0.3 μ. No significant alterations were observed in other regions of the intercalated disc. Widening of the intercellular space does not seem to be a result of preparatory procedures as the control tissues

[1] D. W. FAWCETT, *An Atlas of Fine Structure. The Cell* (W. B. Saunders Co., Philadelphia 1966).
[2] J. W. WOODBURY, *Handbook of Physiology, Circulation* (American Physiological Society, Baltimore 1962), vol. 1, sec. 2, p. 237.
[3] L. BARR, M. M. DEWEY and W. BERGER, J. gen. Physiol. *48*, 797 (1965).
[4] S. WEIDMANN, J. Physiol. *187*, 323 (1966).
[5] R. S. SOHAL, S. C. SUN, H. L. COLCOLOUGH and G. E. BURCH, Lab. Invest. *18*, 49 (1968).
[6] K. KAWAMURA and T. KONISHI, Jap. Circul. J. *31*, 1533 (1967).
[7] C. S. ALEXANDER, Ann. intern. Med. *67*, 670 (1967).