can be obtained through measurements performed in arterial blood.

Our results are very different, and not comparable, with those obtained by other investigators who did not use an adequate mixing time of the indicator[1]. They also differ from those obtained by D'AGOSTINO[2], EVERETT et al.[3] and BOCCI et al.[4] in the rat, who employed a single sample after mixing was completed.

Regional plasma volume was measured in mice by FRIEDMAN[7], KALISS and PRESSMAN[8] and WISH et al.[9], and regional hematocrit calculated on the assumption that there is no difference between arterial blood hematocrit and organs' blood hematocrit. As we have shown in our results their assumption can not be sustained and the regional hematocrit ratio expressed in their results can not be accepted as accurate.

GIBSON et al.[10] measured regional red cell and plasma volumes and hematocrit ratio in dogs; their results are higher than ours in rats, this being due probably to their technique of single sampling of blood and tissues, besides the difference in species.

Table III. Hematocrit ratio, blood volume and interstitial fluid volume in tissues (mean ± S.D.)

	Hematocrit ratio	Blood volume ml/100 g	Interstitial fluid volume ml/100 g
Whole body	42.08 ± 2.55	7.44 ± 0.55	20.68 ± 0.97
Testis	14.63 ± 1.22	2.41 ± 0.11	12.35 ± 1.33
Kidney	11.99 ± 1.11	12.48 ± 0.49	34.64 ± 1.83
Spleen	57.56 ± 6.95	25.85 ± 9.00	10.01 ± 1.35
Liver	19.44 ± 5.79	10.48 ± 1.38	9.10 ± 0.10
Myocardium	35.59 ± 0.97	14.21 ± 0.37	17.54 ± 0.61
Lung	28.19 ± 0.83	22.98 ± 1.42	32.16 ± 2.47
Skin	11.93 ± 0.54	5.16 ± 1.96	32.80 ± 0.30

It should be emphasized that variations of the concentration of indicators are very important, so it is desirable to apply multiple sampling in every determination of total or regional volumes of distribution whenever precision is required.

Conclusions. Determination of regional hematocrit ratio and red cell, plasma and extracellular fluid volume should be done allowing an appropriate mixing time for the indicator and avoiding errors due to changes in local concentration of tracers. Normal values show that hematocrit ratio and volumes are different for each tissue and can no be deduced from arterial hematocrit ratio or corporal red cell plasma and extracellular fluid volumes.

Résumé. La détermination, dans les tissus, du volume globulaire et plasmatique, ainsi que de l'hématocrite, doit être pratiquée après un temps de mélange approprié et en évitant les erreurs dues à la variation locale de la concentration d'indicateur. Les valeurs normales sont fort différentes pour les divers organes étudiés et ne peuvent pas être calculés à partir des résultats obtenus par la mesure de l'hématocrite ou des volumes plasmatiques et globulaires corporels dans le sang artériel.

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[7] J. J. FRIEDMAN, Am. J. Physiol. *196*, 420 (1959).
[8] N. KALISS and D. PRESSMAN, Proc. Soc. exp. Biol. Med. *75*, 16 (1950).
[9] L. WISH, F. FURTH and R. H. STOREY, Proc. Soc. exp. Biol. Med. *74*, 644 (1950).
[10] J. G. GIBSON, A. M. SELIGMAN, W. C. PEACOCK, J. C. AUB, J. FINE and R. D. EVANS, J. clin. Invest. *25*, 848 (1946).

Degeneration Activity after Sympathetic Denervation of the Submaxillary Gland and the Eye

While their parasympathetic postganglionic nerves are degenerating, salivary glands have been found to show a 'degeneration secretion'[1-5] mainly caused by increased leakage of acetylcholine[6]. Transmitter release from degenerating sympathetic fibres could also be demonstrated, but only when the gland cells had been highly sensitized and even then only in 10 cats out of 20. These experiments started about 2 days after denervation, but this may not be the optimal time in the case of sympathetic denervation[7]. Recent experiments show that 'degeneration contraction' of the nictitating membrane starts already 18–30 h after sympathectomy[8].

In 10 cats the right submaxillary gland was sensitized by section of the chorda-lingual nerve, and 2–6 weeks later the right superior cervical ganglion was excised; ether anaesthesia was used; 19–25½ h after sympathectomy, chloralose was given, both submaxillary ducts were cannulated and both nictitating membranes connected to frontal writing levers.

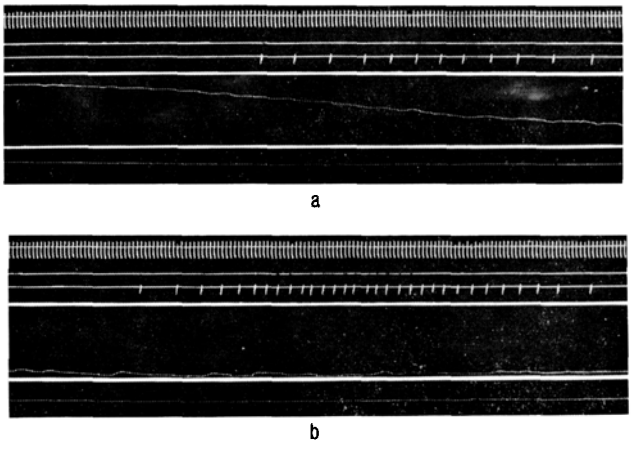
Degeneration contractions of the denervated membranes as previously observed in unanaesthetized[8] or spinal cats[9] were seen in all the cats. In 4 cases they started 20½–24 h after ganglionectomy, to proceed to a maximum and disappear gradually within 7–10 h. In 2 cats the contraction had begun, in 3 it had reached a maximum and in 1 cat the membrane was relaxing when the experiment started (24–25½ h after ganglionectomy). Sometimes small contractions were superimposed on the denervation contraction, as described in the spinal cat[9], and they could occasionally occur when the membrane had relaxed also (see Figure).

