Both regression tests were significant at a 1% level. The regression tests for the control group were not significant, so the respective lines for this group in diagrams represent only the average values.

Discussion. The longer exposure to urethan anaesthesia produced a statistically significant ($P < 0.01$) increase in the liver glycogen content of intact mice. Urethan in a single dose decreases the liver glycogen content of rats being that effect interpreted as a consequence of the liberation of catechol amines. Urethan really stimulates the secretion of adrenaline from the adrenal medulla, and it is reasonable to suppose that under urethan intoxication there is a continual release of catechol amines from the adrenal glands with a consequent reduction in the circulating catechol amines due to depletion of the gland. This fact may explain the results achieved for glycogen. Considering RNA, the decreased cytophotometric values observed are in accordance with the biochemical results obtained by other authors.

Résumé. Utilisant la méthode cytophotométrique on a observé l’accroissement du glycogène et la réduction de ARN dans le foie de souris soumises au traitement toxique par l’uréthane.

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Formation and Decay of Virus-Specific Polysomes in vitro

It has been shown earlier that RNA extracted from Newcastle disease virus (NDV) infected cells forms in vitro virus-specific polysomes after contact with chick embryo ribosomes. However, a mixture of both viral and cellular RNAs was used in that study, which hampered interpretation of the results obtained. The present report records the study of the formation of polysomes of virion NDV RNA and their fate in the process of translation.

Experiments were performed with NDV RNA extracted by the detergent phenol method and precipitated by ethanol with 0.2% sodium acetate. The method for preparation of chick embryo ribosomes and protein-synthesizing system have been described elsewhere. To destroy endogenous polysomes, ribosomes in all experiments were preincubated at 32°C for 40 min with amino acids, energy-regenerating system and cell fraction S100, containing transport RNA and other components of protein-synthesizing system.

In the first series of experiments, NDV RNA was added to the incubation medium for various intervals; the mixture was supplemented with C4-algae hydrolysate and at the end of incubation period was rapidly chilled; polysome formation was analyzed by centrifugation in linear sucrose density gradients. Gradient fractions were sedimented with 10% trichloroacetic acid (TCA), the

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Figure 1. Kinetics of formation and decay of virus-specific polysomes after incubation of NDV RNA with chick embryo ribosomes and C4-amino acids in cell-free protein-synthesizing system at 32°C for 5 (A), 15 (B) and 45 min (C). Samples were centrifuged in linear 17-40% sucrose density gradients at 25,000 g/min for 1 h and 45 min. Designations: A) Optical density at 260 nm (1), radioactivity of fractions without (2) and with (3) the addition of NDV RNA into the system. B) Radioactivity of fractions with the addition of NDV RNA as well as 0.02M EDTA (2) and 50 μg/ml of pancreatic ribonuclease (3). C) Radioactivity of fractions with the addition into the system of NDV RNA as well as 50 μg/ml of puromycin (2) and 100 μg/ml cycloheximide (3).
precipitates were washed with 5% TCA on Millipore filters. Radioactivity was measured in a Packard-Tricarb liquid scintillation counter.

Figure 1 presents the results of this series of experiments indicating that preincubation of ribosomes is accompanied by disintegration of endogenous polysomes into monosomes and ribosomal subunits (Figure 1A-1) in which protein synthesis does not occur (Figure 1A-2). The addition of NDV RNA to the system is coincident with the formation of polysomes with a sedimentation coefficient of 240S (Figure 1A-3). 15 minutes later, a part of the heavy polysomes disappears and polysomes with sedimentation coefficients of 140 to 160S are prevalent (Figure 1B-1), which are destroyed in the presence of EDTA (Figure 1B-2) and ribonuclease (Figure 1B-3). Virus-specific polysomes break up in 45 min (Figure 1C-1); the addition of puromycin into the incubation mixture accelerates the process (Figure 1C-2) and the addition of cycloheximide slows it down (Figure 1C-3). Dissociation of polysomes is due to the synthesis of proteins, the amount of which increased in the upper gradient fractions.

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Fig. 2. Kinetics of formation and decay of virus-specific polysomes after incubation of NDV\(^{3H}\)-RNA with chick embryo ribosomes in protein-synthesizing system for 5 (A), 15 (B) and 45 min (C). Conditions of the experiment are the same as above. Designations: 1. Optical density at 260 nm; 2. Radioactivity of gradient fractions. The first gradient fractions are the pallets (shown by the arrows).

Fig. 3. Kinetics of degradation of NDV\(^{3H}\)-RNA inoculated with the cell-free protein-synthesizing system and extracted from it by phenol 1 (A), 15 (B) and 45 min (C) after incubation. Samples were centrifuged in linear sucrose density gradients at 18,000 g/min for 14 h. Designations: 1. Optical density at 260 nm. 2. Radioactivity of gradient fractions.