than those containing free HCl. It is also demonstrated that in this series the low pepsin content of achlorhydric gastric juices from cancerous stomachs was of the same order as that observed in achlorhydric juices from other sources.

Conclusions. The low pepsin content of achlorhydric gastric juices from carcinomatous stomachs is of the same order as the low pepsin content of achlorhydric gastric juices obtained from patients not presenting carcinoma of the stomach.

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Soluble Antigen in Lymphogranuloma Venereum.

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Neither the fundamental mechanism of the Frei reaction, which until recently was the only basis for the certain diagnosis of lymphogranuloma venereum, nor the nature of the reagent in the antigen which provokes a positive Frei test has yet been clearly defined. It has been observed that high-speed centrifugation of certain Frei antigens yields dermally active sediments containing virus elementary bodies[1] but nonreactive supernatant fluids.[1, 2] Furthermore, the passage of Frei antigens through Berkefeld filters[1, 2] or Seitz K discs[1] was found to reduce greatly or, more commonly, to abolish completely the dermal activity of the filtrate; it should be noted that Seitz K filters are permeable even to bacteria. These observations have led Findlay to the conclusion that the Frei test demonstrates an allergic reaction due to the presence of the elementary bodies of the virus in the material injected.

It has previously been shown that by propagation of the agent of lymphogranuloma venereum in the yolk-sac of the developing chicken embryo[3] or in the lungs of mice[4] there are obtained quantities of virus far greater than those present in other tissues. Inactivated suspen-

[1] Findlay, G. M., *Trans. Roy. Soc. Trop. Med. and Hyg.*, 1938, **31**, 587.

[2] Sullivan, M., and Ecker, E. E., PROC. SOC. EXP. BIOL. AND MED., 1935, **32**, 1024.

[3] Rake, G., McKee, C. M., and Shaffer, M. F., PROC. SOC. EXP. BIOL. AND MED., 1940, **43**, 332.

[4] Shaffer, M. F., Rake, G., and McKee, C. M., PROC. SOC. EXP. BIOL. AND MED., 1940, **44**, 408.

sions of elementary bodies derived from the yolk-sac, and separated from tissue constituents by differential centrifugation, have been successfully employed as antigens for the Frei test[5] and for the demonstration of complement-fixing properties in the serum of persons infected with the virus;[6] in the latter test suspensions of virus from mouse-lung have also been used.

Complement-fixation tests[6] have been performed in which falling dilutions of selected lymphogranulomatous human sera, known to fix strongly, were titrated against falling dilutions of antigen in the form of suspensions of elementary bodies sedimented from yolk-sac or mouse-lung by high-speed centrifugation. From the results it appeared that with increasing dilution of either reagent, greater concentrations of the other were required for fixation of a constant dose of complement. By holding the serum dilution constant it was possible to titrate simultaneously several antigens for their relative fixing potency.

Using this technic, further experiments showed clearly that the complement-fixing properties, found to be associated with the virus-containing sediment deposited at 12 000 rpm, were also demonstrable in the corresponding supernatant fluid which previously had been shown to be largely depleted of live virus.[3] In Table I are summarized results obtained with single or pooled suspensions of 3 different strains of the virus, propagated in the yolk-sac or mouse-lung. It may be seen that, while the complement-fixing titer of the virus sediment, resuspended to the original volume of the 10% tissue suspension and then further diluted with saline, varied somewhat in the different experiments, in each case except Experiment 4 the corresponding high-speed supernate showed a relatively high degree of activity even after passage through Seitz EK filter. This was true for both yolk-sac and mouse-lung suspensions. A sample of the Seitz filtered supernate described in Experiment 5 of Table I was sent to Dr. A. W. Grace of the New York Hospital, who found that in known lymphogranulomatous persons it produced a positive Frei reaction in dilutions through 1:320 (highest tested), while controls with normal yolk-sac antigen were negative in these individuals.

These observations, so markedly at variance with the findings of earlier workers, made it desirable to determine whether the considerable activity of Seitz EK filtrates was due to a residuum of virus

[5] Grace, A. W., Rake, G., and Shaffer, M. F., PROC. SOC. EXP. BIOL. AND MED., 1940, **45**, 259.

[6] McKee, C. M., Rake, G., and Shaffer, M. F., PROC. SOC. EXP. BIOL. AND MED., 1940, **44**, 410.

TABLE I.
Comparative Complement-Fixing Activity of Lymphogranuloma Virus Sedimented at High Speed* and Supernate Therefrom.

