

Allerdings liegen die absoluten Werte für die einzelnen Ionenkonzentrationen bei ZIEGLER erheblich unter den hier mitgeteilten Zahlen, was möglicherweise durch die verschiedenartigen Objekte bedingt ist. Für das starke Überwiegen des Kaliums im Siebröhrensaft liegt noch keine Erklärung vor. Die starke Anreicherung des Magnesiums im Bereich des Phloems⁷ und auch im Siebröhrensaft dürfte mit der besonderen Rolle zusammenhängen, welche dieses Ion bei der Aktivierung von Phosphorylierungsreaktionen und damit indirekt beim Stofftransport im Phloembereich spielt. Interessant ist, dass sowohl Kalium als auch Magnesium, nicht dagegen Natrium und Calcium für Aphiden im besonderen Masse für eine normale Entwicklung notwendig sind⁸. An Anionen kommt ausser Spuren von Chloriden lediglich Phosphat im Honigtau vor, was auch für den Siebröhrensaft zutreffen muss. Dies bestätigt in Übereinstimmung mit den Ergebnissen von ZIEGLER⁷, dass Schwefel und Stickstoff im

Siebröhrensaft ausschliesslich in organischer Form transportiert werden.

Summary. In the honeydew of *Megoura viciae* BUCKT., sucking on the sieve tube sap of *Vicia faba*, the following cations are present in measurable quantities: potassium (13.0–14.1 mg/ml), sodium (0.04–0.051 mg/ml), magnesium (1.8–2.3 mg/ml) and calcium (0.07–0.09 mg/ml). There are only traces of copper, iron, manganese, zinc, cobalt and molybdenum. Amongst the anions, phosphate was found at 1.9–2.5 and chloride at 0.02–0.05 mg/ml, whereas nitrate and sulphate could not be detected.

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The Effect of 7,12-Dimethylbenz(a)anthracene on the Incorporation of Thymidine- H^3 into Deoxyribonucleic Acid in Normal and Regenerating Liver

The administration of DMBA (7,12-dimethylbenz(a)-anthracene) has been reported to depress the incorporation of tritium-labelled thymidine into DNA in rat testis, intestine, and adrenals¹. A single injection of DMBA into male rats has also been reported to inflict selective damage in the testicular cells which actively synthesize DNA². In the present investigation, the incorporation of thymidine- H^3 into DNA in normal or regenerating liver of rats treated with DMBA has been considered.

In the first experiment, male rats (80–100 g) were injected intraperitoneally with DMBA (Fluka AG) (5 mg or 10 mg in 0.5 ml seed oil/100 g); at different times after injection tritium-labelled thymidine (Schwarz Bio-research Inc.) was given intramuscularly twice (2.5 μ C/100 g each time) at 3 h intervals. 3 h after the last injection, the rats were sacrificed and the livers were removed. In the second experiment, male rats (250 g) were partially hepatectomized³, injected intraperitoneally 12 h later with DMBA (5 mg in 0.5 ml seed oil/100 g) and 33 h later with thymidine (5 μ C/100 g). After 3 h (36 h after hepatectomy) the rats were sacrificed and the livers removed. The controls in both experiments received the same treatment, but were injected with oil alone.

DNA extraction was carried out on nuclear fraction following the method of SCHNEIDER et al.⁴. The extracted DNA was dissolved in 0.1 N NH_4OH : an aliquot was used for DNA estimation by diphenylamine reaction⁵, and another one (containing always the same amount of DNA as calculated according to the Dische reaction) was plated on an aluminum disc; after drying, the plated sample was counted in an internal gas flow counter.

Table I shows a noticeable inhibition of thymidine incorporation into liver DNA of growing rats after 24 and 48 h from DMBA injection; after 5 and 7 days an increased incorporation is observed. In other experiments in which double amount (10 mg/100 g) of DMBA was injected, the results were similar to those reported in

Table I, but a high mortality was noted after the third day.

Table II shows a marked decrease in incorporation of thymidine into DNA of regenerating liver in DMBA treated rats, as compared to controls.

From the above experiments, the results of JENSEN et al.¹ can be extended to growing and regenerating liver. Probably the effect of DMBA on DNA synthesis is not due to a necrotic action of DMBA on liver cells. In fact, during the two days following injection of DMBA in the

Table I. Specific activity of DNA (as c.p.m./mg) extracted from the liver of young rats

Time after DMBA injection, in days	1	2	3	5	7
No. of rats	5 (C) 4 (T)	3 (C) 3 (T)	4 (C) 3 (T)	4 (C) 4 (T)	3 (C) 4 (T)
Mean value of control animals	357	351	358	392	341
Mean value of treated animals	245	227	361	643	693
% difference	69	65	100	164	203

Control animals (C): injected with oil only. Treated animals (T): injected with DMBA (5 mg/100 g). The % differences are presented taking the control group as 100. The differences at 1, 2, 5 and 7 days are significant.

¹ E. V. JENSEN, E. FORD, and C. HUGGINS, *Proc. nat. Acad. Sci.* 50, 454 (1963).

² E. FORD and C. HUGGINS, *J. exp. Med.* 118, 27 (1963).

³ G. M. HIGGINS and R. M. ANDERSON, *A.M.A. Arch. Path.* 12, 186 (1931).

⁴ J. H. SCHNEIDER, R. CASSIR, and F. CHORDIKIAN, *J. biol. Chem.* 235, 1437 (1960).

