

Deoxyribonucleic Acid Content of the Cell-Nuclei in the Adrenal Medulla after Exposure to Low Temperatures

As has been shown by H. Roels¹ in other endocrine glands, I have found that, in the adrenal medulla of the white rat, there is a correlation between the mean deoxyribonucleic acid content of the nuclei and cell activity. A preliminary communication on this topic² reports an increase of the nuclear deoxyribonucleic acid content after stimulation by means of daily injections of insulin, and also an important decrease in the resting gland after bilateral splanchnectomy.

The present communication is devoted to experiments on stimulation of the adrenal medulla by means of exposure to low temperature; the stress produces hypersecretion of adrenaline.

Five male rats were kept for a fortnight in a refrigerator at nearly 4° C. They were killed, together with five controls. The adrenals and a control testis were simultaneously fixed in alcohol, formol and acetic acid (75:20:5), dehydrated and embedded. Sections (10 μ) of the different objects were mounted on the same slide, in order to obtain homogeneous Feulgen staining. The deoxyribonucleic acid content of the individual nuclei was measured *in situ* by Lison's histophotometrical method³. As the deoxyribonucleic acid contents obtained were given in arbitrary units, we used as a standard a 'theoretical diploid value', being half the mean content of 50 spermatocytes I measured in the control testis. The results are summarized in Table 1.

Table 1

	\bar{x}	$\log \bar{x}$	s	$s \log \bar{x}$	n
Spermatocytes I	2,663	3.425	0.004	0.006	50
		Theoretical 2s value, 1,332			
Controls					
234	1,308	3.116	0.046	0.006	50
235	1,366	3.135	0.067	0.009	50
236	1,200	3.079	0.045	0.006	50
237	1,210	3.082	0.055	0.007	50
238	1,267	3.102	0.053	0.007	50
Pool	1,268	3.103	0.052	0.003	250
Cold-treated					
289	1,469	3.167	0.043	0.006	50
290	1,426	3.154	0.039	0.005	50
291	1,389	3.142	0.042	0.005	50
292	1,372	3.136	0.034	0.004	50
293	1,446	3.160	0.039	0.005	50
Pool	1,414	3.150	0.041	0.008	250

As the different individual deoxyribonucleic acid contents of the nuclei present a log-normal distribution, the statistical computations are made using the logarithms of these values.

Comparing cold-treated animals and controls, it is clear that the stimulation of the adrenal medulla is accompanied by an increase in the mean deoxyribonucleic acid content of the nuclei, which for the pooled results reaches 11.4 per cent ($t = 7.0$; $P < 0.001$).

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¹Roels, H., *Nature*, **174**, 514 (1954); *C.R. Soc. Biol.*, **150**, 2273 (1956); *Arch. Biol.*, **67**, 211 (1956); *Nature*, **182**, 873 (1958).

²Leeman, L., *Exp. Cell Res.* (in the press).

³Lison, L., *Acta Anat.*, **10**, 333 (1950).

New Types of Salting-out Paper Chromatograms of Antibiotics

ACCORDING to Miyazaki, Omachi and Kamata¹, antibiotics can be grouped according to their salting-out chromatograms. Pure antibiotics and materials containing antibiotics were examined by the aid of ascending paper chromatography. As irrigation solvents in each case and with each antibiotic, distilled water and rising concentrations of ammonium chloride (0.5, 1, 2, 3, 5, 10, 20 per cent and saturated) were used. Location of the spots of the antibiotics, that is, the R_F values, were ascertained bio-autographically.

Antibiotics were divided into four groups. In those of group *A* the R_F value is not correlated with the concentration of the ammonium chloride solution, identical values being found at all concentrations and in distilled water. To this group belong penicillin, grisein, chloramphenicol, aureothricin, and the special substance 2. In group *B* are those antibiotics which have an R_F value 0 in distilled water, and increasing R_F values with increasing concentrations of ammonium chloride. They are: streptomycin, streptothricin, luteomycin, fuscomycin, xanthomycin and flaveolin. Group *C* is different from group *B*, displaying the highest R_F value in distilled water, and lowering of the values with increasing concentration of the solution of ammonium chloride. Members of this group are: actinomycin and griseoflavin. Group *D* consists of antibiotics which do not display any movement whatever in an irrigating solvent from the starting point; eventually increased tailing tendency was observed with higher concentrations of the solutions of ammonium chloride. Aureomycin and terramycin are members of this group.

These results obtained by Miyazaki *et al.* have been confirmed by my experiments with salting-out paper chromatography of antibiotics. Moreover, I have been able to show the existence of other types of salting-out paper chromatograms of antibiotics with antibacterial action, of actinomycetal origin.

In group *E* are antibiotics which have an R_F value 0 in distilled water, and an initial increase with rising concentrations of the solution of ammonium chloride, the maximum being ($R_F =$ nearly 1) in 5 per cent ammonium chloride. Thereafter a decrease occurs, as shown in Fig. 1. The line connecting the different R_F values is of the shape of a truncated parabola. This type of paper chromatogram was

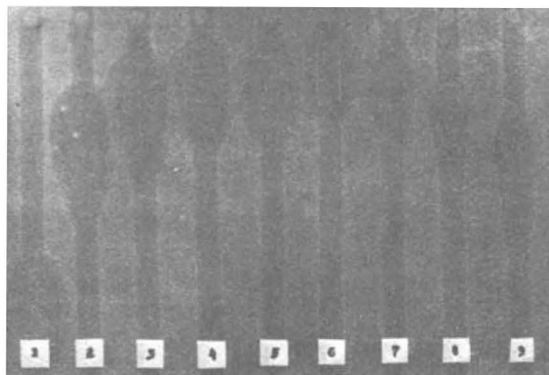


Fig. 1. The salting-out paper chromatogram of erythromycin. 1, Distilled water; 2, 0.5; 3, 1; 4, 2; 5, 3; 6, 5; 7, 10; 8, 20 per cent; and 9, saturated ammonium chloride solutions