

Relation between Nucleolar Volume and Cell Body Content of Ribonucleic Acid in Supra-optic Neuron

DURING an investigation of the influence of increased functional activity on the ribonucleic acid in neurosecretory cells of the supra-optic nucleus of the rat, an observation of more general interest was made. The amount of ribonucleic acid in the cell bodies was measured microchemically¹ and the volumes of their nucleoli were determined on stained slides. Cells from control animals were investigated as well as from animals that had been subjected to administration of sodium chloride for two months. Apart from the finding that increased functional load (increased hormone production) gives rise to statistically significant increases in amounts of ribonucleic acid and nucleolar volumes, it was found that the ribonucleic acid content per cell body was directly proportional to the nucleolar volume (Fig. 1). The ribonucleic acid content per cell body nearly equals that of the cytoplasm in these cells, so it is apparent that there is a proportionality between nucleolar volume and cytoplasmic content of ribonucleic acid. On the other hand, it can be shown that it is unlikely that proportionality between nucleolar volume and nucleolar amount of ribonucleic acid should exist as well as proportionality between nucleolar and cytoplasmic amounts of ribonucleic acid.

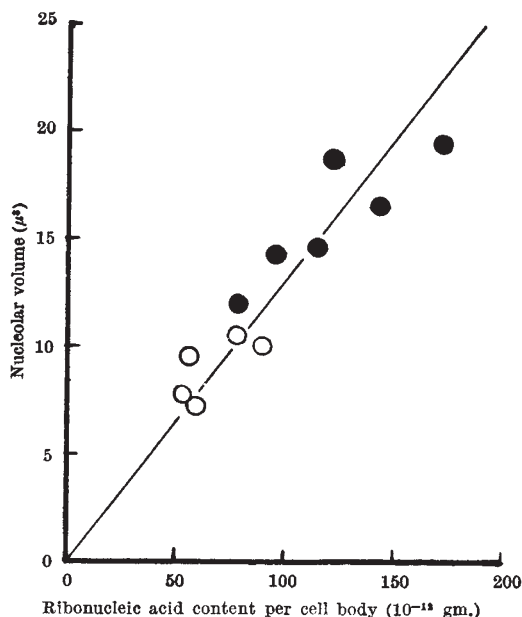


Fig. 1. Values for nucleolar volumes plotted against values for ribonucleic acid content in cell bodies of supra-optic neurons. Mean values per animal are given. Open circles designate controls, filled circles experimental animals

It is of interest to note that LaVelle², from a study of stained slides, came to the conclusion that there exists a proportionality between the size of the nucleoli and the amount of Nissl substance in nerve cells proper. As the basophilia of the Nissl substance is due to ribonucleic acid, these results are in good agreement, indicating that the relations may have more general application.

A full account of this investigation, which was supported by grants from the Swedish Medical

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¹ Edström, J.-E., *Biochim. Biophys. Acta*, **12**, 361 (1958).

² LaVelle, A., *J. Comp. Neurol.*, **104**, 175 (1956).

Antigenicity of Avidin

AVIDIN has been shown to bind the vitamin biotin, but the mechanism of binding is still unknown. Modification of the avidin molecule by acetylation, esterification or by blocking the amide, guanidyl, imidazole and phenolic groups did not result in a loss of its ability to bind biotin¹.

The mechanism and site of binding of an antigen by its homologous antibody is also largely unknown. In an effort to answer the question whether the locus (loci) of binding of biotin by avidin (on the avidin molecule) was (were) the same or one of the loci binding antiavidin to avidin, we have studied the specific precipitation of avidin by rabbit antiavidin serum in the presence and absence of biotin. Avidin concentrates were prepared according to Dhyese², and adult male rabbits were immunized by bi-weekly parenteral administration of these concentrates mixed with Freund's adjuvant supplied by the Difco Co. In all, two courses of immunization were employed. The avidin used for the precipitin reaction was obtained by dialysis of these concentrates, when avidin formed a precipitate. The dialysis was carried out against distilled water over a 48-hr. period with 3-4 changes of the water. The precipitin reaction was carried out according to standard methods³ first at 37° C. for 3 hr. and then at 5° C. for 48 hr. The results are given in Table 1.

Protein was estimated according to Lowry *et al.*⁴ In one series (A) of experiments the precipitation of avidin in the absence of biotin was studied as a control. In the B series, 5γ of biotin were added to the avidin and incubated at 37° C. for 1 hr. At the end of this period, antiserum was added and the incubation continued for 3 hr. at 37° C., and the precipitation continued for 48 hr. in the refrigerator. In the C series, precipitation of avidin was effected first and then 5γ of biotin were added. The results do not show any marked enhancement or inhibition of precipitation of avidin by its homologous antiserum, in the presence or in the absence of biotin. The binding of the antibody was not affected by the previous formation of the avidin-biotin complex

Table 1. PRECIPITATION OF AVIDIN BY RABBIT ANTI-AVIDIN

No.	Amount avidin (γ)	Total protein precipitated, using 1 ml. of serum		
		Exp. A	Exp. B	Exp. C
1	29.6	430	345	364
2	59.2	705	690	664
3	88.8	940	920	1,004
4	118.4	1,100	1,265	1,284
5	148.0	1,530	1,505	1,484
6	222.0	1,835	1,605	1,689
7	296.0	1,605	1,695	1,914