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ABSTRACT

Adenovirus Type 4 continues to be the main agent associated with Acute Respiratory Disease at NTC, Great Lakes, Illinois.

Studies were carried out comparing the effect of live oral monovalent adenovirus Type 4 (LAV) in enteric-coated capsules with inactivated vaccines (IAV) on the prevention of such diseases.

In addition, a special company study was made on the effect of these treatments on the isolation and serological responses to vaccine and natural adenovirus infections. Fifty percent of men fed LAV shed virus rectally within 5 days and 90% within 14 days. Average duration of virus excretion was about 8 days. However, less throat adenovirus was detected in LAV men than in IAV or placebo groups. Moreover, when positive, LAV subjects shed throat adenovirus for less time than the other subjects. Less men in the IAV groups were positive for rectal adenovirus than in the placebo group.

Human embryonic kidney (HEK) tissue cultures appear to be more sensitive than H.Ep.-2 or WI-38, for adenovirus isolation. The latter two were of similar sensitivity.

High adenovirus infection rates occurred in most vaccinated subjects before significant changes occurred in neutralizing antibody status. The measurement of this antibody varied with the specific adenovirus Type 4 antigen and with the tissue culture host system used. Antigens prepared from LAV adapted to HEK showed a greater sensitivity than other neutralization test systems used.

All of the vaccines were effective in reducing Acute Respiratory Disease hospitalization by the 11th day after vaccination. All were about equally effective in reducing these admissions by 50%. The determination of the exact specific protective effect afforded by these vaccines was confounded by the presence of other etiological agents in the population, including influenza A2. None of the vaccines had any appreciable affect on Acute Respiratory Disease that did not require hospitalization.

LIVE ADENOVIRUS VACCINE IN A NAVY RECRUIT COMPANY, NTC, GREAT LAKES, ILLINOIS, 1965

INTRODUCTION:

Initially, the study proposed for use of live Type 4 adenovirus vaccine (LAV) in naval recruits was of two-phase design. Phase I was arranged to determine spread of vaccine virus to cohorts who were given placebo inoculations, as well as to observe untoward vaccine reactions.

Phase II, a much larger study, was designed to compare protection of LAV with inactivated monovalent (Type 4) and bivalent (Types 4 and 7) adenovirus vaccines prepared in monkey kidney tissue cultures. The results of the latter study are presented elsewhere.[1]

However, Phase I was held in abeyance because results of serological surveillance studies, which continually monitor the recruit population, indicated a high rate of adenovirus disease at the time when the LAV program was being contemplated. These natural adenovirus infections would have greatly complicated interpretation of data of LAV spread. Instead, a company study of live and inactivated vaccines was initiated to: (a) attempt to determine adenovirus excretion paterns during a high infection period; (b) to find possible markers to distinguish vaccine virus from wild, and (c) to compare effect of antibody response to LAV and inactivated adenovirus vaccines (IAV) on adenovirus excretion patterns in naval recruits.

MATERIALS AND METHODS:

Population and treatment: A company of newly arrived naval recruits (84 men) was randomly divided into the following four treatment groups:

(a). Twenty-one men received an oral live adenovirus vaccine. This material was secured by Dr. G. G. Jackson from the Board for Vaccine Development, NIAID, NIH. The vaccine was manufactured by Wyeth Laboratories, and contained adenovirus Type 4 isolated and passaged in human cell cultures, of primary and diploid nature[2] (WI-38). Virus material was lyophilized and placed in an enteric coated capsule. All such material was kept at -20°C until given to study subjects.

(b). Twenty-one men received a monovalent Type 4 adenovirus vaccine produced in monkey kidney tissue culture cells by Dr. A. J. Morris, Division of Biological Standards, National Institutes of Health.

(c). Twenty-two men received a licensed, bivalent, adenovirus Types 4 and 7 vaccine, produced for military use by Wyeth Laboratories.

(d). The remaining twenty-one men received a saline placebo preparation.

Recruits were formed into the company approximately on arrival at Great Lakes for recruit training, and received the above treatments 4 days after company formation.

Sampling for virus isolation: Throat and rectal swabs, immersed in a diluent of veal infusion broth with 0.5% bovine serum albumin and antibiotics were obtained from all men remaining in the company. Specimens were collected approximately 3 times a week for 21 days after treatment and thence, once weekly for the 6 remaining weeks of recruit training.

