

Department of Allergology, Karolinska sjukhuset, S-104 01 Stockholm 60, Sweden.

THE RELEASE OF HISTAMINE
FROM BLOOD CORPUSCLES
BY ANTIGEN-ANTIBODY REACTIONS

II. *The Histamine Release from Whole Blood after Adding
Specific Allergen in Hyposensitized Persons*

By

GUNNAR BERGQUIST & ÅKE NILZÉN

Treatment with specific pollen allergen extract causes a reduction in both the patient's general reaction and the skin or conjunctival reaction in functional testing. This can be observed already during the first half year of treatment, but the full effect is only obtained after several years (8, 10, 14). Changes in the serum globulin fractions, reagin and blocking antibody titer, during and after hyposensitization were demonstrated (5, 12). During the first 2-3 months the reagin titer rises, to fall thereafter to the initial value, which usually occurs after about one year's treatment. Blocking antibody, which is not present in untreated persons, appears already in serum after 2-3 months' treatment, but first reaches its maximal value after 10-12 months. These observations were made when treating with specific, aqueous allergen extract, but also when treating with allergen emulsions similar phenomena were noted (5, 13, 15). A certain correlation between high, blocking-antibody titer and good clinical results has been reported (15). Injection of blocking antibody has been shown to produce a desensitizing effect (4).

During recent years the release of histamine *in vitro* from blood corpuscles following incubation with specific allergen

has begun to be used as a diagnostic agent, and for determining whether treatment has been successful (6, 7, 9). Different methods have been devised for the determination of histamine release (HR), which have been discussed in a work published by us, where we also described the method for determining HR, which we use (3).

What is interesting is, that specific antiallergic treatment shows a reduced tendency for histamine release in the blood (2, 9, 11). Thus, van Arsdel and Middleton report that in 2 out of 14 pollen-allergic persons HR was greatly reduced after treatment (2). The duration of treatment varied widely from 4 weeks to 16 months. Van Arsdel present the results of HR determinations for 23 patients before treatment and for 12 after treatment. The initial value of HR was higher than after treatment. Recently an investigation of 12 patients has been published: 6 received high doses of specific allergen extract for a period of 5 months, and 6 lower doses. Four of the patients in the high-dose group showed "a marked decrease" of HR after treatment (9).

MATERIAL AND METHODS

In an earlier work (3) it was pointed out that specific antiallergic treatment seems to decrease HR in plasma after addition of the allergen to whole blood. Our case material at that time was ununiformly treated and in individual cases we found significant HR despite prolonged hyposensitization. There is difficulty in obtaining uniform groups for clinical research which can be used satisfactorily for statistical analysis. In the present work we have tried, however, to standardize to some extent our investigations. Thus, all the subjects were given injections of pollen extract of the allpyral type. (Dome Chemicals Inc., New York, U.S.A.). All the patients were given an injection once a week with the following doses: 0.5 ml, 100 PNU/ml; 0.1 ml, 1000 PNU/ml; 0.2 ml, 1000 PNU/ml; 0.4 ml, 1000 PNU/ml; 0.8 ml, 1000 PNU/ml; 0.15 ml, 10,000 PNU/ml; 0.3 ml, 10,000 PNU/ml; 0.6 ml, 10,000 PNU/ml. 10,000 PNU = Protein Nitrogen Units.

This series comprised 17 subjects all of whom were highly sensitive to pollen. All, excepting one (280719), had been previously treated with aqueous extract or allpyral extract, but at least half a year had elapsed since the previous treatment. Several of the subjects reported on here were included in the earlier investigation (3), when they were selected at random, however, and without taking into account the duration of treatment etc.

