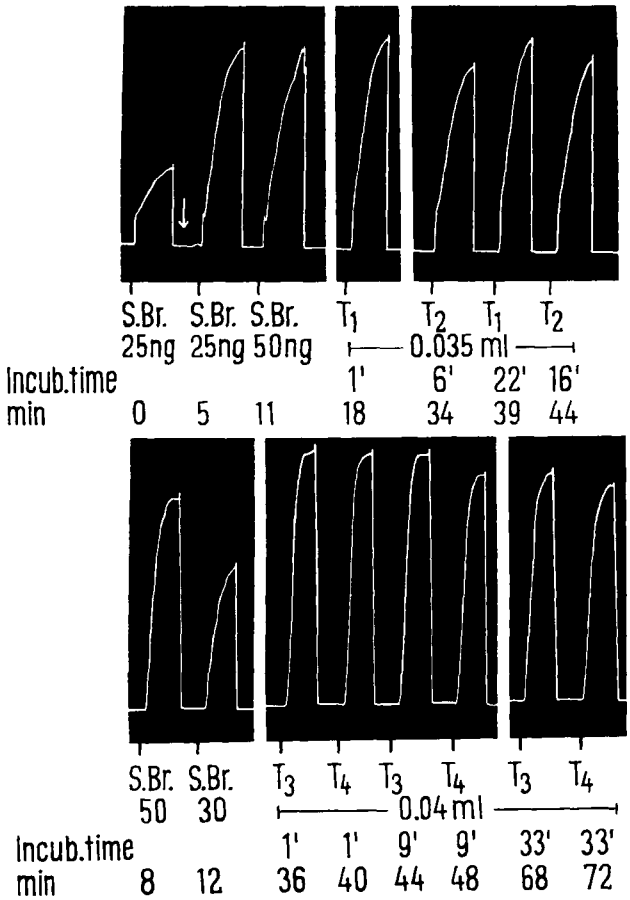

Fig. 3. Inhibition of kininase by cysteine—Assay on the isolated guinea-pig ileum (same conditions as Figure 1) of aliquots of the following incubation mixtures: T_1 = cysteine (2.85 mg/ml) + bradykinin (0.71 µg/ml); T_2 = *washed* guinea-pig ileum + cysteine (2.85 mg/ml) + bradykinin (0.71 µg/ml); T_3 = cysteine (2.48 mg/ml) + bradykinin (0.75 µg/ml); T_4 = *washed* uterus + cysteine (2.48 mg/ml) + bradykinin (0.75 µg/ml). S.Br. = synthetic bradykinin. At the arrow, 100 µg of cysteine were added to the bath. Contact of the *washed* organs with cysteine for 3 min before incubation with bradykinin, prevented the inactivation of the peptide.

the peptide was gradually destroyed, complete inactivation being achieved after 10–30 min (Figure 2). However, if the segments of the *washed* organs were previously left in contact with cysteine for 2–5 min and then incubated with bradykinin, destruction of the peptide was not observed (Figure 3).

These results seem to indicate that the potentiating effect of cysteine on contractions produced by bradykinin on isolated rat uterus or guinea-pig ileum is probably due to inhibition by the amino acid, of kininase present in these tissues. Its smaller effect on the rat uterus could perhaps be related to weaker kininase activity of this organ, which would also contribute to the greater sensitivity of this pharmacological preparation to bradykinin.

Zusammenfassung. Die Wirkung von Bradykinin, aber nicht von Angiotensin I und II auf das isolierte Meerschweinchenileum und den Uterus der Ratte wird durch Cystein verstärkt. Die Sensibilisierung der glatten Muskulatur ist auf die Hemmung der Kininase, die sich in den Geweben jener Organe befindet, durch Cystein zurückzuführen. Fragmente von Uterus oder Ileum von Tieren, deren hintere Extremitäten unter Druck mit Tyrode durchströmt und zur vollkommenen Ausblutung gebracht wurden, inaktivieren das Bradykinin, wenn sie damit inkubiert werden. Diese Inaktivierung kann durch Vorbehandlung der Organe mit Cystein verhindert werden.

