The order of values obtained is in accordance with the new results³ for the heat of hydrolysis of adenosine triphosphate, which is given as -4.7 kcal. per mole, instead of the previously accepted value of -12 kcal. per mole. It is nearly equal to the heat of hydrolysis of pyrophosphate and trimetaphosphate (-5.8 and -6.2 kcal. per mole of phosphate anhydride bond).

Under physiological conditions, the phosphate bond-energy of adenosine triphosphate will change, depending on concentrations of adenosine tri- and di-phosphate and inorganic phosphate, and will be considerably higher. Thus, if we accept the concentration of phosphoric acid as $0.01 M$ (which is usually the case in tissues) and that of adenosine triphosphate as the same as that of the diphosphate, it would be -8.4 kcal.

Hence, in spite of the lower free-energy of adenosine triphosphate hydrolysis under standard conditions, this compound does not lose its biological significance as a universal accumulator of chemical energy.

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Blood Anticoagulant Activity of Sulphated Ovular Mucins of Amphibians

THE mucin constituting the gelatinous layer which envelops the eggs of *Bufo bufo*, if digested with papain and then sulphated, shows a marked anticoagulant activity on the fibrinogen and the whole blood plasma¹. A similar anticoagulant activity has now been shown by the ovular mucins of other species of amphibians, sulphated without any previous proteolysis.

The ovular mucins of *Discoglossus pictus*, axolotl and *Bufo bufo*, removed from the eggs either by hand or by treatment with sodium thioglycolate², were lyophilized, desiccated *in vacuo* over phosphorus pentoxide, and sulphated with chlorosulphonic acid (0.7 ml. per gm. of dry substance) for 12 hr. at $-16^{\circ}C$. The sulphated derivatives were neutralized, dialysed and filtered; no further purification was attempted.

The anticoagulant activity has been determined by the prolongation of the coagulation times of fibrinogen added to thrombin, and of oxalated bovine plasma after recalcification. The determinations were carried out as follows.

(1) *Fibrinogen*. 0.1 ml. of sulphated mucin solution was mixed with 0.5 ml. of a 0.5 per cent solution of fibrinogen (Bovine Plasma Fraction I, Armour Co.) in saline, and 0.1 ml. of a 0.1 per cent solution of Trombasi Roussel (a powder extracted from the blood plasma, and containing thrombin) was added. The coagulation times at $37^{\circ}C$. were determined, as

Table 1

	Concentration ($\mu\text{gm./ml.}$)	Coagulation times	
		Fibrinogen	Plasma
Controls		4 min. 34 sec.	7 min. 10 sec.
Heparin	1	6 min. 18 sec.	10 min. 35 sec.
<i>Discoglossus</i>	2.5	6 min. 20 sec.	—
	60	—	10 min. 28 sec.
Axolotl	9	6 min. 10 sec.	—
	100	—	10 min. 30 sec.
<i>Bufo bufo</i>	2.5	6 min. 20 sec.	—
	25	—	10 min. 22 sec.

well as those of the controls (0.1 ml. of distilled water instead of the mucin) and those given in the presence of a 0.0001 per cent solution of standard heparin (100 units per mgm.).

(2) *Whole plasma*. 0.5 ml. of oxalated bovine plasma (9 parts of blood and 1 part of $0.1 M$ neutral potassium oxalate) was recalcified with 0.1 ml. of 2 per cent calcium chloride, and mixed with 0.1 ml. of sulphated mucin solution, or distilled water, or 0.0001 per cent standard heparin solution. The coagulation times were determined at room temperature (about $18^{\circ}C$.).

The results are summarized in Table 1, which only gives the results for concentrations of mucins showing approximately the same anticoagulant activity as 0.0001 per cent heparin, that is, prolonging the coagulation time of fibrinogen by about 40 per cent, and that of plasma by about 50 per cent. The results in Table 1 are the averages of sixteen determinations for fibrinogen, and of nine for the plasma.

From these results, the anticoagulant activities of the sulphated mucins could be expressed in heparin units as in Table 2.

Table 2. ANTICOAGULANT ACTIVITY IN HEPARIN UNITS

Mucin	Fibrinogen	Plasma
<i>Discoglossus</i>	40	1.5
Axolotl	10	1
<i>Bufo bufo</i>	40	4

The anticoagulant activity induced by sulphation is probably due to the sulphamic groups arising in the esterification with sulphuric acid of the hexoamines present in the mucin substance¹. The much lower activity for the whole plasma than that for fibrinogen, in respect to the heparin, may depend on the inability of the sulphated mucins to combine with the plasma fractions which activate heparin.

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Antigenicity of Antibodies of the Same Species

IN previous papers¹ rare human sera were described which possess the ability to agglutinate human red cells sensitized with incomplete isoantibodies. The possible application of such sera in blood group laboratories was stressed. Based on the absorption experiments, the opinion was expressed that the agglutinating factor contained in these sera is an 'anti-antibody', which reacts with human immune globulins denatured in serological reaction.