

From the Medical Clinic II of St. Göran's Hospital, Stockholm, Sweden.

Head: H. Colldahl, M.D.

BRONCHIAL SENSITIVITY TO ALLERGEN WITH AND WITHOUT PREVIOUS HEPARIN ADMINISTRATION

By

KJELL-ORVAR PEGELOW

INTRODUCTION

The indications for heparin treatment have been widened considerably recently. In addition to acting as an anti-coagulant heparin has also been observed to be effective in lipemia-clearing and to possess anti-inflammatory and anti-allergic properties (14).

Heparin is produced by the mast cells, which are mostly associated with the precapillaries and capillaries but are often also distributed in connective tissue without any close connection with the blood vessels. The mast cells, which contain an abundance of metachromatic granules loaded with heparin, have also been shown to be rich in histamine (17). It is not yet fully clear if histamine is produced by the mast cells themselves or is taken up and bound to the strongly acidic heparin. In any case it seems as if histamine, too, can be produced in tissues lacking mast cells, e g brain tissue (15).

SOME PROPERTIES OF HEPARIN

Chemically heparin is a mucopolysaccharide. In consequence of its strong negative charge it can be bound to positively charged protein molecules such as, for example, antigen or antigen-antibody complexes. Furthermore, because of its acidic reaction it binds basic amines such as histamine and serotonin

(7). Heparin binds not only endogenous amines but also binds or neutralizes basic substances exogenously introduced, such as polymyxin B, compound 48/80 and venoms, *i.e.* well-known histamine liberators. If such substances are injected into mice in lethal doses it has been shown that the mice survive if they get heparin before or soon after the injection of the histamine liberator (11).

Recently *Hansson et al.* (8) have shown that when healthy men and non pregnant women are given normal clinical doses of heparin by intravenous injection they exhibit an increase in plasma histaminase (diamine oxidase). The enzyme activity increases immediately after the injection and after 15 minutes has reached a plateau. Subsequently, a further increase is recorded and this reaches a maximum of up to 100 times the initial value some 60 to 90 minutes after the injection. The plasma concentration of C^{14} -histamine, given by infusion, decreases after an injection of heparin. The increase of plasma histaminase produced by heparin seems thus to be of importance for the inactivation of histamine *in vivo*.

As early as in 1937 *Wising* (19) showed that heparin binds complement in blood. Therefore the Wasserman reaction cannot be carried out on heparinized blood. *Videbæk* (18) and later also *Hartman* (9) and others have reported good results from heparin treatment in some cases of autoimmune hemolytic anemia. A positive Coombs test turned negative after the administration of heparin.

In laboratory experiments *Dougherty* (6) has shown that heparin stimulates fagocytosis and pinocytosis. By pinocytosis is meant the ability of a wide variety of cells to take up very small particles and substances in solution from the surrounding ground substance. The process is very similar to fagocytosis but works on a minor scale. It is interesting that heparin also has been shown to induce and stimulate pseudopod formation in *Amoebae* (1).

Heparin seems to potentiate antibiotics and also to provide protection against the toxic effects of, for example, neomycin and polymyxin B (10, 16).

Johansson (12) has shown that the release of 5-hydroxy-tryptamine in anaphylaxis in rabbits can be inhibited if the sensitized rabbits are pretreated with large doses of heparin 20 minutes before the injection of the anaphylactic dose. Heparin prevents the fall of circulation platelets and increases the survival time of the animals. A positive correlation between the severity of anaphylactic shock and the degree of decrease in platelet count has been shown by *Kopeloff & Kopeloff* (16).

Johansson's investigations indicate that the strongly negatively charged heparin prevents the platelets from agglutinating and thereby disintegrating and releasing the contents of serotonin among other substances. Intravascular coagulation, which seems to be an important link in the anaphylactic reaction, is also prevented by heparin.

HEPARIN TREATMENT IN ALLERGIC CONDITIONS

In later years there have been some reports of clinical trials with heparin treatment in allergic conditions.

Dougherty & Dolowitz (5) have reported on the good effects of heparin in the treatment of acute asthma attacks. They gave 10,000–20,000 units intravenously to the patients within the first 24 hours of an attack. In 23 of 24 cases the breathing was eased both subjectively and objectively. The same authors have also reported positively on the effects of heparin treatment in hay fever and laryngotracheal bronchitis in children.

Cohn (3) has shown that in case of acute asthma heparin reduces almost immediately the flow resistance and increases the vital capacity.