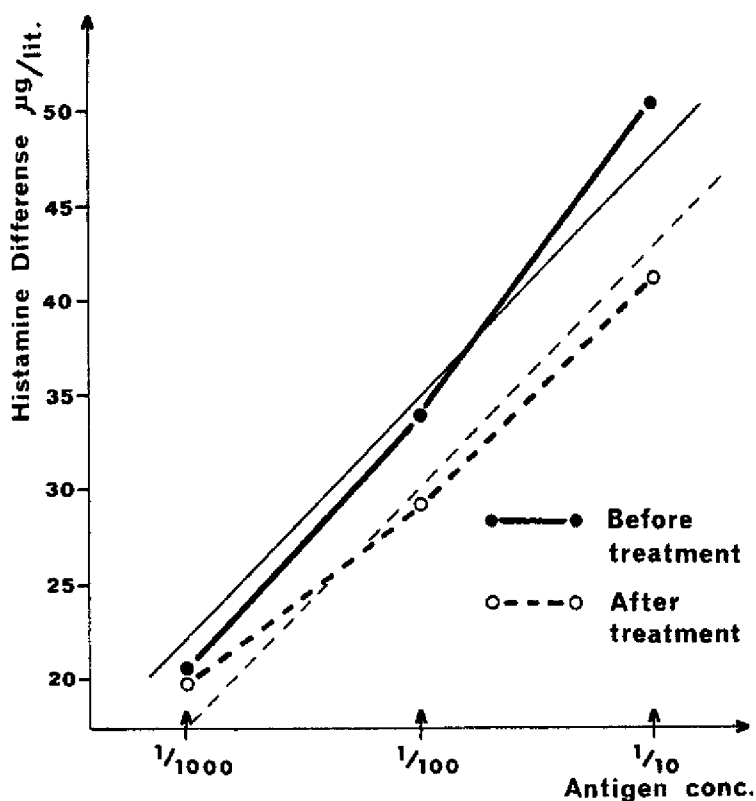


Fig. 1.

The difference between serum histamine level and the physiologic saline control before and after treatment.

RESULTS

Since, as mentioned above, one can expect only to obtain the full serologic and clinical effect of hypsensitization after a relatively long and continuous time of treatment, the results reported on here can only show the results of the initial effect of hypsensitization. The effect of 3 years' continuous treatment will be reported on a later occasion.

The methods differ from that previously described (3), in

Histamine in Serum after Incubation of Blood

Patient	Sex	Allergen	Histamine content in serum					
			Before treatment				Concentration	
			Saline		1:1000		1:100	
			$\mu\text{g/ml}$	%	$\mu\text{g/ml}$	%	$\mu\text{g/ml}$	%
430503	♂	T	0.031	100	0.059	190	0.055	177
180415	♀	B	0.019	100	0.017	89	0.017	89
331202		T	0.039	100	0.072	185	0.090	231
200603	♀	B	0.048	100	0.074	154	0.063	131
100721	♂	T	0.022	100	0.016	73	0.028	127
110817	♂	T	0.016	100	0.027	169	0.034	213
300513	♂	B	0.012	100	0.043	358	0.052	433
550110	♂	T	0.020	100	—	—	0.076	380
380109	♂	B	0.021	100	0.038	181	0.030	143
380109	♂	T	0.024	100	0.058	242	0.040	167
380109	♂	B	0.013	100	0.016	123	0.018	133
231010	♂	T	0.017	100	0.024	141	0.025	147
300913	♀	B	0.019	100	0.052	274	0.055	289
440713	♂	T	0.070	100	0.104	149	0.208	297
280719	♂	T	0.018	100	0.033	183	0.048	267
310220	♂	T	0.034	100	0.052	153	0.067	197
280424	♂	T	0.022	100	0.067	305	0.095	432
480729	♀	T	0.081	100	0.099	122	0.114	141

Allergen: B = Birch pollen extract.

T = Timothy pollen extract.

asmuch as the amount of extract or normal saline added to the whole blood has been decreased to 0.2., which is quite sufficient to obtain manifest HR.

Table 1 shows the values found, expressed in $\mu\text{g/ml}$. plasma before and after treatment, as well as HR in per cent, the control value (incubation with normal saline) being set at 100 per cent

Statistically, there is no significant difference between HR

Appropriate Allergen before and after hyposensitization.

