

Moreover, the augmentary effect could be obtained exclusively in connection with extracts derived from the pituitaries of rats, not with those of men.

Summary. Following protracted injection in 16 rabbits of gonadotropic hormone derived from different sources we succeeded in one case in producing a progonadotropic serum, after treatment consisting of daily injections for a period of 5 months. 0.025 cc of this serum was able to multiply by 8 the gonadotropic effect of an aqueous extract of rat pituitaries. Control experiments showed that this augmentary effect was specific: it was neither obtained with normal serum acting on extracts of the rat pituitary, nor with progonadotropic serum acting on gonadotropic hormones of a different origin, *viz.*, of human pituitaries.

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Allergy in *Brucella* Infections.

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Cutaneous hypersensitiveness to specific antigen as a test for *Brucella* infection has been used by several authors.¹⁻¹¹ Unfortunately, the diversity of preparations used as antigen makes correlation of results difficult. As a rule, a positive reaction has been considered as denoting past or present infection.

Recently, the authors¹² published a method for the preparation of

¹ Fleischner, E. C., and Meyer, K. F., *Am. J. Dis. Children*, 1918, **16**, 268.

² Burnet, E. T., *Arch. Inst. Pasteur del Afrique du Nord*, 1922, **2**, 187.

³ Taylor, R. M., Lisbonne, M., and Vidal, L. F., *Mouvement Sanitaire*, 1935, **12**, 51.

⁴ Dubois, Ch., and Sollier, N., *Annals Inst. Pasteur*, 1931, **47**, 311.

⁵ Bastai, P., and Rotta, C., *Policlinico*, 1928, **35**, 393.

⁶ Levin, W., *J. Lab. and Clin. Med.*, 1930, **16**, 275.

⁷ Trenti, E., *Policlinico*, 1925, **32**, 867.

⁸ Mitra, M., *Pediatrics*, 1924, **32**, 721.

⁹ Bua, F., *Minerva Medica*, 1926, **6**, 394.

¹⁰ Giordiano, A. S., *J. Am. Med. Assn.*, 1929, **93**, 1957.

¹¹ Huddleson, I. F., Johnson, H. W., and Hamann, E. E., *Am. J. Public Health*, 1933, **23**, 917.

¹² Morales Otero, P., and Gonzalez, L. M., *PROC. SOC. EXP. BIOL. AND MED.*, 1938, **38**, 703.

a purified protein-antigen from *Brucella* and decided to use the preparation to investigate cutaneous hypersensitiveness to *Brucella* among milkers and cattle handlers in farms near the city of San Juan, P. R., where endemic abortion in cattle is prevalent. The method used was as follows: The desired amount of the purified protein produced from the *Brucella* organism is accurately weighed and suspended in a few cc of saline; then enough drops of N/10 hydroxide solution are added until a clear solution is obtained, and this is brought back to neutrality with N/10 HCl. The resulting solution is diluted with saline so that it may contain 0.1 mg per cc. By means of a sterile, tuberculin-type syringe and a 26-gauge needle, 0.1 cc of the diluted protein-solution is injected into the skin over the flexor surface of the forearm. The site of the injection is examined 48 hours later. In subjects that react positively, there is marked erythema and edematous induration. As a rule, the area of edema is much larger than the erythematous area. The size of the reaction varies with its intensity and in some cases there is a constitutional reaction shown by general malaise and slight fever, with pain or itching in the injected arm.

Two hundred and twelve adults occupied as milkers or cattle-handlers were tested. At the same time, 15 cc of blood were drawn and agglutination, complement-fixation, and opsono-cytophagic tests for *Brucella* were made. Of the 212 cases tested, 7 or 3.3% showed a positive complement-fixation, 5 or 2.3% gave a positive agglutinative reaction, 46 or 21.7% gave a positive opsono-cytophagic reaction, and 63 or 29.6% showed hypersensitiveness. Of these, 17 or 8% had constitutional reaction as shown by pain in the arm, general malaise, and slight fever, usually lasting 24 hours.

Of all the persons tested, 7 or 3.3% had a history of undulant fever. The reactions in this group are shown in Table I.

Of 7 persons working in our department, 4 were negative to all tests and 3 reacted as follows: One of us (P.M.O.), who has been

TABLE I.

Name	Skin-reaction*	Agglutination	Complement fixation*	Opsono-cytophagic index
F.G.	+++	Negative	++++	Moderate
J.R.	+	1-450	Negative	Slight
J.A.	Negative	1-450	++++	Negative
M.A.M.	+++	Negative	++++	"
F.S.	Negative	1-1600	+++	Marked
J.M.	"	1-1600	++++	Negative
C.R.	+	1-3200	+	"

*The purified *Brucella* protein was used as antigen. In grading the skin reaction the method recommended by Aronson¹³ was followed.

¹³ Aronson, J. D., *Am. Rev. Tuberc.*, 1934, **30**, 727.

handling *Brucella* cultures for the last 10 years, gave a positive (3+) cutaneous reaction and a weak positive (2+) complement-fixation, while the reactions in the opsono-cytophagic and agglutinative tests were negative. The other (L.M.G.), who has been working with us for 5 years, gave a positive (3+) cutaneous reaction, a positive (3+) complement-fixation, a positive opsono-cytophagic reaction and a negative agglutination. In the third (R.C.), who has been in contact with cultures for about 2 years, all 4 reactions were positive. Those 3 of us giving positive reactions are the only ones handling *Brucella* cultures, and we have never shown signs or symptoms of the disease.

The results show that there is no correlation between cutaneous allergy to *Brucella* and other immunological reactions, such as agglutination, complement-fixation and opsono-cytophagic index. As in Evans' report,¹⁴ in our series 15 cases that were positive to the opsono-cytophagic index, gave a negative cutaneous reaction, while 18 showing a positive cutaneous reaction were absolutely negative to the opsono-cytophagic test.

Contact with infected material causes the development of a state of hypersensitiveness as shown by injection of the purified *Brucella* protein, although signs or symptoms of the disease may not have been evidenced. This has been explained by the fact that hypersensitiveness usually appears later than agglutination and complement-fixation and that it lasts longer. However, in the light of recent experiments by Sabin,¹⁵ the possibility of sensitization without infection by coming in contact with infected material through abraded skin is worthy of careful consideration. The cutaneous test should be of value in epidemiological investigations, but until further work is done, the cutaneous reaction should not be considered as proof of present or past infection, since it only shows the development of a state of hypersensitiveness in persons that have been in close contact with infected material.

¹⁴ Evans, A. C., Robinson, F. H., and Baumgartner, L., *U. S. Public Health Rep.*, 1938, **53**, 1507.

¹⁵ Sabin, F. R., and Joyner, A. L., *J. Exp. Med.*, 1938, **68**, 659.