Approximately 2600 specimens were inoculated into duplicate tissue of cultures of H.Ep-2 cells. Six hundred and forty specimens equally selected from LAV and placebo groups were inoculated into WI-38 human diploid cell cultures, originally obtained from Dr. Leonard Hayflick, for comparison of cultural characteristics and tissue sensitivity of vaccine and wild virus with the continuous H.Ep-2 cultures.

All inoculated cell cultures were observed 3 times weekly for evidence of cytopathology, and most specimens received 2 or 3 "blind" passages in appropriate cell cultures.

Identification of viral isolates was done, presumptively, by complement-fixation tests, or by hemagglutination[3] with rat or monkey erythrocytes. Confirmation was done by microplate neutralization test[4] with adenovirus Type 4, type-specific antisera produced in rabbits against WI-38 adapted vaccine virus or Great Lakes adenovirus Type 4 prototype.

Sampling for sera: Blood specimens were obtained from all available study subjects before treatment and after at weekly intervals for 4 weeks, and at the 40th and 62nd day. Sera from such specimens were tested by microplate complement-fixation tests using 4 units of adenovirus group antigen,[5] or by microplate tissue culture neutralization test[4] using 10-30 TCD_{50} of the following antigens pre-incubated for 1 hour at room temperature with equal amounts of 2-fold serum dilutions: H.Ep-2 adapated vaccine Type 4 adenovirus (LAV), Great Lakes prototype adenovirus Type 4 (NMRU-62), "Wild" adenovirus Type 4 (NMRU-65), and Great Lakes adenovirus Types 3 and 7 prototype. Serum antibody titer endpoint was that dilution showing less than 25% CPE after 3 days' incubation at 36°C. In addition, most serum samples were tested against 10-30 TCD_{50} LAV antigen in neutralization tests using microplate tissue cultures of secondary human embryonic kidney.[6] Results of these tests were noted after 5 days' incubation at 36°C.

RESULTS:

Infectivity titer of LAV: Samples of the LAV capsules were titrated in various cell cultures to determine and confirm reported infectivity titers.

Results shown in Table I indicate that human embryonic kidney (HEK) has the greatest sensitivity to LAV infection of all the cells tested. Maximal titers of $10^{6.5}$ in HEK were achieved at 21 days and no further increase was noted. Diploid cells (WI-38) also produced maximal virus infectivity titers at 21 days, but titers were about 1.5 log lower ($10^{5.0}$) in these cells than in HEK. Rhesus monkey kidney tissue culture cells had LAV infectivity titers of only $10^{3.0}$, while H.Ep-2 cell cultures indicated little, if any, sensitivity for LAV upon primary passage.

Vaccine reactions: Little or no untoward reaction to LAV or IAV vaccines was noted. Men were interviewed 3 times a week for 3 weeks and weekly for 6 weeks afterward. Recruits were encouraged to seek treatment at any time during the course of the study. Illness history of this company was comparable to non-treated cohort companies concurrently undergoing training.

Virus isolation: Results of virus isolation attempts from specimens from men in the 4 treatment groups is shown in Tables II-V, and is summarized in Table VI. It can be observed (Table II) that men fed LAV shed adenovirus rectally beginning on day 3 after feeding. Most men showed decreased rectal excretion of such virus after day 14 and only scattered isolates were found after day 16. In contrast, the rectal adenovirus excretion patterns of IAV and placebo subjects shown in Tables III-V, with few exceptions, generally begin about day 13 and end about day 28. Inspection of numbers and distribution of throat isolates shown in Tables II-V show fewer numbers of adenovirus Type 4 recovered from the men in LAV groups than in placebo, with those from the IAV groups somewhere between the two. Moreover, LAV subjects have a shorter duration of shedding of throat adenovirus than do placebo recruits.

The summary of isolation data in Table VI shows that 24.1% of all rectal specimens and 95% of the "LAV" men were positive for adenovirus Type 4 isolates. This compares to 21% of rectal specimens and 100 of men positive in the placebo group. However, only 8.7% of throat specimens and 75% of "LAV" men were adenovirus-positive, whereas, 16.4% of specimens and 86% of placebo men excreted throat adenovirus. IAV recruits show throat virus shedding pattern intermediate between the LAV and placebo groups.

Of all the total viruses isolated, adenovirus Type 4 was in preponderance. However, it is interesting to note the early incidence of a small amount of Herpesvirus in all groups, as well as Respiratory Syncytial virus.

The maximal average duration of viral excretion in rectal or throat specimens from LAV subjects, as interpreted by extrapolation of observed data, is shown in Table VII. It can be seen that the average duration of viral shedding, shown by rectal sampling of LAV subjects, is about 8 days, whereas, data from IAV and placebo groups is somewhere between 5-6 days. However, the data for duration of throat virus excretion is only 2.9 days as compared to 4-5 days for the respective groups.