Exp. No.	Strain of virus	Source of virus	Material tested	Complement-fixing titer†
1	NH	Yolk-sac	Resuspended high-speed sediment	1:640
			Unfiltered " " supernate	1:320
			Seitz-filtered " " "	1:80
2	NH	Mouse-lung	Resuspended " " sediment	1:160
			Unfiltered " " supernate	1:80
			Seitz-filtered " " "	1:80
3	JH	Yolk-sac	Resuspended " " sediment	1:160
			Seitz-filtered " " supernate	1:40
4	G	Yolk-sac	Resuspended " " sediment	1:100
			Seitz-filtered " " supernate	1:10
5	NH + JH + G	Yolk-sac	Resuspended " " sediment	1:150
			Seitz-filtered " " supernate	1:50
6	JH + G	Yolk-sac	Resuspended " " sediment	1:150
			Seitz-filtered " " supernate	1:50

*The sediments and corresponding supernates were obtained by centrifugation at 12,000 RPM in the cold.

†The complement fixation tests were performed in Exp. Nos. 1, 2, 3 and 6 with serum K diluted 1:25, in Exp. No. 4 with serum LV No. 4 (of equal potency) diluted 1:25, in Exp. No. 5 with serum Z (more potent) diluted 1:50.

which had traversed the filters. Accordingly, several experiments were performed in which suspensions of heavily infected yolk-sacs were centrifuged for an hour at 2000 to 3000 rpm in the cold to throw down gross particles. A portion of the supernate was passed through a Seitz EK disc under negative pressure not exceeding 65 cm mercury. Filtrate, as well as unfiltered suspension, was then tested not only for relative complement-fixing titer but also for infectivity by yolk-sac inoculation of the chicken embryo, which is by far the most delicate known indicator of minute amounts of active lymphogranuloma virus. The results are given in Table II, where it will be seen that passage through Seitz EK filters reduced the infectivity of the suspensions by one- to ten-millionfold or more; in all but one case, no virus whatever was detectable when a total of several cc of undiluted filtrate was inoculated into eggs. Nevertheless, complement-fixing potency of the filtrates remained one-eighth to one fiftieth that of the unfiltered material, values comparable to those obtained with Seitz EK filtrates of high-speed supernates (Table I) when possible clogging of the filters with material in these suspensions, which have not been centrifuged at high speed, is taken into consideration. The Seitz EK filtrate in Experiment D of Table II was also

TABLE II.
Infectivity and Complement-Fixing Potency of Lymphogranuloma Virus Yolk-Sac Suspensions Before and After Seitz EK Filtration.

Exp.	Strain of virus	Material tested	Infectivity titer	Complement-fixing titer*
A	NH	Unfiltered suspension	10^{-7}	1:75
		Seitz EK filtrate	$<10^{-1}$	1:10
B	NH	Unfiltered suspension	2×10^{-7}	1:480†
		Seitz EK filtrate	$< 2 \times 10^{-1}$	1:10
C	JH	Unfiltered suspension	10^{-8}	1:75
		Seitz EK filtrate	10^{-1}	1:10
D	JH	Unfiltered suspension	5×10^{-9}	1:400
		Seitz EK filtrate	$< 5 \times 10^{-2}$	1:20

*The complement fixation tests were performed in Exp. A and B with serum Z diluted 1:50, in Exp. C with serum Z 1:100, in Exp. D with serum W 1:10.
†The period of fixation in Exp. B was longer than that usually employed.

submitted to Dr. A. W. Grace. In the highest dilution tested (1:120) it gave a strongly positive Frei response in known reactors who were at the same time negative to the control filtrate of normal yolk-sac.

The combined series of experiment shows clearly that the ability of Seitz EK filtrates, derived from rich suspensions of lymphogranuloma venereum virus, to fix complement specifically in the presence of lymphogranulomatous serum and to elicit a positive Frei reaction is due not to elementary bodies of the virus but to antigen in a less highly organized, possibly soluble, state. The failure of other investigators to obtain similar results is doubtless due to the low concentrations of virus present in the materials which they employed.

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Nature of the Capillary Permeability Factor Present in Extracts of Normal Tissues.

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Menkin[1] has shown that inflammatory exudates of various kinds have the property of increasing capillary permeability as demonstrated by the so-called trypan blue test in rabbits. This consists in

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1 Menkin, V., *Physiol. Rev.*, 1938, **18**, 366.