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Comparison of the Effects of ß-Palmitoyl Lysolecithin and Cardiac Glycosides in two Mammalian Tissues

In 1957 HAJDU et al.[1] reported the isolation of a substance identified as β-palmitoyl lysolecithin from various mammalian tissues. They found that it shared with the cardiac glycosides the properties of: (1) abolishing treppe in the frog's heart, and (2) having positive inotropic effects in the hypodynamic frog heart and the isolated ventricle of the squab. We were interested to see whether these findings could be extended to mammalian tissues. The test systems we used were the human erythrocyte and the isolated guinea pig ventricle. We chose these because of the large amount of data available as to the effects of the cardiac glycosides on them.

The monopalmitoyl-lecithin (β-lysolecithin) used in these studies was prepared as follows: (dipalmitoleyl)-*L*-α-lecithin (containing two unsaturated fatty acids per molecule) was isolated from baker's yeast by the method of HANAHAN and JAYKO[2]. From this material, the corresponding β-lysolecithin, monopalmitoleyl-lecithin, was obtained by enzymatic hydrolysis according to HANAHAN et al.[3], using *Crotalus atrox* venom[4]. This unsaturated lysolecithin was subjected to catalytic hydrogenation[2,3], which resulted in a product with the same melting point (195°) as the monopalmitoyl-lecithin described by HANAHAN et al.[3].

Fresh human red cells were obtained and incubated with $K^{42}Cl$ as previously described[5]. Plasma was separated and counted just before incubation, and at 1, 2 and 3 h.

[1] S. HAJDU, H. WEISS, and E. TITUS, J. Pharmacol. exp. Therap. *120*, 99 (1957).

[2] D. J. HANAHAN and M. E. JAYKO, J. Amer. chem. Soc. *74*, 5070 (1952).

[3] D. J. HANAHAN, M. RODBELL, and L. D. TURNER, J. biol. Chem. *206*, 431 (1953).

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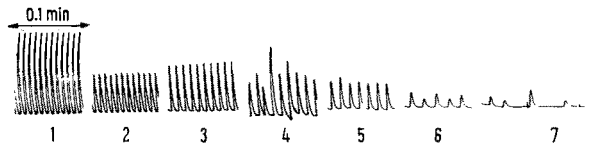
Effects of β-palmitoyl-lysolecithin on isolated guinea-pig ventricles.

Concentration of lysolecithin, μg/ml	Onset of negative inotropic effect after	Onset of irregularities after	Onset of negative chronotropic effects after	Remarks
80	7 min	none	none	Washed with NKR after 10 min no reversal of effects
105	1.5 min	none	1.5 min	Washed with NKR after 3 min no reversal of effects
105	2.5 min	2.5 min	1.7 min	
200	2 min	2.5 min	1.2 min	Slight (20%) positive inotropic effect in 1.5 min lasting for 0,5 min

The isolated guinea-pig ventricles were prepared as before[6], except that contractile force was recorded through a force-displacement transducer on a Grass polygraph. Records were made while the ventricle was perfused with normal Krebs-Ringer solution (NKR), then with the same solution in which half of the Ca was replaced with Na. Lysolecithin was added to the low-Ca solution, so that inotropic effects could be observed in the hypodynamic heart.

Results. (A) Erythrocytes. Four experiments were made with concentrations of lysolecithin from 30 to 200 μg/ml. In no case did the compound affect the rate of loss of K^{42} from the plasma. The highest concentration was about 4×10^{-4} *M*. This is 10^4 times the concentration of ouabain which inhibits K transfer by 50%, and 400 times that of digitoxin[5]. Hence it seems unlikely that palmitoyl lysolecithin shares with the cardiac glycosides the ability to inhibit active K transport in human erythrocytes. Haemolysis was not greater in blood containing lysolecithin than in control samples. On the other hand, dilute suspensions of erythrocytes in buffered saline (0.125% red cells) were haemolyzed by lysolecithin in concentrations as low as 10 μg/ml.