A comparison of the sensitivity of various tissue culture cells is shown in Table VIII. The results of isolations from the routine surveillance program do not indicate any difference in the sensitivity of either H.Ep-2 or WI-38 cells for isolation of adenovirus Type 4 (84% positive). However, rhesus monkey kidney cells show a decreased sensitivity for adenovirus (18%). Furthermore, when recovery of adenovirus Type 4 is compared in H.Ep-2 or WI-38, a similar isolation rate can be seen. However, HEK cell cultures appear to have a somewhat greater sensitivity than either of the other cell cultures. It is interesting to note that the discrepancy of adenovirus Type 4 isolates from LAV group throat specimens to placebo (6-7% LAV as 16-17% placebo) in WI-38 or H.Ep-2 cells is not as great (19% LAV as 29% placebo) when HEK cell cultures are used.

Figures 1 and 2 represent the kinetics of viral recovery from throat and rectal specimens of the various treatment groups, respectively. It can be seen that the curves are virtually identical insofar as onset of linear accumulation of throat virus recovery on day 7 post-treatment. Maximum day and absolute percentage of virus recovery varies somewhat from group to group with the placebo accruing to 85% of men excreting virus by day 28 and the other groups showing somewhat less proportion of men (71-75%) shedding virus. It is interesting to note that the approximate cessation of the rate of linear virus accumulation appears to differ considerably for the various groups with the Bivalent group decreasing sharply by day 15; Monovalent, day 17; LAV, day 18; and Placebo, day 21.

Figure 2 shows a similar pattern for rectal adenovirus excretion. However, in these specimens there is remarkable difference in rate of rectal virus shedding as well as absolute difference in percentage of men infected. It can be seen that LAV subjects appear to commence excreting adenovirus within 24-48 hours after feeding and end linear accumulation within 9 days. Placebo subjects begin virus shedding about day 5 and terminate virus accrual at constant rate by day 21. The IAV groups have a considerably slower rate of throat virus acquisition and a smaller percentage of men shedding adenovirus rectally (95-100% LAV or Placebo as 75% IAV groups).

Serology: Results of neutralization tests for adenovirus Type 4 antibody for each treatment group are shown in Figures 3-6. Tests performed in human embryonic kidney using "LAV" virus for all treatment groups are shown in Figure 7. It can be observed that although dosage of virus antigen was similarly adjusted for each test (10-30 doses) significant difference in absolute titers occurs between viruses used and according to cell system in which the test was performed.

Antibody responses for the prototype (NMRU-62) adenovirus Type 4 were uniformly low for all treatment groups. Considerably higher titers are obtained by using "LAV" H.Ep-2 adapted virus antigen. That this difference is due to individual virus difference and not a specific antibody avidity characteristic of the "LAV" strain is shown by the fact that when a "Wild" 1965 strain (NMRU-65) is used, antibody titers similar in magnitude to those against the LAV antigen are obtained.

However, striking results in increased antibody titer is found when "LAV" antigen is used in HEK cells (Figs. 3-6).

Figure 7 shows geometric mean neutralizing antibody titers of all the treatment groups as determined by "LAV" antigen. It appears that the speed of response is similar for all 3 vaccine groups with the antibody levels of the monovalent group appearing the fastest. Extrapolation from these curves indicate that an average antibody titer of 1:32 is achieved in about 10 days for the monovalent group; 12 days for the LAV group; 17 days for the bivalent group; and at 24 days for the placebo group. It is interesting to note that antibody titers for the IAV groups increased throughout the 62 days of sampling, whereas, the LAV and placebo groups showed no further increase after 28 and 40 days, respectively.

Figures 8 and 9 depict heterologous and homologous neutralizing antibody responses of the various treatment groups. Except for a slight increase in heterologous antibody titer, only the bivalent vaccine groups appear to have significantly altered the status of antibody in the company study subjects. It is interesting to note the significant heterologous response to adenovirus Type 3 in men receiving the bivalent preparation, and is thought to be either due to provocation of such responses by both Types 4 and 7 adenoviral antigens or to the inclusion in the vaccines of adenovirus Type 3 antigen of insufficient potency to be approved for labeling, but nevertheless capable of contributing to the over-all Type 3 antibody response.

Table IX summarizes both the adenovirus isolation and geometric mean antibody responses for individual treatment groups. By comparing the time of adenovirus isolation rates (throat or rectal) of approximately 50% with the time of significant change (4-fold or greater increase) in antibody titer, some conclusions as to expected protection can be made.