⁵ Z. DISCHE, *Mikrochemie* 8, 4 (1930).

first experiment, when the inhibitory effect is evident, no histologically demonstrable necrotic lesions have been observed; furthermore, during a severe experimental necrosis, even quite early, an increase in thymidine incorporation into DNA has been noted⁶. The data of JENSEN et al.¹ and the present results can be related to the early observations of HADDOW⁷ on inhibition exerted by carcinogenic hydrocarbons on growth of normal and neoplastic tissues. The mechanism of this effect is not yet clear. Some recent data can be quoted as providing some work hypotheses. BOYLAND and GREEN⁸ and LIQUORI et al.⁹ have reported the solubilization of benzo(a)pyrene and related compounds by dilute solution of DNA, and

considered their results as providing evidence for the intercalation of the planar hydrocarbons between the base pairs of DNA; GIOVANELLA et al.¹⁰ consider these data as the result of a non-specific adsorption. An in vivo binding of 1, 2, 5, 6-dibenzanthracene to mouse skin DNA was reported by HEIDELBERGER and DAVENPORT¹¹, but subsequently MCKINNEY and HEIDELBERGER (unpublished data, quoted in ¹⁰) did not confirm this result.

Riassunto. L'idrocarburo cancerogeno 7,12-dimetilbenz(a)antracene somministrato con iniezione endoperitoneale inibisce l'incorporazione della timidina nel DNA del fegato normale e del fegato rigenerante di ratto.

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Table II. Specific activity of DNA (as c.p.m./mg) of regenerating rat liver

	No. of rats	Specific activity	% difference
Mean value of control animals	4	4375	100
Mean value of treated animals	5	1767	40

See Table I. The difference is highly significant.

⁶ M. C. LEEVY, *Ann. N.Y. Acad. Sci.* 104, 939 (1963).

⁷ A. HADDOW, *Nature* 136, 868 (1935).

⁸ E. BOYLAND and B. GREEN, *Br. J. Cancer* 16, 507 (1962).

⁹ A. M. LIQUORI, B. DE LERMA, F. ASCOLI, C. BOTRÉ, and M. TRACIATTI, *J. molec. Biol.* 5, 521 (1962).

¹⁰ B. C. GIOVANELLA, L. E. MCKINNEY, and C. HEIDELBERGER, *J. molec. Biol.* 8, 20 (1964).

¹¹ C. HEIDELBERGER and G. R. DAVENPORT, *Acta Un. int. Cancr.* 17, 55 (1961).

Effect of Synthetic Vasopressins on the Myocardial Contractile Force in the Isolated Guinea-Pig Atria

It has been known for some years that pitressin constricts coronary arteries and decreases coronary blood flow in dogs¹⁻³. Very recently, WANG⁴ observed that synthetic 2-phenylalanine-8-lysine vasopressin (PLV-2) decreased coronary arterial blood flow, thereby reducing myocardial contractility in dogs. However, it remains uncertain whether synthetic vasopressin per se would possess the direct negative inotropic action of the heart. The present study was undertaken to investigate the comparative effect of three synthetic vasopressins, i.e. 8-arginine vasopressin, 8-lysine vasopressin and 2-phenylalanine-8-lysine vasopressin, on the myocardial contractility in the isolated guinea-pig atria.

Guinea-pigs of either sex weighing approximately 500 g were killed by cervical dislocation. Their hearts were removed immediately and the atria excised. The atria were washed twice with and suspended in a bath containing oxygenated Ringer-Locke solution (32°C) through which 95% O₂ and 5% CO₂ (pH = 7.35) was bubbled⁵. The frequency of atrial contraction was kept constant at a rate of 90/min using a Grass stimulator (Model S4). The force of atrial contraction was measured and recorded continuously using a Grass force-displacement transducer (FT-03) and a Grass polygraph (Model 5), respectively. 45 min after the preparation was completed, the effect of the three vasopressins in concentrations between 10 and

320 mU/ml on the myocardial contractile force was examined.

The results of the effect of the three vasopressins in the isolated guinea-pig atria were consistent in all experiments and are summarized in Figure 1. Tracings of representative experiments are illustrated in Figure 2. It was found that concentrations of less than 10 mU/ml of the three synthetic vasopressins in the isolated bath caused essentially no change in the myocardial contractile force. However, higher concentrations (more than 20 mU/ml) of the vasopressins decreased contractile force in proportion to the concentration. In no instance was any increase in the force of myocardial contraction observed. There was no essential difference in the magnitude of the decrease in contractile force between 8-arginine vasopressin and 2-phenylalanine-8-lysine vasopressin. However, the decrement in contractile force by these two vasopressins was slightly greater than that by 8-lysine vasopressin.

From the present study, it is evident that small doses of the three synthetic vasopressins did not show any

¹ E. BULBRING, J. H. BURN, and J. M. WALKER, *Quart. J. Med.* 18, 73 (1949).

² H. E. ESSEX, R. G. E. WEGRIA, J. F. HERRICK, and F. C. MANN, *Am. Heart J.* 19, 554 (1940).

³ H. D. GREEN, R. WEGRIA, and N. H. BOYLE, *J. Pharmacol. exp. Therap.* 76, 378 (1942).

⁴ H. H. WANG, *Fed. Proc.* 23, 565 (1964).

⁵ S. GARB, *Am. J. Physiol.* 182, 601 (1955).