Action d'une dose unique de rayons X sur les acides nucléiques de la moelle osseuse (700 r).

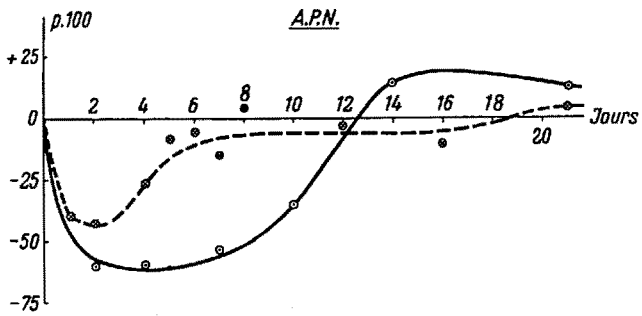

Fig. 2. Action d'une dose unique de rayons X (700 r) administrée en irradiation totale et locale sur les *acides ribonucléiques* (ARN.) de la moelle osseuse des membres postérieurs chez le rat. Résultats exprimés en pour-cent des valeurs témoins.

------ Irradiation unilatérale —— Irradiation corporelle totale

La Figure 1 présente l'évolution de la quantité absolue de l'ADN. de la moelle osseuse des membres postérieurs après irradiation soit de l'animal total, soit des membres gauches seulement. On notera que la réduction de l'ADN. est bien plus accusée dans le cas de l'irradiation corporelle totale puisqu'elle peut atteindre en moyenne – 75 %, alors qu'après irradiation locale, la réduction ne dépasse pas – 60 %. De plus, le retour à la normale est beaucoup plus rapide après irradiation locale.

La Figure 2 représente les modifications subies par l'APN. dans les mêmes conditions expérimentales. Là encore, nous notons en moyenne une réduction de – 60 % après l'irradiation corporelle totale, pour une diminution de – 45 % seulement dans le cas de l'irradiation locale. L'appauvrissement de la moelle en APN. est également de plus longue durée après une irradiation corporelle totale.

Les modifications que nous venons de décrire prouvent que la réduction de l'activité de la moelle, à savoir de la prolifération nucléaire que reflète l'ADN. et des synthèses cytoplasmiques qu'indique l'APN., est bien plus importante quand le corps entier est irradié. Il convient donc d'admettre que, même pour des tissus très sensibles aux rayons X, ce n'est pas seulement l'action directe des rayons qui en est cause mais qu'il existe des effets secondaires consécutifs à l'irradiation corporelle totale; ces effets contribuent à aggraver considérablement la destruction de la moelle et à retarder sa régénérescence.

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Summary

The biochemical effects on bone marrow of a same dose of X rays (700 r) induced by a total body irradiation or a local irradiation are compared. The ribonucleic acid and the desoxyribonucleic acid of bone marrow show a fall during days 2 to 14 after irradiation, followed by complete recovery on day 21. A significant difference is noted during this period between animals exposed to a total or local irradiation system. The changes consecutive to X rays induced by a total irradiation are prominenter and remain longer than in local irradiation. The direct effect of X rays is probably associated with secondary injury of physiological connexions or other substances affected in total body irradiation.

The Nature of Brdička's Cancer Test

BRDIČKA[1] studied catalytic polarographic waves produced by adding denatured blood serum to an ammoniacal cobalt solution. He found that the catalytic wave was lower for carcinoma serum than for normal serum. On treating the serum with sulphosalicylic acid (S.S.A.) as a deproteinating agent, the catalytic wave of the filtrate was higher for cancerous serum than for normal serum[2]. This last phenomenon has been used in cancer diagnosis[3] but the test is also sometimes positive with sera from patients with high fevers. There have been suggestions[4] that the substance responsible for the catalytic wave of the S.S.A. filtrate is of mucoid nature.

We have isolated the substance responsible for the BRDIČKA filtrate reaction by dialysis of the S.S.A. filtrate of cancer sera, followed by alcohol fractionation according to the procedure of MEYER[5]. This substance, the "cancer substance", is a white powder, and it gives catalytic waves in the BRDIČKA test solution identical with those given by the S.S.A. filtrates from cancerous serum. For comparison with a known mucoprotein, we prepared ovomucoid from egg white using FREDERICQ and DEUTSCH's[6] method. When the ovomucoid was added to the BRDIČKA test solution, it gave a catalytic wave identical with those given by the "cancer substance" and the S.S.A. filtrates.

The "cancer substance" and ovomucoid were then partially hydrolysed to liberate the sugars only. Paper chromatographic analysis of the hydrolysates showed that both "cancer substance" and ovomucoid contained the same sugars–galactose, mannose, and glucosamine. Complete hydrolysis of the "cancer substance" and ovomucoid, followed by paper chromatography of the hydrolysates, showed that both substances contained the same amino acids in so far as these could be identified. A quantitative estimation of cystein was made by SULLIVAN's[7] method, giving 3·4 % cystein in ovomucoid and 4·12 % cystein in the "cancer substance". YOUNG[8] reports 3·95 % ovomucoid and FREDERICQ and DEUTSCH[6] 6·7 %. These determinations confirm that the "cancer substance" is a mucoprotein.

