this pneumonic area. In general, the pathological process found in the lungs of this group of animals differed from that observed in the second group of rabbits in that the nodules were considerably larger and contained more abundant caseous material. Microscopical examination of the material from nodules was negative for *Monilia,* but culturally *Monilia tropicalis* was obtained in 2 instances.

These results suggest the conclusion that previous sensitization renders rabbits susceptible to the subsequent intratracheal administration of *Monilia tropicalis.* The lung process which may be designated as experimental bronchomoniliasis is confined to the formation of multiple firm nodules containing caseous material. The positive culture examination of this material indicates that the development of lung lesions is dependent upon the pathogenic activity of the fungus. A similar experiment with passively sensitized rabbits gives an essentially negative result. It is also shown that old fungous culture rich in filamentous growth is somewhat more effective in the production of lung lesions than a young culture containing mostly budding cells.

7116 C

Stimulating Effect of Alum and T.A.B. Vaccine in Tetanus Prophylaxis.

W. K. CHEN. (Introduced by C. E. Lim.)

From the Department of Bacteriology and Immunology, Peiping Union Medical College.

Ramon and Zoeller[1] in their study of active immunization against tetanus with tetanus toxoid have referred to the use of an immunizing agent consising of toxoid plus T.A.B. vaccine on human beings in the first dose to be followed next by toxoid alone. Glenny[2] by the addition of potash alum to tetanus toxoid produced on experimental animals far better results than the use of the original toxoid and he attributed the increased antigenic response of alum-toxoid to delayed absorption. As alum is very irritating to the tissues, it may cause much discomfort when applied to human beings; while T.A.B. vaccine associated with toxoid will have the advantage of serving the dual function of preventive against enteric fever

[1] Ramon, G., and Zoeller, C., *C. R. Soc. de Biol.,* 1929, **100,** 92.

[2] Glenny, A. T., *Brit. Med. J.,* 1930, **2,** 244.

and also a stimulus to the antigenic value of tetanus toxoid, should its stimulating effect be established as equal to that of alum. The present experiment was carried out with the object of comparing this property of alum and T.A.B. vaccine in tetanus immunization on guinea pigs.

Three groups of guinea pigs weighing about 300-350 gm. were immunized subcutaneously by 3 injections of 0.5 cc. doses of 3 kinds of antigen mixture at 14 day intervals. The first group was immunized with tetanus toxoid and T.A.B. vaccine mixture of equal parts. The second group immunized with tetanus alum-toxoid mixture (0.25% alum forming the most precipitation by titration) and the third immunized with alum toxoid (0.25% alum) and T.A.B. vaccine mixture of equal parts were utilized for comparison of their antigenic efficiency. One month after the last injection all the immunized animals (weight increased to 450-550 gm.) were tested against fresh tetanus toxin as well as the whole culture without filtration.

All the materials used in the experiment were supplied from the National Epidemic Prevention Bureau, Peiping. The tetanus toxoid was prepared by addition of 4 cc. formalin per liter of fresh tetanus toxin containing 5,000 M.L.D. per cc. and after one month incubation at 37°C. tested on guinea pigs for toxicity. The tetanus toxin was prepared by inoculating a 48-hours growth of *B. tetani* in glucose agar to 1% glucose broth (pH 7.0) under vaseline and incubating for 10 days before filtration through Berkefeld "V" candles. The triple typhoid vaccine (T.A.B.) was a saline suspension of heat-killed bacteria of 500 million *B. typhosus* and 250 million each of *B. paratyphosus* "A" and "B" per cc. with 0.25% tricresol added as preservative. The L+ dose of the toxin and the M.L.D. of the whole culture were determined preliminarily on white mice of about 20 gm. in weight and also on guinea pigs of 350 gm. in weight with dosage of toxin 10 times the mouse dose. In the determination of L+ dose 1/5 of an international unit of standard tetanus antitoxin was mixed with different amounts of toxin before injection into the animals.

From the results summarized in Table I, it is evident that the antigenic efficiency of the tetanus toxoid and T.A.B. vaccine mixture is equal to that of the alum-toxoid which has been proved by many workers as the best antigen for active immunization. Also further addition of alum to the mixture of toxoid and T.A.B. vaccine does not increase the antigenic value of the original simple combination of toxoid and T.A.B. vaccine. All the animals immunized with the

TABLE I.
Active Immunization Against Tetanus.

Antigen for immunization	No. of animals	Test dose	Results
Tetanus toxoid and T.A.B. vaccine	5	100 L+ dose toxin	All survived
	"	200 " " "	" "
	"	400 " " "	1 "
			4 died
	"	10 cc. whole culture	4 survived
			1 died
Tetanus alum-toxoid	4	100 L+ dose toxin	All survived
	5	200 " " "	" "
	"	400 " " "	2 "
			3 died
	4	10 cc. whole culture	3 survived
			1 died
Tetanus alum-toxoid and T.A.B. vaccine	4	100 L+ dose toxin	All survived
	"	200 " " "	" "
	5	400 " " "	1 "
	4	10 cc. whole culture	4 died
			All survived
Control	2	1/100 L+ dose toxin	Died 36 hr.
	"	1/200 " " "	" 3 days
	"	0.0002 cc. culture	" 48 hr.
	"	0.0001 " "	" 4 days

above three kinds of antigen mixtures possess a very high protective power against tetanus toxin over 200 L+ doses (or over 40,000 M.L.D.) and most of them can tolerate even a 10 cc. subcutaneous injection of a 10-day whole tetanus culture (toxin with spores).

Conclusions. The stimulating effect of T.A.B. vaccine in tetanus toxoid is established in the present experiment in comparison with alum-toxoid which has hitherto been recommended as the best preparation for active immunization but is unsuitable for human beings on account of its irritating action. The associated T.A.B. vaccine in tetanus toxoid is found to have the advantage of serving the dual function of a preventive against enteric fever and also a stimulus to the antigenic property of tetanus toxoid. In actual application to human cases, especially soldiers during war time, active prophylaxis against tetanus is preferable as it gives far better protection than immunization with specific antiserum. The combination of tetanus toxoid and T.A.B. vaccine for the double prophylaxis against tetanus and enteric fever by a series of 3 injections is, therefore, to be recommended to military hygienists for further trial.