Dadurch ist die unterschiedliche Lage der linearen Regressionslinien im Diagramm ersichtlich.

Statistische Erfassung. Die Abweichung des Kurvenverlaufs von Fall Nr. 2111 gegenüber dem des Falles Nr. 2113 erwies sich im Bereich zwischen A = 25% und A = 87,5% als hochsignifikant ($p < 0{,}001$), desgleichen zwischen Fall Nr. 2111 und Nr. 2019 im Bereich von A = 12,5–62,5%. Auch die Abweichung zwischen Fall Nr. 2113 und Nr. 2019 war in den Bereichen A = 12,5 – 50% und A = 75 – 100% hochsignifikant ($p < 0{,}001$).

Histologisch zeigen alle vier untersuchten Lymphknoten einen normalen Aufbau.

Diskussion. Bei dem Vergleich der erhaltenen Ergebnisse zeigt sich sowohl im Histogramm als auch bei den Absorptionsniveaukurven eine beträchtliche Streubreite der zytophotometrischen Messwerte.

In der Literatur wird das Auftreten einer solchen Streuung an bestimmten Geweben innerhalb einer Tiergattung gelegentlich beschrieben[14,15]. Zum Teil wurde dies als eine tatsächlich vorliegende Variation auf Grund erhöhter Stoffwechselaktivität in verschiedenen Zellpopulationen interpretiert[16,17], zum Teil wurde die Bedeutung der von der DNS-Konstanz abweichenden Befunde vollkommen verneint und auf Messfehler zurückgeführt[17,18]. Es sind ausserdem auch präparative Ungenauigkeiten bei der Herstellung, Fixierung und Färbung in Betracht zu ziehen, die trotz gleicher und gleichzeitiger Behandlung unwillkürlich auftreten können[19].

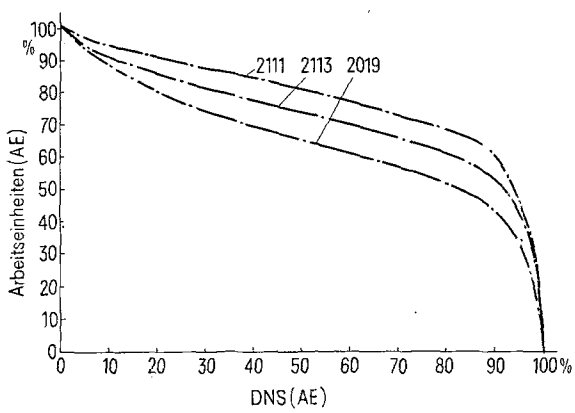

Fig. 2. Normierte Absorptionsniveaukurven (arithmetische Mittelwerte).

Bei den von uns untersuchten Schaflymphknoten ist die Möglichkeit eines präparativen Fehlers sowohl durch eine rasche und gleichzeitige Herstellung als auch durch die stets gleichzeitige weitere Behandlung der Präparate auf ein Minimum reduziert worden. Dadurch wird deutlich, dass die hier aufgezeigten Schwankungen zwischen den Normalpopulationen weitgehend von der Behandlung unabhängig sind und demnach bei vergleichenden Untersuchungen berücksichtigt werden müssen. Vergleicht man die hier gefundenen Ergebnisse mit entsprechenden Untersuchungen an leukotisch veränderten Lymphknoten[12], so zeigen letztere eine ähnlich breite Streuung der Messwerte wie die Ergebnisse aus der Normalpopulation. Eine exakte Interpretation erscheint daher nicht möglich. Aus diesem Grund erachten wir bei zytophotometrischen Untersuchungen die Kontrolle mehrerer Normalpopulationen als unbedingt notwendig.

Summary. Cytophotometric investigations of lymphocytic populations of 4 sheeps were carried out to determine the diploid standard value for comparative investigations of lymph node cells from sheep with lymphatic leucosis. Inspite of exact and simultaneous preparation there were marked and significant differences in the histograms as well as in the diagrams of absorption level separation. Therefore, the control of several normal cell populations is essential before starting cytophotometric investigations of neoplastic cells.