(B) Guinea-pig ventricles. Four experiments were made with this preparation which are summarized in the Table. In all four hearts the phosphatide had negative inotropic effects which progressed to the point of diastolic arrest. Contracture (rise of the base line) was never seen. All of these effects are different from those of cardiac glycosides which have been described before[7]. Negative chronotropic effects were seen with doses above 80 μg/ml. The irregularity, seen in one experiment with 105 μg/ml and in the one with 200 μg/ml, was a series of ventricular extrasystoles. A typical sequence of events is illustrated in the Figure. In three of the four experiments with lysolecithin, perfusion with NKR was resumed after the effects of the phosphatide were seen. In every case, the effects of the drug were unaffected by washing, and progressed to the termination of the experiment. In the experiment with the highest concentration of phosphatide, there was a slight transient positive inotropic effect (see Figure) which was quickly replaced by a more severe and longlasting negative inotropic effect. This was the only case in which there was any evidence of increased contractile force.



Effects of β-palmitoyl lysolecithin (200 μg/ml) on an isolated guinea-pig ventricle.

[1] NKR control. [2] Low-Ca control. [3] 1.8 min after lysolecithin. Note slight positive inotropic effect (not observed in the other experiments). [4] 3 min after lysolecithin. Irregularities have begun; contraction height decreasing. [5] 8 min. [6] 14 min. Severe effects, which progressed in 2 min to diastolic arrest. [7] 16 min. Diastolic arrest.

Our results do not agree with those of HAJDU et al. In view of the differences in the test systems used and of the different starting materials and procedures applied for the preparation of the lysolecithins, it is difficult to interprete the discrepancies. We feel, however, that our data present conclusive evidence that pure β-palmitoyllysolecithin, as prepared according to HANAHAN et al., does not possess any activity similar to that of the cardiac glycosides on the human erythrocyte and the isolated guinea-pig ventricle[8].

Zusammenfassung. 1957 sind von HAJDU et al. Versuche veröffentlicht worden, wonach sich aus verschiedenen Geweben eine Substanz mit Herzglykosid-ähnlicher Wirkung auf Kaltblüterherzen extrahieren lässt, welche als β-Palmitoyl-lysolecithin interpretiert wurde[1]. In unseren Versuchen wurde aus Hefe Dipalmitoleyl-lecithin isoliert und daraus durch enzymatische Abspaltung der α-ständigen Fettsäure und anschliessende katalytische Hydrierung reines, hämolytisch wirksames β-Palmitoyl-lysolecithin hergestellt. Diese Verbindung zeigte jedoch keine den Effekten der Herzglykoside verwandte Wirkung am isolierten Meerschweinchenventrikel sowie an menschlichen Erythrocyten.

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Departments of Pharmacology, University of Cincinnati and Northwestern University, Chicago (USA) and Pharmakologisches Institut der Universität Bern (Switzerland), November 27, 1961.

[6] R. L. VICK and J. B. KAHN, J. Pharmacol. exp. Therap. *121*, 389 (1957).

[7] R. L. VICK, J. Pharmacol. exp. Therap. *125*, 40 (1959). See effect of dihydro-ouabain in Figure 2 of this reference.

[8] This work was supported in part by a grant from the Life Insurance Medical Research Fund.

Occurrence of Virus-Like Particles in Cultured Cloudman S-91 Melanoma

In connection with recent cytological studies of cultured cells of the Cloudman S-91 mouse melanoma[1], further electron microscopic observations have revealed the presence of virus-like bodies in tumor cells cultivated *in vitro*. The present report concerns itself with a description of these virus-like particles, indistinguishable struc-

[1] R. BARISHAK, S. R. WELLINGS, and B. V. SIEGEL, Amer. J. Path. *38*, 371 (1961).