of hydrogen peroxide from a 0.01 molar solution of I in 1/15 molar phosphate buffer of pH 7 is presented as a function of time. The titanous sulphate method[10] was used for the determination of hydrogen peroxide. After 140 h about 78% of the theoretical amount of hydrogen peroxide is formed. It is supposed that the reaction follows a similar scheme as the autoxidation of hydrazobenzene[11].

The same pattern of behaviour as I both with respect to the degradation of DNA and to the formation of hydrogen peroxide is observed with 1-methyl-2-benzyl-hydrazine phosphate and with 1-methyl-2-*p*-allophanoyl-benzyl-hydrazine hydrobromide (II[12]). All these compounds have in common that they autoxidize rather slowly. It may be assumed that *slow* release of hydrogen peroxide is an essential requirement for cytotoxic activity.

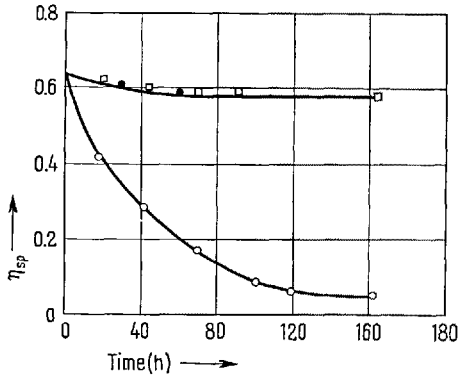

Fig. 1. Change of specific viscosity of a 0.0005 molar aqueous solution of I containing 0.07% w/v of DNA.
○ in air; ● in nitrogen; □ in air, solution contains catalase

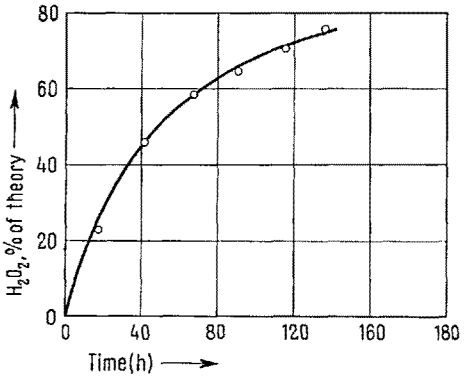

Fig. 2. Formation of hydrogen peroxide in a 0.01 molar aqueous solution of I in the presence of air.

From the experimental results it is concluded that the effect on the viscosity of aqueous DNA solutions of the above mentioned methylhydrazine derivatives is due to autoxidation of the latter compounds, leading to the formation of hydrogen peroxide. It is generally accepted that the action of hydrogen peroxide on DNA proceeds via OH radicals[8]. Therefore, an analogy of the effect of the methylhydrazine derivatives on DNA with the indirect effect of ionizing radiation is evident as the latter is assumed to be due mainly to the action of OH radicals[13]. The question remains open whether the inhibition of tumour growth depends on an effect on preformed DNA, on the synthesis of DNA, or on other biochemical effects of the hydrogen peroxide, like the inhibition of glycolysis[14].

Zusammenfassung. Methylhydrazinderivate wie 1-Methyl-2-*p*-(isopropylcarbamoyl) benzyl-hydrazin-hydrochlorid und 1-Methyl-2-*p*-allophanoylbenzyl-hydrazin-hydrobromid bewirken unter aeroben Bedingungen einen Viskositätsabfall der wässerigen Lösungen von Desoxyribonucleinsäure. Es wird gezeigt, dass dieser Effekt auf die Bildung von Wasserstoffperoxyd bei der Autoxydation der Methylhydrazinverbindungen zurückzuführen ist. Auf die Analogie zum indirekten Effekt von Röntgenstrahlen wird hingewiesen.

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Preliminary Studies on the Hemostatic Activity of the Isonicotinyl Hydrazone of Acetaldehyde

Introduction. The isonicotinyl hydrazone of acetaldehyde (IHA) was studied by us as a possible metabolite of isonicotinic acid hydrazide[1], arising from the decarboxilation of the isonicotinyl hydrazone of pyruvic acid[2]. Studies on the toxicity and bacteriostatic activity of the said hydrazones[3] led to the finding of a pronounced hemostatic activity of the acetaldehyde derivative, and these are the results we wish to describe.