If the placebo group is taken to represent rates of natural infection, then it is apparent that approximately 50% of such infections occurred within 14 days after the study was initiated. Antibody response to these infections occurs about 14 days later (21-28th day). Men fed LAV capsules have a 50% throat virus isolation rate at about the same time, but also have already established a significant antibody response to the oral vaccine at this time. This effect is also found in subjects of the monovalent group who have the highest antibody titer (1:62) of all groups at this time. The antibody response to bivalent vaccine appears to follow the 50% level of throat virus acquisition.

It should be noted that I/ with 50% or greater isolation rate of rectal adenovirus is delayed one eek later than placebo men, although they do not differ in relative rate of throat virus isolation.

DISCUSSION AND SUMMARY:

Although no differentiating markers were detected between the "LAV" and "Wild" adenovirus isolated during the study, a slight difference in the sensitivity of HEK tissue culture for these agents was observed. Further study will be necessary to determine if any significance can be attached to this observance.

Further, a difference in sensitivity of adenovirus for antibody was observed between the "LAV" and "Wild" adenovirus Type 4 antigens. However, this difference was not confined to the LAV preparation since at least one other adenovirus Type 4 antigen selected at random from pre-study isolates exhibited the same avidity for antibody as the "LAV" antigen.

In the absence of differentiating markers, it cannot be stated with certainty that spread did not occur, although studies in marine recruits elsewhere have repeatedly shown this phenomenon when men are vaccinated in non-epidemic periods of adenovirus Type 4 infections. However, it appears unlikely that the potential for such spread is not possible. Many cycles of multiplication of initially potent virus occur in LAV vaccinated subjects who live in intimate proximity with non-vaccinated cohorts. Ample opportunity for aerosols of high titered adenovirus Type 4 to infect contacts occur daily in latrines. One would have to conclude either that aerosols are not the optimal method for dissemination of the LAV virus or that its affinity for site of tissue multiplication has been altered from that of the natural oropharynx cavity to the enteric site. Of further and probably greater importance are the factors which enable virus to spread from human to human under one environment may not be present at another. Previous data[7] has shown that adenovirus infection rates among marine recruits at San Diego are considerably lower than among navy recruits at the same training center. Differences in environmental and training procedures were concluded to be the major factor for the differences in adenovirus disease rates in the two military groups. Further studies are now underway to obtain more information in this respect. Although the number of subjects is small, it would appear from the data obtained from the neutralization tests performed, that antibody responses elicited by inactivated vaccines were comparable to LAV. In fact, the higher titers obtained by IAV may be due to booster responses of infections in men who developed insufficient antibody to prevent adenovirus infection, but sufficient to result in high secondary response.

Less adenovirus was recovered from throat specimens of LAV than Placebo subjects. In addition such viruses were shed for a shorter time in the former group. Although no difference in rate of excretion of throat adenovirus was observed between IAV and placebo groups, considerable decrease in rectal excretion was obtained. The significance of this observation is not clear unless humoral antibody of the gut is in greater concentration or availability than in the oropharyngeal cavity and thus is capable of aborting or limiting such infections to the upper areas.

It is evident from the data presented that adenovirus Type 4 infections appear to begin very shortly after men arrive at Great Lakes. Thus, any vaccine (live or inactivated) must be given as soon as possible to be effective. In fact, for dramatic vaccine effect, men should be well endowed with sufficient antibody before arriving at the endemic area.

REFERENCES

1. Naval Medical Research Unit No. 4: Annual Report to the Commission on Influenza, Armed Forces Epidemiology Board, 1964-1965.

2. Hayflick, L. and Moorhead, P.S.: The Serial Cultivation of Human Diploid Cell Strains. Exp. Cell Res. 25:585-621, 1961.

3. Rosen, L.: A Hemagglutination-inhibition Technic for Typing Adenoviruses. Am. J. Hyg. 71:120-128, 1960.

4. Rosenbaum, M.J., Sullivan, E.J., Phillips, I.A. and Edwards, E.A.: A Simplified Method for Virus-Tissue Culture Procedures in Microtitration Plates. Proc. Soc. Exper. Biol. and Med. 113:224-229, 1963.

