

Basic Study of Cernilton

Immuno-Serological Findings

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4. Results

A) Serological Study of Rabbit-Serums Immunized with Cernitin T-60 and Cernitin GBX

Serological study was made of the serums (7 rabbits) immunized according to method 1).

A. Precipitin Reaction (double layer method):

Table 1

		Precipitin Reaction (diluted antisera)			
		Undiluted Antiserum	X2	X4	X8
Rabbits Group A (T-60 Administration Group)	No. 1	—	—	—	—
	2	—	—	—	—
	3	—	—	—	—
	4	—	—	—	—
Rabbits Group B (GBX Administration Group)	No. 1	—	—	—	—
	2	—	—	—	—
	3	—	—	—	—

Notes: Cernitin T-60 (30 mg/ml) was used as precipinogen for antisera of Group A (Nos. 1—4). For antisera of Group B (Nos. 1—3) Cernitin GBX (1.5 mg/ml) was used.

As may be noted from the Table, results were negative in all cases, revealing no antibodies at all.

B. Haemagglutination Reaction:

Results of haemagglutination reaction test carried out according to method 4) are as given in Table 2.

Table 2

		X10	X20	X40	X80	X160	X320	X640
Rabbits Group A (T-60 Administration Group)	No. 1 a	+++	+++	++	+	+	—	—
	b	+++	+++	++	+	—	—	—
	No. 2 a	+++	+++	++	+	+	—	—
	b	+++	+++	+	—	—	—	—
	No. 3 a	+++	+++	+++	++	+	+	—
	b	+++	++	++	+	—	—	—
	No. 4 a	+++	+++	++	+	+	—	—
	b	+++	+++	++	+	—	—	—
Rabbits Group B (GBX Administration Group)	No. 1 a	—	—	—	—	—	—	—
	b	—	—	—	—	—	—	—
	No. 2 a	—	—	—	—	—	—	—
	b	—	—	—	—	—	—	—
	No. 3 a	—	—	—	—	—	—	—
	b	—	—	—	—	—	—	—

Notes: a . . . Corpuscles sensitized with Cernitin T-60

b . . . Corpuscles sensitized with Cernitin GBX

The agglutination values of the rabbit-serums of Group A (immunized with Cernitin T-60) were 160—320 with Cernitin T-60 sensitized corpuscles. Even with Cernitin GBX sensitized corpuscles the values were as high as 40—80. The rabbit-serums of Group B (immunized with Cernitin GBX) showed no agglutination at all with Cernitin T-60 or Cernitin GBX sensitized corpuscles.

C. Gel-Precipitin Reaction (Ouchterlony's Method):

Precipitin reaction test as carried out according to Ouchterlony's method, with the antiserums (Group A 4 cases, Group B 3 cases) placed in the center and the antigens in the peripheral areas, as shown in the left chart, revealed negative results in all cases with no appearance of precipitation lines.

B) Sensitigenocity of Cernitin T-60 and Cernitin GBX

A. Anaphylactic Shock (Guinea Pigs): Antigens (15 mg/ml) were administered intravenously at a dose of 1 ml to guinea pigs sensitized according to method 2) and observation was made as to the presence of anaphylactic shock.

Table 3

	Shock Injection	Guinea Pigs	Symptoms
Cernitin T-60 Sensitized Group	Cernitin T-60 (30 mg/ml) 1 ml i.v.inj.	1. 240 g	— (Survived)
		2. 260 g	— („)
		3. 250 g	— („)
		4. 280 g	— („)
		5. 260 g	— („)
Cernitin GBX Sensitized Group	Cernitin GBX (15 mg/ml) 1 ml i.v.inj.	6. 250 g	— („)
		7. 270 g	— („)
		8. 240 g	— („)
		9. 250 g	— („)
		10. 260 g	— („)

As may be seen from the Table, no cases showed anaphylactic shock and all cases survived.

B. Arthus' Phenomenon (Rabbits): Rabbits were immunized according to method 1). After shaving off the hairs, the antigens (0.1 ml) were administered subcutaneously to the animals at 6 sites and observation was made as to the presence of the symptoms of reddening and induration. Results are given in Table 4.

Table 4

		Cernitin T-60			Cernitin GBX		
		15 mg/ml	3 mg/ml	0.6 mg/ml	15 mg/ml	3 mg/ml	0.6 mg/ml
Rabbits Group A Immunized with T-60	No. 1	1.2 × 1.1 cm	—	—	—	—	—
	2	1.4 × 1.2 cm	—	—	—	—	—
	3	1.0 × 0.8 cm	—	—	—	—	—
	4	0.7 × 0.8 cm	—	—	—	—	—
Rabbits Group B Immunized with GBX	No. 1	—	—	—	—	—	—
	2	—	—	—	—	—	—
	3	—	—	—	—	—	—

Note: Figures indicate sizes of reddening (in diameters).

As the results would show, there was observed a slight degree of reddening when Cernitin T-60 was injected subcutaneously at a concentration of 15 mg/ml in rabbits immunized with Cernitin T-60. No bleeding, necrosis or induration was noted, however.

4. Summary and Conclusion

Pollen extracts Cernitin T-60 and Cernitin GBX were studied immunoserologically to examine their antigenicity and sensitogenicity, with results as summarized below.

1) Examination was made as to the antibody-producing properties of Cernitin T-60 and Cernitin GBX using the serums of immunized rabbits. Results were negative in all cases by means of the precipitin reaction (double layer method) and gel-precipitin reaction (Ouchterlony's method) tests. By means of haemagglutination test the agglutination value was slightly elevated in Cernitin T-60 immunized rabbit-serums but not in Cernitin GBX immunized serums.

2) Observation was made as to anaphylactic shock in guinea pigs strongly sensitized with Cernitin T-60 and Cernitin GBX, but the results were negative in all cases.

3) Observation was also made as to Arthus' phenomenon using rabbits strongly sensitized with Cernitin T-60 and Cernitin GBX. When Cernitin T-60 was used as the antigen and given at a concentration of 15 mg/ml, there was observed a slight degree of reddening in rabbits immunized with Cernitin T-60. At lower concentrations no symptoms were revealed at all. Results were negative in all rabbits immunized with Cernitin GBX.

It may be said in conclusion that both Cernitin T-60 and Cernitin GBX have either no or, if any, an extremely slight degree of antigenicity or sensitogenicity.