chapters that many teachers might desire to omit some entire chapters. The planning of these separate chapters is expected to afford wide freedom of choice for such omission, or for rearrangement, without interfering with the student's progress. The chief claim to novelty lies in the wide variety of fields from which verbal problems have been gleaned. The text has been prepared with somewhat more than average care, although the reviewer notes several definitions and formal explanations which seem not above criticism. Answers are provided for odd-numbered problems. Approximately the last hundred pages are devoted to numerical tables, reference formulas and (somewhat incomplete) index.

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SPECIAL ARTICLES

AN INFECTIOUS AGENT FROM CASES OF ATYPICAL PNEUMONIA APPARENTLY TRANSMISSIBLE TO COTTON RATS[1]

RECENTLY a primary atypical pneumonia of unknown etiology has been a rather common disease.[2] Observations made in this laboratory since March, 1941, suggest that in some cases of this disease an infectious agent is transmissible to cotton rats (*Sigmodon hispidus*) and produces pulmonary consolidation after the first intranasal inoculation of sputum or lung under ether anesthesia. Both the eastern cotton rat (subspecies *hispidus*) and the western cotton rat (subspecies *eremicus*) are susceptible.

The results with material from a total of 78 cases of atypical pneumonia are summarized in Table 1.

TABLE 1

RESULTS OF INOCULATING COTTON RATS WITH SPUTUM OR LUNG TISSUE FROM CASES OF ATYPICAL PNEUMONIA

	Days after onset	Number of specimens causing lung lesions	Number of specimens causing no lung lesions
Sputum	5 or less	8	11
Sputum	6 to 9	4	15
Sputum	10 or more	1	19
Sputum	unknown	2	9
Lung	.....	2	7
Total		17	61

Similar material gave negative results in mice, ferrets, hamsters and other animals. Sputums taken early in the disease often produced lung lesions rather consistently when several cotton rats were inoculated with the same specimen. Fully grown or old animals were more susceptible than those 3 to 7 weeks of age. Of the total of 131 cotton rats receiving material from cases of atypical pneumonia 35 developed lung lesions. Thirty-four control cotton rats inoculated intranasally with throat washings from cases of influenza or with heated sputum, horse serum broth or other materials did not develop significant lung lesions. All animals were sacrificed 7 days after inoculation. Only one out of more than 50 cotton rats used in experiments not connected with atypical pneumonia has shown lung lesions at autopsy.

By serial intranasal passage of lung suspensions from animals which had lesions on the first passage, strains of an infectious agent from 6 cases of atypical pneumonia were adapted to cotton rats. In 2 cases this adaptation was repeated, starting from the original sputum, but using cotton rats of a different subspecies. After 4 to 6 passages the adapted strains produced gross evidence of lung involvement in over 90 per cent. of the animals inoculated, but seldom caused death. With sputum from 11 cases lung lesions were produced on first inoculation, but no adaptation by serial passage was obtained. When the lungs of normal cotton rats or of animals which developed no lesions after inoculation of sputum were passed serially, the results were uniformly negative.

The lung lesions were patchy red-gray with maximum intensity at 6 to 8 days. Microscopic examination of sections of lungs showed an infiltration of the septa with polymorphonuclear leucocytes and mononuclear cells and hyperplasia of the alveolar epithelium. No inclusion bodies, elementary bodies, rickettsiae or visible microorganisms were seen in sections or in impression smears stained by the methods of Gram, Giemsa or Macchiavello. Cultures on blood agar and horse serum broth were negative. In 2 out of 6 filtration experiments using Berkefeld N candles passage of the agent was demonstrated.

Strains which had been adapted to cotton rats produced lung lesions after intranasal inoculation of Syrian hamsters (*Cricetus auratus*), but caused no detectable disease in mice, rabbits or guinea pigs. Animals which had recovered (in about 14 days) from intranasal inoculation of the pneumonia agent were solidly immune to reinoculation by the same route. Infected cotton rat lung produced neither illness nor

[1] The studies and observations on which this paper is based were conducted with the support and under the auspices of the International Health Division of The Rockefeller Foundation in cooperation with the California State Department of Public Health. Most of the material for laboratory studies was obtained through the courtesy of physicians at the Medical Center and the Cowell Memorial Hospital of the University of California.

[2] For literature review and references see J. H. Dingle and M. Finland, *New England Journal of Medicine*, 227: 378, 1942.

immunity when inoculated intracerebrally or intraperitoneally into cotton rats.