ml and expressed in %

Allergen	After treatment									
	Concentration of allergen									
	Saline									
	1:10				1:1000		1:100		1:10	
$\mu\text{g/ml}$	%	$\mu\text{g/ml}$	%	$\mu\text{g/ml}$	%	$\mu\text{g/ml}$	%	$\mu\text{g/ml}$	%	
0.069	223	0.050	100	0.053	106	0.066	132	0.070	140	
0.036	189	0.027	100	0.020	74	0.017	63	0.027	100	
0.103	264	0.038	100	0.058	153	0.060	158	0.065	171	
0.096	200	—	—	0.009	—	0.006	—	0.024	—	
0.035	159	0.025	100	0.025	100	0.027	108	0.058	232	
0.036	288	0.011	100	0.023	209	0.024	218	0.033	300	
0.038	483	0.061	100	0.084	138	0.068	111	0.091	149	
0.085	425	0.031	100	0.064	206	0.068	219	0.046	148	
0.055	262	0.022	100	0.048	218	0.050	227	0.070	318	
0.051	213	0.022	100	0.052	236	0.060	273	0.070	318	
0.015	115	0.013	100	0.031	238	0.031	238	0.048	369	
0.034	200	0.017	100	0.035	206	0.032	188	0.037	218	
0.091	479	0.031	100	0.035	113	0.050	161	0.058	187	
0.251	343	0.026	100	0.126	485	0.178	685	0.210	808	
—	—	0.048	100	0.071	148	0.084	175	0.088	183	
0.064	188	0.026	100	0.030	115	0.033	127	0.045	173	
0.122	555	0.042	100	0.074	176	0.116	276	0.130	310	
0.141	174	0.015	100	0.015	100	0.037	247	0.061	407	

before and after treatment whether the effect is measured in per cent of the physiologic saline controls or according to the difference from the physiologic saline control ($\mu\text{g/ml}$). A certain tendency for HR to decrease after treatment is reflected, however, in the figure where the effect, according to the difference from the physiologic saline control, is recorded. There is no statistical difference, however, for any dosage level, between before and after treatment.

DISCUSSION

As was mentioned in the introduction, there are investigations which indicate that specific treatment increases the ability of plasma to eliminate the effect of an added allergen *in vitro*. The authors of the present work were unable to verify this with the technique applied. But a tendency for HR to decrease was observed. However, continuous treatment with the extract was given only for 8 weeks, which may be one of the reasons why the difference before and after treatment was comparatively slight.

SUMMARY

Histamine release in plasma after the addition of specific allergen before and after hyposensitization has been studied. No statistically significant difference before and after treatment has been observed. In this series duration of treatment was 8 weeks.

REFERENCES

1. *van Arsdel, J. P.*: Effect of Specific Hyposensitization on Histamine Release from human leukocytes. *Fed. Proc.* 24, 632, 1965.
2. *van Arsdel, P. P. & Middleton, E.*: The effect of Hyposensitization on the in vitro histamine release by specific antigen, *J. Allergy*, 62, 348, 1961.
3. *Bergquist, G. & Nilzén, A.*: The release of histamine from blood corpuscles by antigen-antibody reactions. *Acta Allergol.* 23, 363, 1968.
4. *Bernton, H. S., Chambers, D. C. & Qeurry, M. W.*: Therapy of ragweed pollinosis with blocking antibody. *J. Allergy*, 33, 356, 1962.
5. *Cooke, R. A., Barmard, J. H., Hebal, S. & Stull, A.*: Serological evidence of immunity with coexisting sensitization in a type of human allergy hay fever. *J. Exp. Med.*, 62, 733, 1935.
6. *Katz, G. & Cohen, S.*: Experimental evidence of histamine release in allergy. *J.A.M.A.*, 117, 1782, 1941.
7. *Lichtenstein, L. M. & Osler, G. A.*: Histamine release from human leukocytes, *J. Exper. Med.*, 120, 507, 1964.
8. *Loweless, M. H.*: Studies of pollinosis, *J. Immun.*, 47, 165, 1943.
9. *van Metre, T., Lichtenstein, L. M. & Turk, A.*: Hay fever symptoms blocking antibody levels and leukocytes histamine release in patients receiving very high dosage immunotherapy with ragweed pollen extract. *J. Allergy*, 43, 181, 1969.

10. *Prigal, S. J.*: Fundamentals of Modern Allergy. New York 1960.
11. *Pruzansky, J. J. & Patterson, R.*: Histamine release from leukocytes of hypersensitive individuals. *J. Allergy*, 39, 44, 1967.
12. *Sherman, W. B.*: Changes in serological reactions and tissue sensitivity in hay fever patients during the early months of treatment. *J. Immun.* 40, 289, 1941.
13. *Sherman, W. B.*: Status of knowledge of desensitization. Congress of Allergy, New York, 1961.
14. *Sherman, W. B.*: Hypersensitivity, London, 1968.
15. *Sherman, W. B. & Conell, J. T.*: Changes in skin sensitizing antibody titer following two to four years of injection (aqueous treatment). Annual meeting of Am. Acad. of Allergy 1966 and *J. Allergy*, 43, 22, 1969.

This document is a scanned copy of a printed document. No warranty is given about the accuracy of the copy. Users should refer to the original published version of the material.