5. Sever, J.L.: Application of A Microtechnique to Viral Serological Investigations. J. Immunol. 88:320-321, 1962.

6. Sullivan, E.J. and Rosenbaum, M.J.: Further Observations of Microplate Tissue Cultures. Bact. Proc. 1964, p. 134.

7. Arlander, T.R, Pierce, W.E., Edwards, E.A., Peckinpaugh, R. O. and Miller, L. F.: Epidemiology and Prevention of Acute Respiratory Disease in Naval Recruits. Iv. An Epidemiologic Study of Respiratory Illness Patterns in Navy and Marine Corps Recruits. Am. J. Pub. Health 55:67-80, 1965.

TABLE I

TISSUE CULTURE INFECTIVITY OF LAV CAPSULES* USED IN 1965 STUDY

Capsule	Tissue culture infective dose$_{50}$**									
		HEK				WI-38			H.Ep-2	MK
	Day:	3	7	14	21	3	7	14	21	14	14
I (LAV)		<1	3.5	5.0	6.5	<1	2.5	3.5	5.0	<2.0	3.0
II ("A")		<1	2.5	4.5	6.5	<1	2.0	3.0	5.0	ND	ND

* 60 mgm capsule reconstituted in 0.94 ml 199

**Calculated by method of Karber - neg. $\log_{10}$ of dilution 50% CPE endpoint

TABLE II

Virus Isolates from Recruits Immunized with Live Oral Adenovirus Type 4 Vaccine -- "LAV" Company, Great Lakes, Illinois, 1965

Recruit No.	Sampling Day Post Immunization												
	1	3	5	7	9	13	14	16	19	21	28	35	62
03			/A	/A	/A	A/			A/A				
06			/A	/A	/A	A/A	A/A	/A					
08					/A	/A	/A				/A		
23		/A	/A	/A	/A		A/A						
26			/A	/A	/A	/A	A/A						
30												/A	
37	H/	/A	H/A	/A	/A	/A					A/A	A/	
41				A/								A/	
44							/A				RS/		
47					/A	/A	/A	A/					
52			/A		/A	/A	/A	ND/	A/	A/			
53						/A	/A	/A	A/	A/			
58		/A	/A	/A	/A	/A	/A	A/					
60			/A	/A	/A	A/A	A/	A/A	A/	/A			P/
61				/A	/A		/A						
62			/A	/A	/A	/A	/A						
63			/A	A/A	ND	/A	/A						
64				/A	/A	/A		A/					
76					/A	/A	/A	/A			A/		
79				/A	/A	A/A		A/	A/				

T/R = positive virus isolate, throat/positive virus isolate, rectal

A = Adenovirus; H = Herpes

RS = Respiratory syncytial

P = Polio; ND = Not done

TABLE III

Virus Isolates from Recruits Immunized with Inactivated Monovalent Type 4 Vaccine -- "LAV" Company Great Lakes, Illinois, 1965

Recruit No.	Sampling Day Post Immunization												
	1	3	5	7	9	13	14	16	19	21	28	35	40
04						/A	/A	A/A	A/A	A/A			
05					A/	A/	A/	A/A	A/A	/A			
09								A/A	A/A	/A	H/		
13								/A	/A	/A	/A	/A	
15											/A		
18													
22	H/	H/	H/	H/		A/							/A
33						A/A	A/A	A/A	A/A	A/A			
35													
36	H/				A/			/ND					
39							A/	A/	A/A	/A			
43					A/A	A/A	A/A	A/A					
46								A/	/A	/A			
48								A/A	A/A	/A	/A		
50								A/	A/	A/A	/A		
58								A/	A/A	A/A			
65	A/				A/	A/	A/	A/	A/				
67				A/				H/					
72								/A	ND		ND	ND	ND
74	/A	/A	/A	/A	/A						/A		RS/
83						A/A	A/A	A/A	A/A	/A			

T/R = positive virus isolate, throat/positive virus isolate, rectal

A = Adenovirus; H = Herpes

RS = Respiratory syncytial

P = Polio; ND = Not done

TABLE IV

Virus Isolates from Recruits Immunized with Inactivated Bivalent (adenovirus Type 4 and 7) Vaccine -- "LAV" Company
Great Lakes, Illinois, 1965

Recruit No.	Sampling Day Post Immunization												
	1	3	5	7	9	13	14	16	19	21	28	35	4
02						/A	A/	A/A	A/A				
11		ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
12						A/A	A/A	A/A	A/A	/A			
14										A/A	/A		
16											/A	A/	
17													
19									A/	A/A	A/A		
24						A/	/ND		A/A	A/			
27													
31						A/	A/	A/	A/A	/A			
38					/A	A/A	A/A	/A	/A				
40													
42											/A		
45						/A							
59	H/			H/									
66			/A	/A	/A	A/A							
69							A/	A/	A/		/A		
71	A/												
73						A/A	A/A	A/A	A/A			RS/	
77					A/A	A/A	A/A	A/A	A/A				
82								/A			A/A	/A	
84						A/			/A				