[1] N. EMMELIN and B. C. R. STRÖMBLAD, J. Physiol. *143*, 506 (1958).
[2] N. EMMELIN, J. Physiol. *162*, 270 (1962).
[3] D. A. COATS and N. EMMELIN, Experientia *18*, 177 (1962).
[4] P. OHLIN, Experientia *19*, 156 (1963).
[5] I. NORDENFELT, Q. Jl exp. Physiol. *49*, 104 (1964).
[6] N. EMMELIN, in *Secretory Mechanisms of Salivary Glands* (Ed. C. and L. SCHNEYER; Acad. Press, New York 1967).
[7] D. A. COATS and N. EMMELIN, J. Physiol. *162*, 282 (1962).
[8] S. Z. LANGER, J. Pharmac. exp. Ther. *151*, 66 (1966).
[9] S. Z. LANGER and U. TRENDELENBURG, J. Pharmac. exp. Ther. *151*, 73 (1966).

The pupils were inspected at intervals. In 2 of the cats the denervated pupil was found to be considerably larger than the normal one while the membrane was contracted, and even, to a smaller extent, after its relaxation. Degeneration contraction of the pupillary dilator, difficult to find in unanaesthetized cats[8], can thus occasionally occur.

Unprovoked degeneration secretion from the sympathetically denervated submaxillary gland was found in 4 cats. It appeared later than the contraction of the membrane, while the membrane was relaxing or later, even at the end of the experiments 38–40 h after ganglionectomy. The rate of flow varied and between periods of activity there were intervals of secretory rest, as shown in the Figure and described earlier[7]. In 4 other cats degeneration secretion could be provoked 32–37 h after ganglionectomy by injecting noradrenaline i.v. After the immediate secretory response to this drug, there was a pause of 5–7 min, and secretion was then recommenced. In 2 cats there was no degeneration secretion.

The Figure shows degeneration secretion appearing $26^1/_2$ h after ganglionectomy, when the degeneration contraction of the membrane was wearing off. A second, and more marked period of secretion 30 h after ganglionectomy is also shown. At that stage the membrane had relaxed fully. It is evident that contraction and secretion occur independently of each other; they cannot be due to catecholamines in the blood but must be caused by local events. On the whole, secretion appears later than the contraction. This may reflect some difference between the secretory fibres of the gland and the motor fibres of the membrane; it seems questionable whether an explanation can be provided by the finding[10] that the time of appearance of degeneration activity depends on the length of the degenerating nerve. That degeneration secretion was obtained less regularly than contraction of the membrane is very likely due to the fact that the sympathetic secretory innervation of the cat's submaxillary gland is variable, being sometimes extensive, sometimes very scanty[11,12].



Degeneration contraction and degeneration secretion. (a) Started $25^1/_2$ h, (b) $29^1/_2$ h after sympathectomy. Records in each section from above: time in min, secretion from the left, from the right submaxillary gland and contractions of the right and the left nictitating membrane.

Zusammenfassung. Nach Sympathectomie erscheint eine «Degenerationskontraktion» der Nickhaut nicht nur in wachen[8] oder spinalen[9], sondern auch regelmässig in narkotisierten Katzen. Die «Degenerationssekretion» von der Submaxillarisdrüse tritt später auf; aber auf Grund der Variabilität der sympathisch sekretorischen Innervation sieht man sie nicht bei allen Tieren.

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[10] N. Emmelin, J. Physiol. *188*, 44P (1967).

[11] N. Emmelin, in *Handbook of Physiology, Alimentary Canal I* (Waverly Press Inc., Baltimore 1967).

[12] Supported by a grant from the Swedish Medical Research Council.

A Histoautoradiographic Study of Protein Loss in the Intestine of Antifolic-Treated Rats

Since 1962 in our laboratory we have been able to work up a model of enteropathy in rats treated with different dosages of antifolic drugs or antibiotics. More recently, histological changes of intestinal mucosa and metabolism of Cr^{51}-albumin have been investigated in these animals (Marano et al.[1]). Our experiments showed albumin losses from the bowel and a parallel fall of protein plasma levels.

The purpose of this study is to clarify the mechanism of protein loss and to establish a relationship between experimental and human pathology.

One hundred male albino rats (weighing between 200 and 250 g) were divided into 3 groups: the first group of 40 rats received by stomach tube 0.2 mg/kg body weight of aminopterine daily for 7 days; the second group of 40 rats was given the same drug at the dosage of 0.04 mg/kg daily for 60 days; the rats of the third group, used as a control, received 2 ml of saline daily.

At the end of the treatment, each rat was injected with 15 μg of Cr^{51}-albumin into the vein of the tail. The rats were sacrificed in groups of 7, respectively 6, 12, 24, 48 and 72 h following injection of albumin, and a loop of the small bowel was taken for histology. This sample was washed in saline and fixed in 10% formol saline. The presence of labelled albumin in the intestinal mucosa was demonstrated by means of the autoradiographic technique; radioactive Cr in the sections appeared as black spots.

Autoradiographic technique. After fixation, the specimens were washed in water, dehydrated and embedded in paraffin at 56–58 °C.

The sections, 5–7 μ thick, were placed in water at 37 °C, distended on clean slides and kept overnight at 37 °C; the paraffin was then removed and the sections were left for 5 min in absolute alcohol to dry at room temperature.

The procedure was carried out in a dark room (Wratten Safelight No. 1 lamp) as follows. Rectangular pieces of film (Kodak autoradiographic plates type A) as large as

[1] R. Marano, G. Pastore and O. Schiraldi, Rass. Fisiopat. clin. terap. *37*, 544 (1965).