Cardiac Glucosides and Lundsgaard Contracture

The striped muscle of vertebrates poisoned with monohalogeno acetate shows *when stimulated* a progressive, irreversible contracture and inexcitability. This contracture, the Lundsgaard phenomenon, is associated with the inhibition of carbohydrate metabolism. All the substances which react with the –SH groups of cysteine, glutathione, and proteins (i. e. "*thioloprive*" substances[1]), when applied to isolated amphibian muscle, give the same contracture curve when the muscle is stimulated: for instance, heavy metal ions, arsenic compounds[2], vesicants and lacrymators, maleic acid (but not fumaric) and oxidizing substances including quinones and peroxides.

Further work has shown that the Lundsgaard contracture is the physiological translation of any biochemical lesion of carbohydrate metabolism[3].

At first sight, nothing is closer to the Lundsgaard phenomenon than the contracture of the frog's heart poisoned with a cardiac glucoside, and this similarity is not accidental[1].

A series of papers of KRAYER, R. MENDEZ, and others[4] have shown the following facts:– (1) substances (synthetic or natural) containing the unsaturated lactone ring, which in the molecule of cardiac glucosides is essential for digitalis-like action, cause a progressive ventricular contracture of the isolated frog's heart; (2) ascorbic acid, which can be considered as an unsaturated lactone, has a digitalis-like action on the frog's heart, and this action is due to hydrogen peroxide formed during the oxidation of ascorbic acid; (3) besides H_2O_2 (a confirmation of RICHTER'S[5] observations), various sulfhydryl reagents applied to the frog's heart lead to progressive contracture and inexcitability.

Recent work in my laboratory has proved that isolated amphibian striped muscle, when poisoned with digitoxin, shows an incomplete contracture when periodically excited with KCl; but we have not observed a reaction *in vitro* between cardiac glucosides and sulfhydryl substances in an atmosphere of nitrogen.

Many substances have the same general effect (with quantitative differences) on striped and cardiac muscle; the digitalis ventricular contraction of the frog's heart may be considered as a particular case of the Lundsgaard contracture. The biochemical background of the Lundsgaard contracture is an inhibition of glycolysis by blocking of the SH enzymes; lactic acid is not produced; the phosphagen stores are rapidly exhausted.

Several investigations by various german authors show that cardiac glucosides inhibit glycolysis *in vitro*:– (1) strophanthin 0·001 to 0·1% decreases oxygen consumption and CO_2 production of mouse liver and guinea-pig's brain; anaerobic glycolysis is inhibited by strophanthin concentration as low as 0·001%[6]; (2) according to HAARMANN[7] and TOBBEN[8], lactic acid formation in hearts poisoned with strophanthin is nearly completely inhibited, although it might be increased when small concentrations of strophanthin are used. Further discussion of these facts is given by LENDLE[1]. In this field of biochemical action of cardiac glucosides, much more information is needed before reaching a definite conclusion; all that can be said at present is that already a few data are available by favour of the hypothesis which assimilates cardiac glucoside contracture to the Lundsgaard phenomenon on striped muscle.

Quite naturally, many arguments can be raised against this idea; conflicting observations are numerous. We agree with MENDEZ[2] when he stresses the point that therapeutic and toxic actions are very different phenomena in digitalis action; but we like to give a final clinical argument.

The typical disease where inhibition of carbohydrate metabolism (at the level of pyruvic acid) plays the dominant role is beriberi. Sudden death from cardiac failure and vasomotor collapse is common in human severe thiamin deficiency; it is usual in criminal digitalis intoxications. The T wave of the electrocardiogram is typically flattened or inverted in beriberi and in digitalis poisoning; this anomaly disappears when both conditions are cured; failure of the beriberi heart to respond to digitalis therapy has been noted, and considered as a criterium[3].

Thus, if one considers the pharmacological evidence, the clinical data and the little we know about the biochemical action of cardiac glucosides, one gathers without difficulty the elements of a sound working hypothesis which may be formulated as follows: the action of cardiac glucosides on the heart is the consequence of a discrete inhibition of carbohydrate metabolism, presumably by the indirect blocking of some –SH enzyme (or enzymes).