T/R = positive virus isolate, throat/positive virus isolate, rectal

A = Adenovirus; H = Herpes

RS = Respiratory syncytial

P = Polio; ND = Not done

TABLE V

Virus Isolates from Recruits Inoculated with Saline Placebo --
"LAV" Company
Great Lakes, Illinois, 1965

Recruit No.	Sampling Day Post Immunization												
	1	3	5	7	9	13	14	16	19	21	28	35	41
01					A/A	/A	/A	/A	A/A	A/A			
07		A/	A/A	A/	A/	/A							ND
10										/A			
20	H/								A/A	A/A	A/A	/A	
21										/A	A/A	/A	
25								A/	A/	/A			
28				/A			A/	A/A	A/A	/A			
29								A/A		A/	/A		
32	H/							/A			A/	/A	
34						A/	A/	A/A	A/A	/A	ND	ND	ND
49						A/	A/	A/A	A/A	A/A	A/		
51					/A	A/A	A/A	A/A	A/A				
54							/A	A/	A/				
55	RS/					/A	/A	/A	A/A				
57										A/	/A		
68					A/	A/A	A/A	A/A	A/A				
70						/A			/A				
75						A/A	A/A	A/A	A/A	/A			
78						/A	A/A	A/A	A/A	/A			
80						/A	/A	/A	/A	/A	/A		
81	A/				/A	A/A	A/A	/A	A/A			A/	RS/

T/R = positive virus isolate, throat/positive virus isolate, rectal

A = Adenovirus; H = Herpes

RS = Respiratory syncytial

P = Polio; ND = Not done

TABLE VI

SUMMARY OF ADENOVIRUS ISOLATIONS FROM LAV STUDY IN A NAVAL RECRUIT COMPANY, NTC, GREAT LAKES, ILL., FEB-MARCH 1965

Vaccine group (no. men)	Specimen (number)	Percent specimen positive	Percent recruits positive
LAV (20)	Rectal (320)	24.1	95
	Throat (320)	8.7	75
Bivalent (21)	Rectal (336)	12.8	76
	Throat (336)	11.3	76
Monovalent (21)	Rectal (336)	15.5	76
	Throat (336)	13.1	71
Placebo (21)	Rectal (336)	21	100
	Throat (336)	16.4	86

TABLE VII

AVERAGE DURATION OF ADENOVIRUS EXCRETION FROM LAV STUDY IN A NAVAL RECRUIT COMPANY, NTC, GREAT LAKES, ILL. FEBRUARY-MARCH 1965

Vaccine group	Specimen	Average time virus excretion (days)
LAV	Rectal	8.1
	Throat	2.9
Bivalent	Rectal	5.2
	Throat	4.1
Monovalent	Rectal	5.8
	Throat	4.9
Placebo	Rectal	6.0
	Throat	4.5

TABLE VIII

COMPARISON OF VARIOUS TISSUE CULTURE SYSTEMS FOR ISOLATION OF ADENOVIRUS

Study specimen	Tissue culture			
	H.Ep-2	WI-38	HEK*	MK
	Percent positive ($\frac{\text{No. pos.}}{\text{No. treated}}$)			
Routine survey	54.6 (90/165)	54.0 (89/165)	ND	18.2 (30/165)
LAV company study:				
Vaccine group				
LAV				
Rectal	21.8 (35/160)	18.1 (29/160)	32.7 (16/49)	ND
Throat	6.8 (11/160)	5.6 (9/160)	18.8 (9/48)	
Placebo				
Rectal	22.5 (36/160)	20.0 (32/160)	29.5 (13/44)	ND
Throat	16.8 (27/160)	15.6 (25/160)	28.6 (12/42)	

*Data from University of Illinois -- Dr. G. G. Jackson.

TABLE IX

TEMPORAL RELATION OF GEOMETRIC MEAN NEUTRALIZING ANTIBODY RESPONSES AND ADENOVIRUS TYPE 4 ISOLATION FOLLOWING IMMUNIZATION

Vaccine group	DAYS POST IMMUNIZATION									
	0		7		14		21		28	
	GM* antibody titer	% Pos. isolation (R/T)**	GM antibody titer	% Pos. isolation (R/T)	GM antibody titer	% Pos. isolation (R/T)	GM antibody titer	% Pos. isolation (R/T)	GM antibody titer	% Pos. isolation (R/T)
LAV (20 men)	8	0/0	11	65/10	(36)	90/45	50	90/70	76	90/75
Placebo (21 men)	9	0/5	9	10/10	11	57/47	17	95/76	(48)	100/85
Bivalent (21 men)	8	0/5	10	5/5	23	33/57	(42)	57/62	85	76/71
Monovalent (21 men)	9	0/5	15	5/10	(61)	24/43	53	67/71	97	71/71

Parenthesis ( ) = first 4-fold increase.