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[14] M. P. VIOLA-MAGNI, J. Cell Biol. *25*, 415 (1965).
[15] H. ROELS, Int. Rev. Cytol. *19*, 1 (1966).
[16] S. R. PELC, Expl. Geront. *5*, 217 (1970).
[17] M. ALFERT und H. SWIFT, Expl. Cell Res. *5*, 455 (1953).
[18] K. PATAU und H. SWIFT, Chromosoma *6*, 149 (1953).
[19] C. LEUCHTENBERGER, in *General Cytochemical Methods* (Ed. J. F. DANIELLI; Academic Press, New York 1958), vol. 1.

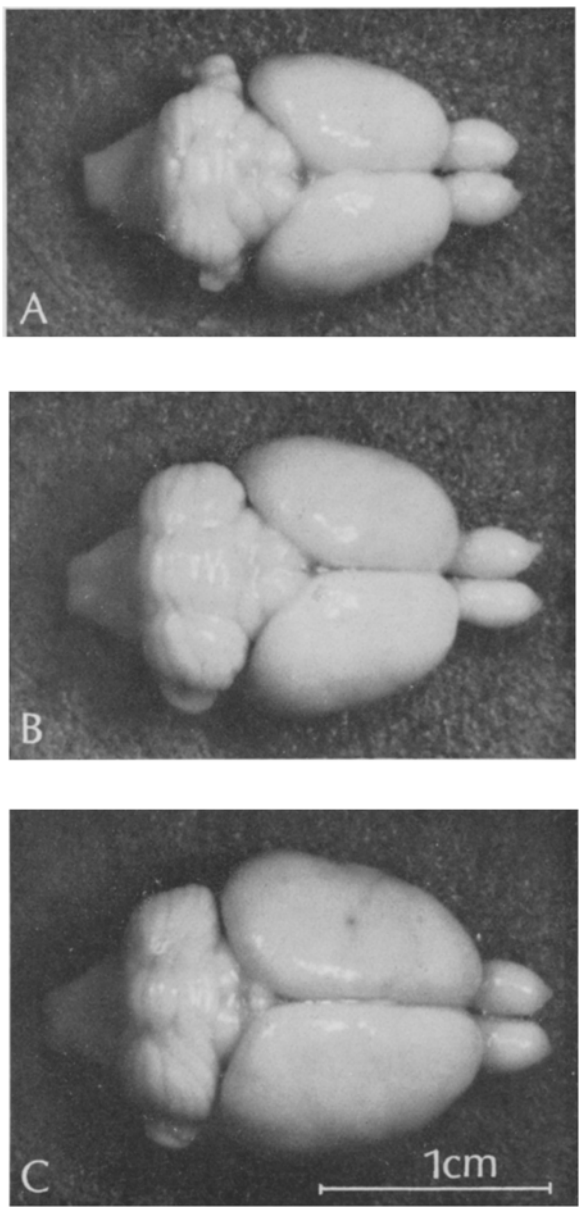
Effects of Ethylnitrosourea on the Development of Brain

DRUCKREY[1–3] and his associates, in their pioneering studies on experimental brain tumors, have shown that administration of Ethylnitrosourea (ENU) to pregnant rats produces brain tumors in the offsprings of these animals. These investigations strongly suggest that ENU can reach the developing embryos in rats transplacentally. From neuroembryological viewpoint this technique provides a valuable experimental tool for the study of neuroembryogenesis in the rat.

Laboratory-bred Wistar albino rats were administered ENU i.v. between days 14 and 21 of gestation (dose: 60 mg/kg body weight). On each day of gestation at least 1 animal was injected with the chemical. From each litter born to these animals 2 pups each were sacrificed on days 1, 3, 10, 15, 21 and 75 after their birth.