MEHL and co-workers[9] observed that mucoprotein levels in normal blood are in general lower than those in cancerous blood. They isolated the mucoproteins from normal blood and found on electrophoresis that the mucoprotein fraction consisted of at least 3 components. They prepared an electrophoretically homogeneous fraction by ammonium sulphate fractionation[10]. Through the kindness of Dr. MEHL, we have examined a sample of his mucoprotein and find it also to contein galactose, mannose and glucosamine. We also found that in the BRDIČKA test solution the catalytic wave of MEHL's mucoprotein and the "cancer substance" are qualitatively and quantitatively identical. On the other hand, the catalytic wave of ovomucoid is identical with the

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wave produced by ten times greater quantities of MEHL's mucoprotein or the "cancer substance". The "cancer substance" and MEHL's mucoprotein show practically identical electrophoretic patterns.

1

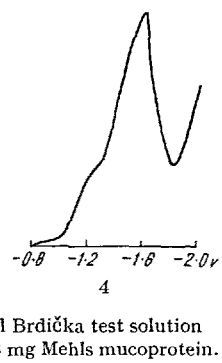
5 ml Brdička test solution
Sensitivity × 200.

2

5 ml Brdička test solution
+ 0·1 mg ovomucoid
Sensitivity × 200.

3

5 ml Brdička test solution
+ 1 mg cancer substance
Sensitivity × 200.



4

5 ml Brdička test solution
+ 0·8 mg Mehls mucoprotein.
Sensitivity × 200.

It has thus been demonstrated that the substance responsible for the BRDIČKA reaction is a mucoprotein very similar if not identical with MEHL's mucoprotein, which is a component of normal blood. This suggests that BRDIČKA's reaction indicates an increase in the level of normal mucoproteins in the blood and not the presence of a new substance which is absent in the blood of healthy subjects.

The authors thank Dr. J. W. MEHL of the University of Southern California for a sample of mucoprotein, Dr. H. H. MACKINNON of the Victoria Public Hospital, Fredericton, for samples of cancer sera, and also Dr. A. SEHON and Mr. KENNEDY of McGill University for carrying out electrophoretic measurements.

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Zusammenfassung

Es wird gezeigt, dass die Brdičkasche Filtratreaktion durch eine Erhöhung der Konzentration des normalerweise im Serum vorkommenden Mucoproteins verursacht wird. Dieses Mucoprotein enthält Cystein, so dass keine Modifikation der ursprünglichen Interpretierung der Reaktion durch BRDIČKA erforderlich scheint.

Acid-soluble Nucleotide Derivatives in Cell Nuclei Isolated in a Non-aqueous Medium

In recent studies on the composition of cell nuclei isolated by non-aqueous procedures (KAY, SMELLIE, HUMPHREY, and DAVIDSON[1]) it has been shown that considerable amounts of material are removed from such nuclei when they are extracted with dilute citric acid. This extractable material includes some of the nuclear protein and a considerable proportion of the nuclear ribonucleic acid (RNA) together with other acid-soluble substances including simple nucleotides. The presence of acid soluble nucleotides and related nucleosides and free bases is of considerable interest inasmuch as they are likely to be connected in some way with the metabolic role of the nucleus. In this brief note some observations on these components of cell nuclei are recorded.

Samples of nuclei prepared from a variety of tissues according to the method described by KAY *et al.*[1] were extracted in the cold with 0·7 *N* perchloric acid in the proportion of 5 ml/100 mg nuclei. The mixture was centrifuged and the residue was washed twice with 2 ml cold 0·7 *N* perchloric acid. The residues were reserved for estimation of RNA and DNA by the modification of the SCHMIDT-THANNHAUSER method described by SMELLIE, HUMPHREY, KAY, and DAVIDSON[2]. The combined extracts were treated with butanol to remove nucleosides and free bases leaving the nucleotides and inosine in the aqueous phase according to the method of GOLDWASSER[3]. The butanol extracts were taken to dryness and digested with 12 *N* perchloric acid at 100° to yield the individual purine and pyrimidine bases. The aqueous phase containing the nucleotides and inosine was also evaporated to dryness and digested with perchloric acid. Portions of the digest were applied to WHATMAN 3MM paper and the bases separated by two dimensional chromatography, using descending isopropanol: HCl (WYATT[4]) followed by ascending butanol: ammonia (MAC NUTT[5]). Appropriate blanks were also run. The bases were located in ultraviolet light and were eluted with 0·1 *N* HCl for all except guanine which required 1·6 *N* HCl. From readings of the optical density at appropriate wavelengths in the ultraviolet the amounts of each base were calculated and were then related to the DNA-phosphorus present in the original extracted sample. The values were thus obtained as μg base per mg DNA-P.

In some samples the total ultraviolet absorption of the perchloric acid extract was measured. For this estimation the perchloric acid extracts were diluted so that 1 ml was derived from an amount of nuclei containing 10 μg DNA-P. Absorption readings in the ultraviolet at 260 mμ were then made in the Beckman spectrophotometer, using appropriate perchloric acid blanks.

The values for the total ultraviolet absorption at 260 mμ of perchloric extracts of rabbit nuclei are shown in Table I. Since the figures quoted are related to the same amount of DNA-P it may be assumed that the various extracts are derived from comparable numbers of nuclei. The values show a wide scatter but the general pattern which emerges is that the nuclei from adult

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