fusion, 24 h and 1 week after operation (taken from patients). The plasma kininogen was estimated with the method of DINIZ and CARVALHO[4]. The biological activity of the samples was calculated with the 4-point method on the isolated guinea-pig ileum. Synthetic bradykinin (VEB Berlin-Chemie[5]) was used as standard. The concentration of plasma kininogen is expressed in µg bradykinin per ml plasma released by trypsin incubation (BEq µg/ml).

The first series (8 patients) did not get any protease inhibitor. The second series (6 patients) were treated with the kallikrein-trypsin-inhibitor Contrykal® (VEB Arzneimittelwerk Dresden). The inhibitor activity of this protease inhibitor is expressed in anti-trypsin-units (ATrU). 1 ATrU inhibits the activity of 1 trypsin-unit (according IUB). In the starting of perfusion 20,000 ATrU Contrykal® were given into the heart-lung machine, 20,000 ATrU were administered to the patient during the perfusion and 20,000 ATrU Contrykal® were applicated to the patient after perfusion was finished.

The student-*t*-test was used in the statistical evaluation of the data (p 1%). All values are expressed as mean plus or minus standard error of mean.

Results. First series – without protease inhibitor. After a perfusion time of 5 min the kininogen content of plasma is not significantly different from the value 1 day before operation. During 30–60 min of perfusion the kininogen level drops but not to a significantly different level compared with the value of 5 min perfusion. However, 2 h after perfusion is finished, the plasma kininogen reaches a level which is significantly lower than the level after 5 min perfusion. 24 h after operation the initial kininogen content is regained (Figure).

Second series – with protease inhibitor. The kininogen content of plasma does not change significantly during and after extracorporal circulation. The kininogen values of the first and second series are significantly different 2 h after perfusion is finished (Figure).

The decrease of plasma kininogen level during and after extracorporal circulation and the prevention of kininogen depletion with the protease inhibitor refer to an activation of the kinin-forming system in extracorporal circulation. The mechanism of kinin formation may be related to the activation of fibrinolysis and blood clotting system in extracorporal circulation. It can be supposed that the common link which initiates each of these processes is the Hageman factor. The Hageman factor, activated by contact of blood with glass and other adsorbing surfaces, activates not only plasminogen and plasma thromboplastin antecedent but also the kinin-forming system (MARGOLIS[6], WEBSTER[7]). The kinin formation in this condition can be achieved both by activation of kallikreinogen induced by Hageman factor and plasmin and by the proteolytic action of plasmin on kininogen (EISEN[8]).

Besides the activation of Hageman factor, another mechanism of kinin formation can be discussed. TICE et al.[9] suggested, as a cause of increased fibrinolysis during hypothermic perfusion in open-heart surgery, the activation of lysosomal hydrolytic and proteolytic enzymes. Lysosomal kininogenases may also influence the plasma kininogen content in extracorporal circulation in cases of hypothermia.

A decrease of the plasma kininogen level was found by GOMAZKOV[10] in experiments on rabbits during blood perfusion through glass cuvettes. Using siliconized cuvettes no change of plasma kininogen could be observed. The author discussed kinin formation induced by glass contact as the cause of the fall of systemic blood pressure, blood pH and body temperature in his experiments. But it seems difficult to recognize whether kinins play a role in the very complex haemodynamic changes during and after extracorporal circulation. However, as described in this paper, treatment with a kallikrein-trypsin-inhibitor could be of use not only for inhibition of fibrinolysis (MAMMEN et al.[11]) but also for prevention of decrease of kininogen in extracorporal circulation.

Zusammenfassung. Beim extrakorporalen Kreislauf mit der Herz-Lungen-Maschine wurden die Veränderungen des Kininogengehalts im Plasma und ihre Beeinflussbarkeit mit einem Proteasen-Inhibitor untersucht. Der beobachtete Abfall des Kininogenspiegels im Plasma während und nach der Perfusion und die Hemmung der Kininogendepletion mit dem Proteasen-Inhibitor sprechen für eine Aktivierung des Kininogen-Kinin-Systems im extrakorporalen Kreislauf. Beziehungen zur Aktivierung des fibrinolytischen Systems werden angenommen.

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Bronchodilator Activity of Prostaglandin E_2 when Administered by Aerosol to Three Species

The prostaglandins are a series of hydroxy unsaturated fatty acids widely distributed throughout body tissues. Depending upon the species, pharmacologic preparation, and type of compound studied, the prostaglandins can either stimulate or inhibit bronchial smooth muscle[1].

The bronchodilator properties of aerosolized prostaglandin E_2 (PGE_2) became evident through our observation that it could protect guinea-pigs from a histamine-induced bronchoconstriction[2]. The present study evaluates the response of 3 species to the bronchodilator properties of this compound.

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PGE_2 was prepared by the method of LAPIDUS et al.[3] and kept frozen in a solution of 0.06 *M* sodium phosphate buffer (pH 7.4) at concentrations of 1 and 10 mg/ml. Appropriate working dilutions were freshly made from these stock solutions by diluting with distilled water or saline at the time of experiment.