On macroscopic examination it was found that the overall size of most of the brains was smaller than that of normal brains of the same age, and this suggested that ENU had interfered with the normal development of the central nervous system. Cerebral cortex and cerebellum were consistently affected, and brain stem and olfactory bulbs did not appear affected. When the brains from all the animals of 21 days age were placed in a series and

[1] H. DRUCKREY, S. IVANKOVIČ and R. PREUSSMANN, Nature, Lond. *210*, 1378 (1966).
[2] S. IVANKOVIČ, H. DRUCKREY and R. PREUSSMANN, Naturwissen *53*, 410 (1966).
[3] S. IVANKOVIČ and H. DRUCKREY, Z. Krebsforsch. *71*, 320 (1968).



Brains of 21-day-old animals, that received ethylnitrosourea on days 14(A), 17(B) and 20(C) of gestation transplacentally.

compared, it was evident that the cerebral cortex and cerebellum had been affected in a graded fashion. In the animals that received ENU on day 14 of gestation cerebral cortex and cerebellum appeared very small in size (Figure A), and these two neural structures became larger and larger in graded manner as we approached the animals injected on higher stages of gestation (Figures B and C). In other words, ENU had severe effects on the brains of animals injected on earlier stages of embryonic development and these effects became less and less pronounced in the brains of animals injected at increasingly advanced stages of embryogenesis. Identical observations were made on the brains of animals that were sacrificed on days 15 and 75 of postnatal life, which suggested that these gross developmental defects could be detected during weaning period and that they were permanent in nature.

When these brains were dissected into blocks for histology it was found that even the internal structures such as basal ganglia, thalamus and hypothalamus, which could be compared under dissecting microscope, showed similar graded changes in their size. Gross changes in the brains of animals treated with ENU during their embryonic development reflect the possibility of massive changes in the histogenesis of the nervous system as well as in the differentiation of the existing stock of neurons. However, exact nature of these changes will be determined from histological evaluation of this material. It is worthwhile to comment that similar graded changes in the overall size of the brain have been produced by low level doses of X-ray irradiation of the rat embryos in our laboratory as well as at the hands of other investitigators[4,5].

Zusammenfassung. Wiederholte Applikation von Äthylnitroso-Harnstoff bei trächtigen Ratten bewirkt bei Föten eine Hemmung der Gehirnentwicklung, wobei vor allem die Hemisphären und das Cerebellum unterentwickelt bleiben. Diese Hemmung ist am ausgeprägtesteniw wenn die Behandlung am 14. Tag der Trächtigkeit einsetzt.

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Department of Biological Sciences, Purdue University, Lafayette (Indiana 47907, USA), 16 February 1972.

[4] S. P. Hicks, Physiol. Rev. *38*, 337 (1958).
[5] R. Rugh, A. Rev. nucl. Sci. *3*, 271 (1953); *9*, 493 (1959).

Milk Microsomes, Viruses, and the Milk Fat Globule Membrane

In 1954 Morton[1] isolated lipoprotein particles from milk fat globules which he later demonstrated had considerable biochemical homology with microsomes derived from endoplasmic reticulum of mammary gland.

Recent examination of mammary tissue of cows and goats by conventional transmission microscopy has shown that the milk fat globule membrane (MFGM) rapidly changes its structure after release of the globule from the secretory epithelium[2,3] thus confirming the prediction of Knoop, Bargmann and their co-workers (for a review, see reference[4]). Immediately after release from the cell the fat globules have a continuous unit membrane plus a thin coat of cytoplasmic material (initial MFGM). This appears to rapidly bleb off in small vesicles or lift off in sheets into the milk serum leaving small blebs and plaques remaining on the 'secondary MFGM'[3], which is seen as a continuous single dense line around the globule[3]. This description has now been found true for milk fat globules

[1] R. K. Morton, Biochem. J. *57*, 231 (1954).
[2] F. B. P. Wooding, J. Cell Sci. *9*, 805 (1971a).
[3] F. B. P. Wooding, J. Ultrastruct. Res. *37*, 388 (1971b).
[4] W. Bargmann and U. Welsch, in *Lactogenesis* (Eds. M. Reynolds and S. J. Folley; University of Pennsylvania Press, Philadelphia 1969), p. 43.