*Reciprocal of dilution.

** R = rectal; T = throat

FIG. 1

ADENOVIRUS TYPE 4 ISOLATIONS (IN H.Ep$_2$) FROM THROAT SPECIMEN-LAV COMPANY STUDY, NTC, GREAT LAKES, ILLINOIS 1965

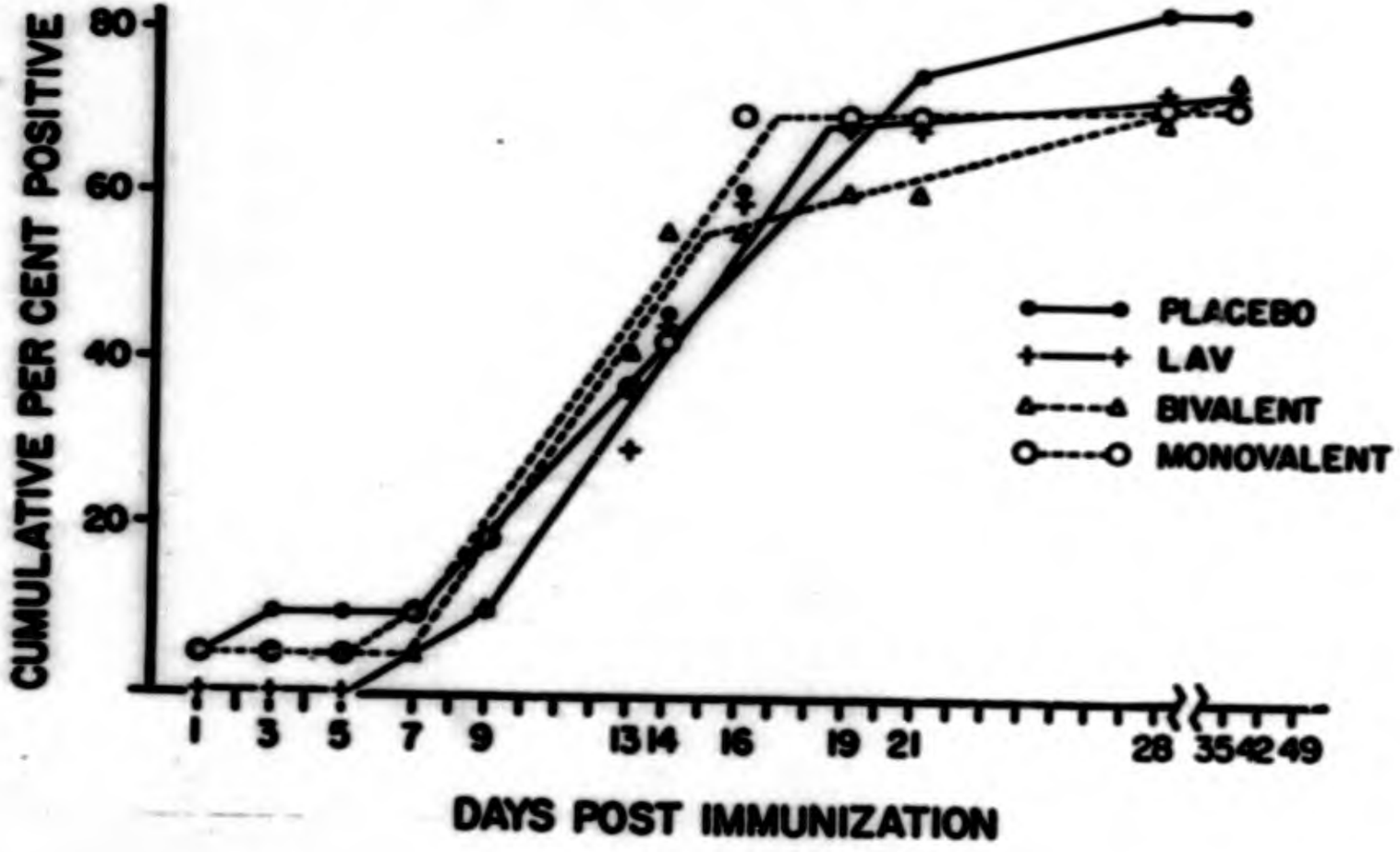


FIG. 2