Bronchoconstriction was produced in guinea-pigs by placing them in a closed chamber which had been sprayed (DeVilbiss nebulizer No. 40) for 60 sec with a 0.2% (base) histamine diphosphate solution. The control time of onset of respiratory distress and convulsions was recorded. Only animals with a control time in the range of 50–100 sec were used in the subsequent experiments. After a 4-h recovery period, these animals were exposed in a second chamber to an aerosol of the test compound for 2 min, allowed an additional 1-min period in the chamber before being exposed again to the histamine aerosol in the separate chamber as previously described. A comparison of the test and control preconvulsion ratios (T/C) for PGE_2 and isoproterenol indicated that PGE_2 is quite active and somewhat more potent than isoproterenol as an aerosol (Table). In similar experiments β-adrenergic blockade by pretreatment with 5 mg/kg of propranolol failed to prevent the bronchodilating properties of PGE_2. Exposure of a group of guinea-pigs to a concentration of 0.2% PGE_2 for 5 min failed to elicit any adverse effects. In a separate study, PGE_2 was shown to be effective in preventing respiratory symptoms of anaphylactic shock in guinea-pigs passively sensitized to horse serum and then exposed to an aerosol of the same.

Bronchodilator properties of PGE_2 and isoproterenol were compared in dogs and monkeys using modifications of the methods of KLIDE and AVIADO[4] and DIAMOND[5]. Male and female mongrel dogs (15–25 kg) and male Rhesus monkeys (7–10 kg) were anesthetized with Dial-Urethane and artificially respired with a Harvard pump. For the determination of pulmonary resistance the respiratory flow was measured with a Fleisch pneumotachograph (No. 0 or 00) and a Sanborn pressure transducer (No. 270). Transpulmonary pressure was measured with a Sanborn differential pressure transducer (No. 268B) bridged between the trachea and the intrapleural space. These measurements were simultaneously displayed on both axes of an oscilloscope, producing a respiratory loop. The electronic subtraction of compliance by means of a Hewlett-Packard respiratory preamplifier resulted in closure of the loop. Measurement of its slope with a precalibrated protractor allowed the determination of pulmonary resistance. Simultaneous values for pulmonary compliance were similarly obtained by determining the relationship between volume, derived by integrating air flow, and transpulmonary pressure. Histamine diphosphate was used in a 2% aqueous solution as the standard bronchoconstrictor. Drugs were administered as aerosols by means of a DeVilbiss nebulizer No. 42. A bypass to the inspiratory limb of the respiratory circuit was arranged so that the inspired air passed through the nebulizer chamber before entering the animal's lungs. Compression of the nebulizer bulb at the onset of the inspiratory cycle thus assured virtually complete evacuation of the chamber of the nebulizer and subsequent passage of drug mist into the lungs. Administration of known doses of drugs was achieved by calibrating the nebulizer and varying the number of inhalations and the strength of drug solutions. Histamine (as base) was given at a constant dose of 5 sprays (total of 1 mg), while PGE_2 was given in 5 to 20 sprays, depending upon dosage. The maximum constrictor effect of histamine occurs 2 min after administration; thus all drug effects were measured at 2 min after histamine spray. Percentage inhibition of the effects of histamine were calculated by comparison with a control histamine response. A minimum recovery period of 30 min was allowed between control histamine challenges. As soon as a control histamine response was reestablished, another dose of the same drug was generally then tested in the same animal. No animal was ever used to test more than a single bronchodilator.

Histamine is a potent bronchoconstrictor, capable of causing an extreme (generally greater than 100%) increase in resistance and decrease in compliance. Figure 1 illustrates, in dogs and monkeys, the comparative ability of PGE_2 and isoproterenol to prevent the subsequent effects of histamine given immediately after various aerosolized doses of these compounds. Statistical comparison of the regression equations was done by analysis of covariance[6].

In a series of 7 dogs, isoproterenol was an effective bronchodilator, capable of causing 100% reversal of histamine-induced changes in lung resistance. PGE_2, in contrast, was quite weak in antagonizing histamine in 10 dogs at doses up to 8 times as great as the maximally effective doses of isoproterenol. There was a statistically significant difference between the slopes of the dose-response curves for these 2 drugs ($F = 7.2$). In monkeys, however, both PGE_2 (8 monkeys) and isoproterenol (3 monkeys) were active bronchodilators. There was no significant difference between the slopes describing the effects of either drug on the change in resistance ($F = 0.8$),

