

La consommation d'oxygène a été mesurée à l'aide du ludion de LINDERSTRØM-LANG ET HOLTER⁶. Elle a été suivie sur des amibes témoins et traitées à la RNase, mises par groupes de 30 dans les ludions cylindriques déjà utilisés par BRACHET^{1,3}. Comme nous n'avons pas tenu compte des lectures de la première heure et que le remplissage des ludions est assez long, les premières mesures commençaient au bout d'1 heure $\frac{1}{2}$ à 2 heures d'action de la RNase. La concentration a été la même que précédemment.

Les résultats du Tableau II montrent que la RNase n'a pas d'effet massif sur la respiration: la moyenne est pratiquement identique dans le cas des amibes témoins et des traitées.

Néanmoins, dans les expériences de longue durée, on constate souvent au bout d'une heure $\frac{1}{2}$ à 2 h d'action de la RNase, une augmentation appréciable de la consommation d'oxygène (50 à 90 %), qui devient légèrement inférieure à la moyenne une heure plus tard.

Conclusions. Sans exercer d'effet massif, la RNase semble provoquer, au bout d'une heure, une augmentation sensible de la teneur en ATP, suivie d'une légère baisse. Au bout d'1 h $\frac{1}{2}$ à 2 h la consommation d'oxygène augmente aussi momentanément, pour diminuer ensuite. Ces résultats montrent que, comme dans le cas des racines d'oignon étudiées par BRACHET⁴, la RNase ne modifie pas profondément les mécanismes producteurs d'énergie dans les cellules traitées.

TABLEAU II
CONSOMMATION D'O₂ PAR AMIBE EN 10⁻³ μ l/h

Exp.	Amibes témoins	Amibes traitées
I	0.23	0.27
II	0.27	0.28
III	0.28	0.25
IV	0.28	0.27
V	0.26	0.24
VI	0.23	0.43
Moyenne	0.25	0.29

Y. ŠKREB-GUILCHER*

Laboratoire de Morphologie animale, Faculté des Sciences de
l'Université libre de Bruxelles (Belgique)

¹ J. BRACHET, *Nature*, 168 (1951) 205.

² J. BRACHET, *Nature*, 175 (1955) 851.

³ J. BRACHET, *Biochim. Biophys. Acta*, (sous presse).

⁴ J. BRACHET, *Nature*, 174 (1954) 876.

⁵ M. KUNITZ, *J. Biol. Chem.*, 164 (1946) 563.

⁶ K. LINDERSTRØM-LANG ET H. HOLTER, *Compt. rend. trav. lab. Carlsberg*, 24 (1943) 334.

⁷ B. L. STREHLER ET J. R. TOTTER, *Arch. Biochem. Biophys.*, 40 (1952) 28.

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* Boursière des accords culturels franco-belges. Adresse permanente: Institut de Zoologie, Université de Zagreb, Yougoslavie.

Blood group substance activity of intrinsic factor preparations

SPRINGER, ROSE AND GYÖRGY¹ have found that high blood group A, B and H substance activities were present in all of 8 highly purified intrinsic factor preparations. Furthermore, LATNER, MERRILLS AND RAINE² have reported that their intrinsic factor preparation possessed well-marked blood group substance activity, although ultracentrifuge experiments did not reveal the presence of the usual blood group substances.

In view of these facts we decided to test some of our intrinsic factor preparations for possible A, B and H antigen activity and to compare this with the clinical activity.

Intrinsic factor activity was ascertained by daily oral administration of the preparation together with cyanocobalamin (5–10 micrograms, mostly bound) to patients with pernicious anemia in relapse with an initial blood count of less than 2 millions of erythrocytes per mm³. Blood counts and hemoglobin determinations were performed before and at least on the first, seventh, fourteenth and twenty first day of the treatment. Reticulocytes were counted daily. The preparations were considered as fully active if they gave an increase of at least 1.2 millions of erythrocytes per mm³ in 21 days, as moderately active with an increase of 0.6 million–1.2 millions, and as inactive with an increase of less than 0.6 million. Although an occasional patient with pernicious anemia may show a slight or transient hematopoietic response following the daily oral administration of 5–10 micrograms of vitamin B₁₂ without added intrinsic factor, we did not regularly administer this dose of vitamin B₁₂ as an oral control therapy before the test. We believe that the remote possibility that some response

might occur following such small daily doses of vitamin B₁₂ does not justify a prolongation of the test period in every case.