Boyle et al. (2) treated with double-blind procedure 37 patients with different types of obstructive pulmonary disease. One group included patients with acute bronchospasm classified as "status asthmaticus". They were at once given a single intravenous injection of 20,000 units heparin or placebo (saline) as the only treatment. If adequate relief was not obtained at

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Results of Provocation Tests with Specific Allergen in Patients with Extra

Initials Date of birth Sex	Date of inhalation	Strongest antigen solution inhaled	Heparin inj. before inhalation	Vital capacity (lit.)		
				Before antigen inhalation	After strongest antigen sol. inhal.	After inhalation of epi- nephrine
B.S. 420909 female	650524 650531	Horse ⁻⁴ Horse 10 ⁻⁴	— 12500 IU	2.9 3.0	2.0 2.3	2.9 2.7
G.L. 341009 female	650512 650520	Dog 10 ⁻² Dog 10 ⁻²	— 12500 IU	3.5 3.3	3.1 2.9	3.4 3.5
M.L. 461112 female	651115 651124	Timothy 10 ⁻³ Timothy 10 ⁻³	— 15000 IU	3.3 3.0	1.8 2.2	2.8 2.8
S.G. 481013 male	651125 651208	Cat 10 ⁻² Cat 10 ⁻²	— 15000 IU	4.3 4.6	1.6 2.0	3.5 4.3
O.Ä. 451006 male	660119 660202	Horse 10 ⁻⁴ Horse 10 ⁻⁴	— 15000 IU	4.6 4.4	3.3 3.0	4.5 4.4
K.I. 490825 male	660314 660330	Horse 10 ⁻⁴ Horse 10 ⁻⁴	— 15000 IU	4.5 5.0	2.2 4.1	4.2 4.7
G.L. 380115 male	660414 660427	Horse 10 ⁻⁴ Horse 10 ⁻⁴	— 20000 IU	6.3 6.7	4.1 5.4	5.8 5.4
U.C. 320105 female	660512 660525	Horse 10 ⁻⁴ Horse 10 ⁻⁴	— 20000 IU	4.3 4.5	3.5 3.8	4.6 4.0
M.A. 460425 female	660512 660518	Horse 10 ⁻⁵ Horse 10 ⁻⁵	— 20000 IU	3.3 3.3	2.4 2.5	2.8 2.3
N.H. 430930 male	660404 660504	Horse 10 ⁻² Horse 10 ⁻²	— 20000 IU	5.6 5.7	4.1 4.6	5.3 5.4

Chronic Asthma, with and without Previous Heparin Administration.

FEV _{1,0} (lit./sec.)			Peak flow (lit./min.)		
Before antigen inhalation	After strongest antigen sol. inhal.	After inhalation of epi- nephrine	Before antigen inhalation	After strongest antigen sol. inhal.	After inhalation of epi- nephrine
2.5	1.3	2.0	310	155	265
2.2	1.4	1.9	360	175	285
2.6	2.1	2.6	295	185	300
2.6	2.2	2.6	320	205	335
2.8	1.5	1.5	380	180	240
2.1	0.9	2.1	340	180	295
3.5	1.2	2.7	390	130	260
4.1	0.9	2.4	420	130	340
3.2	1.8	2.9	400	180	340
3.2	1.4	2.9	440	160	340
2.8	1.0	2.8	320	220	340
3.9	2.5	3.6	435	220	400
4.6	2.7	4.1	585	300	540
4.7	3.5	3.8	580	450	560
2.8	1.7	3.3	345	180	350
3.5	2.2	2.6	420	210	310
2.8	1.6	2.2	410	280	320
2.5	1.5	1.5	410	255	230
4.9	3.9	5.0	540	450	520
5.2	4.0	4.5	560	460	540

the end of 15 minutes the patients received conventional therapy. Another group included patients with severe chronic bronchopulmonary disease with respiratory distress due to bronchospasm or tenacious secretions, or both, who were already receiving maximal conventional therapy including, in many cases, corticosteroids. All patients were considered resistant to their treatment and had exhibited a stable or deteriorating course for 24 hours or more, prior to inclusion in the study. Instead of increasing or adding corticosteroids or otherwise modifying therapy, 20,000 units intravenous heparin or placebo were administered followed by 10,000 units at 12-hours intervals for three additional doses. No other change in therapy was permitted and the effect was judged in each case before 24 hours had elapsed. The results are given for the two groups taken together. 19 patients had been given heparin and 18 placebo. In the heparin treated group 68 per cent of the cases showed complete relief or considerable improvement, while in the placebo groups 72 per cent manifested no response.

Johansson (13) has given patients with food allergy intradermal injections of the specific allergen in a dose large enough to provoke temporary thrombocytopenia and produce local or generalized reactions. If he gave 10,000–20,000 units of heparin 30 minutes before the test the platelet count remained unchanged and no local or generalized symptoms appeared.