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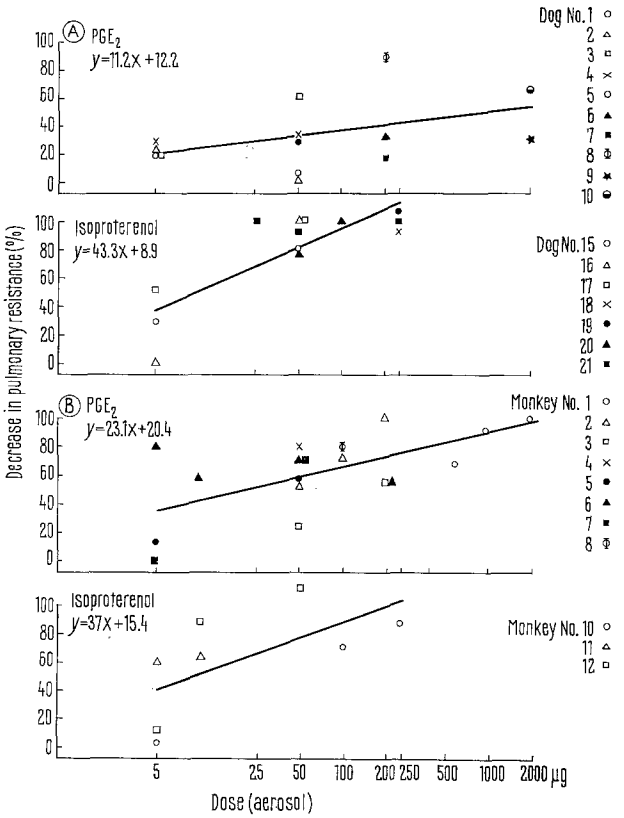
Effect of aerosolized PGE_2 and isoproterenol on histamine-induced bronchoconstriction in the unanesthetized guinea-pig

Aerosol spray concentration (%)	PGE_2			Isoproterenol		
	No. of animals	No. completely protected[a]	T/C score[b]	No. of animals	No. completely protected[a]	T/C score[b]
Buffer control	6	0/6	0.92 ± 0.10	–	–	–
0.0001	10	1/10	1.66 ± 0.16[c]	21	0/21	1.35 ± 0.10
0.0005	10	2/10	2.19 ± 0.19[c]	30	4/30	1.63 ± 0.12[c]
0.001	10	8/10	2.78 ± 0.15[c]	27	13/27	2.31 ± 0.15[c]

[a] Ability to withstand 3 times the control preconvulsion time (T/C = 3.0). [b] Mean $\pm$ S.E. [c] Significant protection ($p < 0.05$).

indicating that in the monkey PGE_2 is equivalent to isoproterenol in antagonizing this histamine-induced change. Pulmonary compliance was increased in a manner coincident to the decrease in pulmonary resistance. PGE_2 generally caused hypotensive effects of short duration in both dogs and monkeys. Pretreatment of monkeys with either propranolol, hexamethonium or atropine failed to prevent the bronchodilator effects of PGE_2.



Antagonistic effects of PGE_2 and isoproterenol on histamine-induced increases in total lung resistance in the dog (A) and monkey (B).

These experiments demonstrate that PGE_2 has bronchodilating properties when administered as an aerosol. They also indicate that marked species differences are characteristic of this compound; the bronchodilator properties were superior in the guinea-pig and monkey compared with the dog. A lack of naturally occurring PGE_2 in the dog lung could be important here[7]. The effectiveness of another of the prostaglandins, PGE_1, when administered by aerosol has recently been described in guinea-pigs[8] and humans[9] and confirms our initial observations on PGE_2[2]. PGE_2 is a normal constituent of human lung tissue[10] and can relax isolated human bronchial muscle[11,12]. The potency, natural occurrence and rapid metabolism of the prostaglandins in the lungs of several species suggest a role for these substances both in the natural regulation of bronchial smooth muscle[1] and therapeutically as a bronchodilator aerosol.

Zusammenfassung. Es wird gezeigt, dass Prostaglandin in Aerosolform bei mehreren Arten als Bronchodilator wirkt. Die Wirksamkeit, natürliches Vorkommen und schnelle Metabolisierung der Prostaglandine sprechen für eine therapeutische Verwendung als bronchodilatierende Aerosole.

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Filamentous Structures in Normal Taste Bud

Many observations have been made concerning the existence of filamentous structures in different cell types[1], particularly in sensory receptors[2] and specifically in the taste bud[3]. The subject of the present note is in fact the summary description of the fine features of the filamentous structures present in the main cell types of the taste bud.

Materials and methods. Foliate papillae of adult male rabbits were fixed in glutaraldehyde and post-fixed in osmium according to the usual electron microscopic techniques.

Results and discussion. Filaments permeate the cytoplasm of the I and II type cells of the taste bud. Their diameter varies from 70 to 100 Å. They follow an irregular course often assembling as interweaving bundles in the perinuclear zone (Figure I). The filaments are frequently situated near the plasma membrane taking part in the formation of intercellular junctional complexes of a desmosomal type[4] (Figure 2): in fact they insert on the cytoplasmic side of both the thickenings of the plasma membrane. Dense filamentous bundles can also be found within the processes of the type I cells (Figure 3). In the apical region of this cell type they become closely packed and parallel to each other occupying, besides the osmiophil granules and the centriole, the whole cytoplasmic matrix. Sometimes their cross-sections are clearly evident in the inside of the microvilli. In the type II cells, filaments interpose among the characteristic tubulo-vesicular profiles of the smooth endoplasmic reticulum, often accumulating around the lipid droplets (Figure 4). But they become most evident in the more advanced phases of the physiological degeneration of such a cell type: in fact this is subject to a continuous degenerative

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