Z. M. BACQ

Laboratoire de pathologie et thérapeutique générales, Université de Liège, Belgique, le 8 janvier 1949.

Résumé

De nombreux arguments d'ordre pharmacologique, en accord avec quelques expériences biochimiques et des observations cliniques, montrent que la contracture cardiaque provoquée par les corps digitaliques peut-être assimilée à la contracture de Lundsgaard du muscle strié. Une hypothèse de travail est proposée, selon laquelle l'action des glucosides digitaliques sur le cœur est la conséquence d'une inhibition partielle du métabolisme des hydrates de carbone.

[1] Z. M. BACQ, Exper. *2*, 349, 385 (1946).

[2] J. M. GERNAY and J. LECOMTE, Arch. Int. Pharm. Thér., *77*, 318 (1948).

[3] J. LECOMTE, M. GOFFART, and Z. M. BACQ, Arch. Int. Physiol., *51*, 63 (1948).

[4] O. KRAYER, R. MENDEZ, E. MOISSET DE ESPANES, and R. P. LINDSTEAD, J. Pharm. Exper. Ther., *74*, 372 (1942). – O. KRAYER, R. P. LINDSTEAD, and D. TODD, ibid., *77*, 113 (1943). – R. MENDEZ, ibid. *81*, 151 (1944). – R. P. LINDSTEAD and O. KRAYER, Science *95*, 332 (1942).

[5] H. RICHTER, Arch. Exper. Path. Pharmak. *197*, 137 (1941).

[6] H. WEESE and CH. WIEGAND, Medizin and Chemie *2*, 148 (I. G. Farben, 1934).

[7] W. HAARMANN, Bioch. Z. *255*, 103, 142 (1932).

[8] H. TOBBEN, *Thesis* (Münster 1932).

[1] L. LENDLE, Heffter Handbuch Exper. Pharm., Ergänzungswerk *1*, 207 (1935).

[2] R. MENDEZ, Archives Inst. Card. Mexico *17*, 83 (1947).

[3] E. S. GORDON, *Nutritional and Vitamin Therapy* (The Yearbook Publishers, Chicago, 1947).

Protecting Effect of Heparin on the Oxidation of Thrombin[1]

There can be found many statements in the widespread literature on heparin about its antithrombin–antiprothrombin–and antikinase effect.

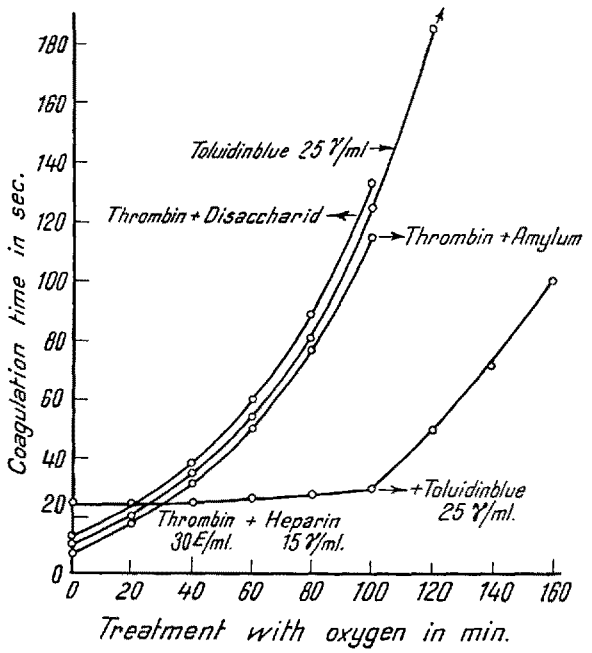
[1] With the help of the «Roche»-Studienstiftung.

The present work demonstrates a property of heparin, unknown till now, from which one could conclude–under certain circumstances–on a thrombin-protecting effect of the heparin.