Materials and Methods. IHA was prepared by dissolving isonicotinic acid hydrazide in commercial acetaldehyde, evaporating the excess aldehyde and precipitating the hydrazone by the addition of water. The precipitate was washed with water and with ethanol, being then dried under an infrared lamp. The purity of the product was controlled by paper chromatography[1,4] and its composition was confirmed by acid hydrolysis (HCl) and paper chromatography[1,4,5]. IHA is a yellowish powder, insoluble in

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water, alcohol, ether and all the usual organic solvents, soluble in alkaline media (aqueous) and in alkaline organic solvents, such as pyridine and liquid amines. Acute toxicity (LD_{50}) for mice was found to be 800 μg/g.

Coagulation times were measured by the usual method of Sabrazes (capillary tube), using blood collected from the marginal vein of the ear of rabbits, before and at different times after the intravenous injection of slightly alkaline (pH 7.5) aqueous solutions of the hydrazone. Control animals were injected with the same volumes of alkaline solutions of identical pH and bled simultaneously.

Results and Discussion. Upon the examination of more than 60 rabbits, we found the normal coagulation time to lie between 30 sec and 1 min. About 20% of the animals had coagulation times as high as 4 min, though being apparently healthy, and among them we selected 12 of the ones presenting the highest values. Two of them we kept as controls, and the rest was injected with different amounts of the hydrazone.

The coagulation time was measured every 30 min during 3 h, and typical examples of the results so obtained are shown in the Figure, where we compare the values found for the controls with the ones of 2 rabbits injected with 5 mg/kg IHA.

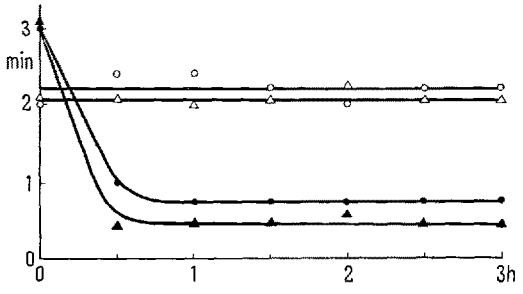
As can be seen, under these conditions the coagulation time drops at a very fast rate during the first 30 min after the injection of the drug, till the lower normal values are reached. These lower values were found to be stable during all the period of observation, the rate of that decrease being roughly proportional to the amount of drug injected.

The Table shows the initial values found for the coagulation times of all the animals used in the present experiment, as well as the lower ones found after the injection of different amounts of the drug.

No explanation of the hemostatic activity of IHA can be offered at the present moment, though the possibility exists of its being due to the interference of the drug on the acetylation of thrombin, leading to a higher clotting power[6].

Coagulation times before and after the intravenous injection of different doses of IHA

Animal No.	Coagulation time (min) initial	final	Dose mg/kg
1	2:45	1:05	2
2	4:00	1:30	2
3	2:50	0:40	4
4	2:50	1:20	4
5	2:35	0:30	4
6	3:00	1:00	5
7	3:00	0:45	5
8	2:30	0:30	5
9	3:05	0:25	10
10	3:00	0:30	10



Coagulation times of two rabbits injected intravenously with 5 mg/kg of IHA (black marks) compared with the values found for two untreated animals (empty marks).

Résumé. Les auteurs ont étudié l'activité hémostatique de l'isonicotinyl hydrazone de l'aldéhyde acétique, utilisant des lapins normaux à temps de coagulation sanguine relativement long (2–4 min). L'injection intraveineuse de 5 mg/kg de la drogue cause une chute très rapide du temps de coagulation vers les valeurs normales (0,5–1 min), valeurs qu'ils ont trouvées constantes pendant toute la période d'observation (3 h).

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Selective Inhibition of the Multiplication of Phage T1 in *E. coli* K12

During a screening of antibiotics from Actinomyces, we isolated a product which is chemically correlated with Netropsin[1] and with Congocidin[2]. In previous communications[3,4] we reported some preliminary data about the activity of the antibiotic named by us Distamycin A on experimental tumours.

This compound (Distamycin A)[5] shows the interesting ability to inhibit the multiplication of phage T1 in *E. coli* K12 in a selective way, at concentrations quite harmless to the growth of the host microorganism. From tests carried out on solid medium, determining the number of plaque-forming units present in a preparation of phage T1, it appeared that the inhibiting activity of the product was proportional to the dose. In fact, in the presence of this compound, the number of plaque-forming units decreased

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