Neutralization tests with serum-virus mixtures incubated for 1 hour at 37° C. and then kept in the icebox overnight were performed by inoculating cotton rats and hamsters intranasally. Sera from hyperimmunized cotton rats, hamsters and rabbits gave definite neutralization of the agent. Partial or irregular neutralization was observed with sera of human beings convalescent from atypical pneumonia, with sera of cotton rats inoculated once only or of guinea pigs inoculated repeatedly with cotton-rat adapted strains, and with sera from rabbits immunized with human lung infectious for cotton rats.

By cross-inoculation and neutralization tests antigenic relationships between 6 established strains were demonstrated. Cotton rats immunized by two successive intranasal inoculations with adapted strains were solidly immune to reinoculation with a specimen of infectious human lung which produced marked lesions in the controls. Cotton rats immunized with human material were partially resistant when tested with adapted strains.

During the course of serial passages from cotton rats which developed lung lesions on the first inoculation, two strains not antigenically related to those just described were obtained. These two "aberrant" strains may have been carried by the cotton rats and had apparently replaced the agent present in the first passages.

The appearance of non-bacterial lung lesions in cotton rats after inoculation of material from cases of atypical pneumonia suggests that a virus-like agent was transmitted and established by serial passage. The strains adapted to cotton rats were related to the agent in human material by cross-immunity tests. This agent, which is presumably a filterable virus, differs from the psittacosis-like virus previously described[3] and also from other known viruses which can infect cotton rats by the intranasal route. At present the evidence for the causal relation of this agent to the most common form of atypical pneumonia must be considered incomplete because of irregularities in the neutralization tests, particularly those with human serum. Further investigations on the influence of the amount of the infecting dose on the neutralization test are in progress.

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[3] M. D. Eaton, M. D. Beck and H. E. Pearson, *Jour. Exp. Med.*, 73: 641, 1941.

DESTRUCTION OF HYPERTENSIN AND PEPSITENSIN BY AN AMINOPEPTIDASE OBTAINED FROM YEAST

THE vasoconstrictor properties of hypertensin (angiotonin) and pepsitensin—a substance formed by the digestion of proteins with pepsin—can be entirely destroyed by an aminopeptidase (a.p.) enzyme obtained from yeast and purified by the Johnson method.[1]

These two hypertensive substances incubated with that enzyme at 38° and neutral pH. lose their vasoconstrictor properties in a few minutes.

Approximately 0.01 cc of the final purified solution obtained from 2 kg of compressed yeast destroys them after 5- to 10-minutes incubation (2 or 3 units of hypertensin or pepsitensin). The degree of destruction of these two products under the influence of the enzyme was controlled by the method previously described, using the Loewen Trendelenburg test and the arterial pressure of the cat.[2]

The mixture of hypertensin or pepsitensin with the enzyme was injected after different periods of incubation and the vasoconstrictor or pressor effects obtained were compared with those produced by an equal dose of substrate and the enzyme mixed immediately before injecting. Sometimes as a means of controlling the results, a mixture of enzyme was used, incubated for the same length of time, and previously inactivated by boiling with the vasoconstrictor substance (Fig. 1).

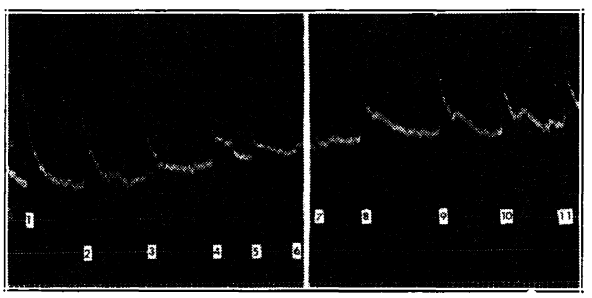

FIG. 1. Arterial pressure increase produced by: 1: 0.4 cc hypertensin; 2,3,4,5,6: 0.5 cc hypertensin incubated 5′ with 0.0025, 0.005, 0.01, 0.02 and 0.03 cc of a.p. 676 F respectively 7: 0.5 cc water with 0.03 cc a.p. 672 F 8,9,10,11: 0.5 hypertensin.

Titration of polypeptid nitrogen during incubation showed a progressive and considerable diminution. A similar result was obtained when renal hypertensinase acted upon hypertensin or pepsitensin.

The hydrolytic activity of yeast enzyme on synthetic substances allows one to classify this enzyme as a.p.[3] Hypertensinase extracts of pig kidneys, purified by

[1] M. J. Johnson, *Jour. Biol. Chem*, 127: 575, 1941.
[2] H. Croxatto and R. Croxatto, SCIENCE, 42: 101, 1942.
[3] J. S. Fruton, G. W. Irving, Jr. and M. Bergmann, *Jour. Biol. Chem.*, 141: 763, 1941.