In a previous work[1] it was reported, that thrombin is oxidizable and that it loses its coagulation activity in this oxidized form. In the actual work the oxidation of thrombin could be reached by streaming it through with molecular oxygen, as our starting material (thrombin, Hoffmann - La Roche) contains some unknown compound which activates molecular oxygen. This proceeding can be shown in a typical curve (see Figure). It is evident from this curve that thrombin loses its activity to coagulate plasma (0·2% sodium oxalate) or fibrinogen (0·6%) gradually and finally gets inactive totally, when it is streamed through with oxygen.

If heparin up to a concentration of 15 γ/ml is added to a solution of thrombin the coagulation time against plasma increases from 10 to 26 seconds. When, on the other hand, the same heparin thrombin mixture and the solution of thrombin whithout heparin are streamed through with oxygen at the same time, so the heparin-free solution of thrombin only loses its coagulation activity, meanwhile the mixture of thrombin and heparin keeps its original coagulation activity completely.

Therefore heparin protects the thrombin from oxidation by oxygen.



As the heparin is an esterified polysaccharide (Liquemin, Hoffmann-La Roche ester of polysulfuric acid of mucoitin), the experiment was repeated with other non-esterified saccharides. It was shown that neither disaccharides nor the polysaccharide starch has the oxidation-inhibiting quality of heparin.

The specific activity of heparin is proved by the fact that, if the heparin in the mixture of thrombin and heparin is combined with toluidine blue (25 γ/ml) the activity of heparin is excluded and an oxidation resp. inactivation of thrombin takes place (see Figure).

[1] L. A. PÁLOS, Nature, in print.

Experiments on the behaviour of heparin during oxidation of the plasma and the other coagulation factors, are in process.

The acting mechanism of heparin in this reaction is not known and should be further investigated.

L. A. PÁLOS

Institute for Hygiene and Bacteriology of the University of Basle, Switzerland, December 30, 1948.

Zusammenfassung

Das Thrombin ist oxydierbar und wird in seiner oxydierten Form inaktiv. Die Oxydation des Thrombins kann durch sehr kleine Mengen Heparin gehemmt werden. Wenn das Heparin mit Toluidinblau gebunden wird, kann die Oxydation vor sich gehen.

Effect of Thiamin, Riboflavin, Pyridoxin, and Nicotinic Acid on the Intestinal Absorption of Dextrose

In earlier experiments[1] I found that after the administration of unphysiologically large doses of a vitamin used in the metabolism in phosphorylated form, the apparatus of the phosphorylation becomes exhausted. I further found[2] that the absorption of one vitamin—in my experiments that of thiamin—from the intestine was greatly impaired, when other vitamins of the B complex were given in large doses before the experiment. This fact could also be demonstrated by using dextrose instead of the vitamins. So I could explain the cases of the so-called vitamin imbalance, i. e. the deficiency symptoms which became manifest in a deficiency state during the treatment with large doses of a single vitamin, symptoms of pellagra, for instance, after administration of riboflavin, or symptoms of polyneuritis during the treatment with nicotinic acid. The high unphysiological dosis of a vitamin gives rise to the exhaustion of the phosphorylation apparatus and other vitamins cannot be utilized.

To get further evidences of my hypothesis, I examined the absorption of dextrose–a substance which must be phosphorylated too when absorbed and utilized–from the intestine of rats, after having administered large doses of the B-complex vitamins.

My method was that of LASZT's procedure[3] in which 200 mg of dextrose in 2 cc phys. sodium chloride solution were given into a 30 cm long tied bowel section of narcotized rats and after 30 minutes the unabsorbed quantity of the dextrose was determined. Details of the method are given elsewhere[2].

The results of the control experiments were the following:

Table I

No.	mg dextrose found after 30 min.	mg dextrose absorbed	absorbed dextrose %
1	120	80	40
2	137	63	31
3	112	88	44
4	121	79	38
5	109	91	45
6	124	76	38

[1] I. MAGYAR, Acta medica Hungarica *1*, 46 (1948).
[2] I. MAGYAR, Int. Z. Vitaminforsch., in print.
[3] L. LASZT and L. DALLA TORRE, Schweiz. med. Wschr. *22*, 1416 (1941).