Dahl (4) has described 4 cases of asthma with a strong hypersensitivity to birch, dog, horse and housedust respectively. By giving 10,000–20,000 units of heparin intravenously 2–15 minutes before the inhalation of specific allergen he was able to reduce the sensitivity 10–100 times. 3 cases of food allergy, who had gastrointestinal troubles and asthma after the administration of allergen, were also able to tolerate the food in question very well, if they had 10,000 units of heparin immediately before the provocation meal and 5,000 units one hour later. In the cases with inhalation allergy, *Dahl* has not given any further description of the methods for the provocation tests, nor has he indicated whether he has judged the results only from auscultation of the lungs or from lung function studies as well.

OWN INVESTIGATIONS

Material and Methods

In order to investigate with objective methods the possible effect of heparin on the bronchial sensitivity to specific allergens I have studied 10 cases with extrinsic asthma, on whom the provocation tests have been performed with and without previous heparin administration. The composition of the material appears in Table 1.

The provocation was made by letting the patients inhale 1 ml nebulized antigen solution each time, in increasing concentrations at ten minute intervals. A de Vilbiss nebulisator and compressed air was used. In addition to auscultation of the lungs the patients were studied before and during the provocation as regards vital capacity, timed vital capacity ($FEV_{1.0}$) and peak flow. The provocation test was considered positive if the reduction of these values occurred promptly and if the mean change amounted to 15 per cent or more. The first provocation was made without preceeding heparin administration, while the second was performed 1-2 weeks later and then the patients were given heparin intravenously 15-60 minutes prior to the expected asthma reaction. No change in the status or treatment of the patients before the second provocation occurred. Before the second test, 12,500 units of heparin were given to 2 cases, 15,000 units to 4 cases and 20,000 to 4 cases.

In one case platelets were counted before the test and 15 and 45 minutes after the appearance of the asthmatic reaction. This was done both in the test with a previous injection of heparin and in the one without any such injection. In 4 cases the platelets only were counted in the same way in the test where heparin was used.

RESULTS

All the 10 patients thus examined showed almost identical results with both provocations (see Table 1). The bronchial sensitivity in this material has consequently not been influenced by the administration of heparin. In the few cases where platelets were counted no significant change in number was registered.

DISCUSSION

Why then did the effect of heparin on the bronchial sensitivity fail to appear in these provocation tests?

With the rather cautious methods of exposure which we use, the patients seldom will get more severe asthmatic reactions and we have not in previously made studies seen any platelet decrease. If, as some authors state, the main "anti-allergic"

effect of heparin is that it prevents the platelets from agglutinating, and thereby prevents the release of 5-hydroxytryptamine and the onset of intravascular coagulation, then it is probable that the reaction to allergen exposure with the method mentioned above is too weak and that these effects of heparin are of no real significance.

In his cases Dahl has provoked severe asthma and in one case allergic shock as well, and this might explain why he obtained a protective effect from heparin when repeating the provocation in the same way. Another explanation could be that after the allergic shock his patients, by analogy with anti-anaphylaxis in guinea pigs, remained in an anti-allergic state. If this was the case, then the fact that the antibodies were already saturated could prevent any more severe reaction from being provoked by a further administration of the allergen within one week.

If heparin did have a real histamine and/or serotonin binding capacity, it is very likely that this ought to be evident even when the provocation tests give rise to more moderate asthma.

Even if histamine is only one of several substances which produce asthmatic symptoms with allergen exposure, it seems reasonable to assume that the increase in plasma histaminase consequent on an injection of heparin and thereby the inactivation of histamine, should be demonstrable even in provocation tests.

In one of the cases in these series the threshold value for histamine inhalations has also been fixed. The patient had a bronchospasm at a histamine concentration of 1 mg/ml (normal threshold value for healthy individuals is 32 mg/ml). The test was repeated one week later and the patient was than pretreated with 12,500 units of heparin. The same threshold value for histamine inhalation was found.

SUMMARY

Past work has shown the modifying effect of heparin on allergic reactions, but the mechanism behind this effect is not

fully understood. It may include the binding of histamine and serotonin, the prevention of local intravascular coagulation and thereby the release of serotonin from platelets, the neutralization of histamine liberators, an increase of the plasma histaminase activity or even other processes.

In provocation tests with allergen on 10 patients with specific asthma no change in bronchial sensitivity could be shown after pretreatment with heparin. However, further investigations and clinical trials will be needed if we are to learn more about the alleged therapeutic value of heparin in the treatment of allergic diseases.

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