The serological activity was determined with the agglutination inhibition test of MORGAN AND VAN HEININGEN³. The anti A serum had a titer of 1:64 towards A₂ erythrocytes and the anti H serum of 1:8 towards O erythrocytes. Both antisera were of human origin. The results of the determinations of the A and H antigen activity given in Table I are expressed in terms of the reciprocal inhibition titer. None of the preparations showed any reaction in the test for B antigen activity.

TABLE I
RESULTS OF THE DETERMINATIONS OF BLOOD GROUP SUBSTANCE ACTIVITY
IN INTRINSIC FACTOR PREPARATIONS

Preparation	Clinical tests		Blood group substance activity* (reciprocal inhibition titer)			
	Daily dose** (mg)	Activity	A		H	
			per mg	per daily dose	per mg	per daily dose
I.F. 656 A	4.8	inactive	1000	4800	10	48
I.F. 860 A	—	inactive***	1000	—	0.2	—
I.F. 922	5.0	inactive	1000	5000	20	100
I.F. 1187	1.0	moderately active	1000	1000	5	5
I.F. 1344	1.0	moderately active	2000	2000	75	75
I.F. 550	2.7	active	1000	13000	75	950
I.F. 762	2.0	active	1000	2000	10	20
I.F. 1342	1.0	active	1000	1000	75	75
I.F. 1345	1.0	active	1000	1000	35	35

* Blood group B substance activity could not be demonstrated in any of the preparations.

** Active preparations were given in a daily dose of 1 or 2 oral U.S.P. units. Inactive preparations represented an equivalent amount of starting material (hog pyloric mucosa) except for I.F. 922, where a much higher dose was given.

*** I.F. 860 A has not been tested clinically but would certainly have been inactive because afterwards the starting material was found to be inactive.

As may be seen from the table the A antigen activity per mg substance is practically the same for clinically active and inactive preparations. Per daily dose the clinically inactive preparations I.F. 656 A and I.F. 922 show an even higher titer than the clinically active I.F. 762, I.F. 1342 and I.F. 1345, whereas the most impure preparation I.F. 550 has the highest A antigen activity. For the H antigen activity per mg substance more differentiation was found. Here, too, the titer per clinical daily dose of the inactive preparation I.F. 922 is much higher than that of the fully active I.F. 762, and again the highest antigen activity is shown by the crude preparation I.F. 550.

From these data it can be concluded that there is no correlation between intrinsic factor activity and the blood group A or H substance activity. Therefore, at least a great deal of the blood group A and H substance activities in our intrinsic factor preparations must result from contaminants. However, we do not exclude the possibility that intrinsic factor itself might show some blood group A or H substance activity, since none of the clinically active intrinsic factor preparations tested so far by us, by SPRINGER *et al.*¹ or by LATNER *et al.*², was free from serological activity.

Our preparations did not show any blood group B substance activity and this proves that no relationship at all exists between intrinsic factor activity (from hog mucosa) and blood group B substance activity.

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Research Laboratories, N.V. Organon, Oss (Netherlands)

Blood Transfusion Service of the Netherlands Red Cross, Central Laboratory,
Amsterdam (Netherlands)

JAN P. W. VAN BAAL

HARRY G. WIJMEGA

MIA VAN DER HART

¹ G. F. SPRINGER, C. S. ROSE AND P. GYÖRGY, *J. Lab. Clin. Med.*, 43 (1954) 532.

² A. L. LATNER, R. J. MERRILLS AND L. C. D. P. RAINE, *Biochem. J.*, 57 (1954), Proc. xix.

³ W. T. J. MORGAN AND R. VAN HEININGEN, *Brit. J. Exp. Pathol.*, 25 (1944) 5.

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