ADENOVIRUS TYPE 4 ISOLATIONS (IN H.Ep_2) FROM RECTAL SPECIMEN-LAV COMPANY STUDY, NTC, GREAT LAKES, ILLINOIS 1965

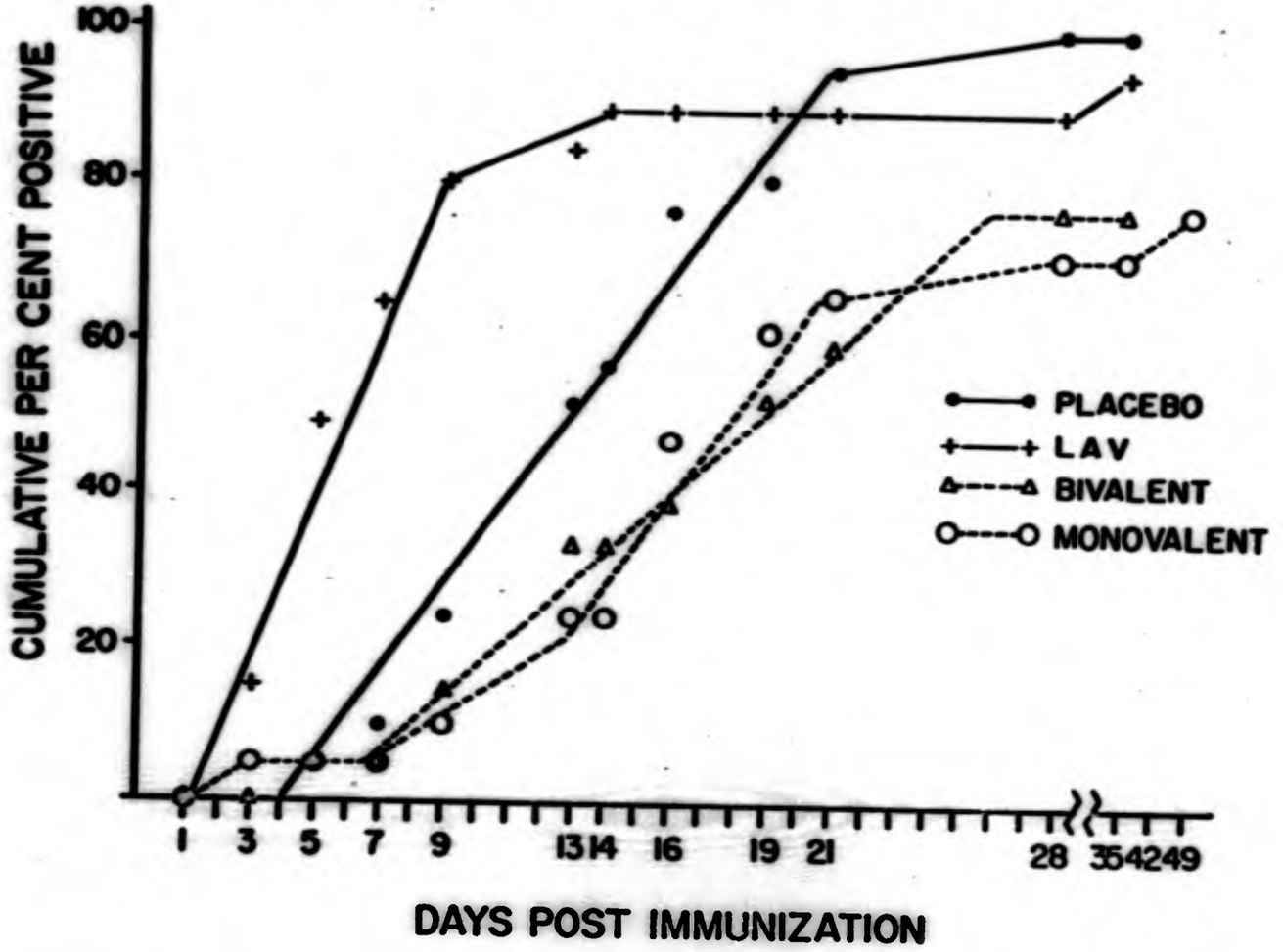


FIG. 3

ADENOVIRUS TYPE 4 ANTIBODY RESPONSES IN VARIOUS NEUTRALIZATION TESTS."LAV" COMPANY STUDY, LAV GROUP GREAT LAKES, ILLINOIS, 1965

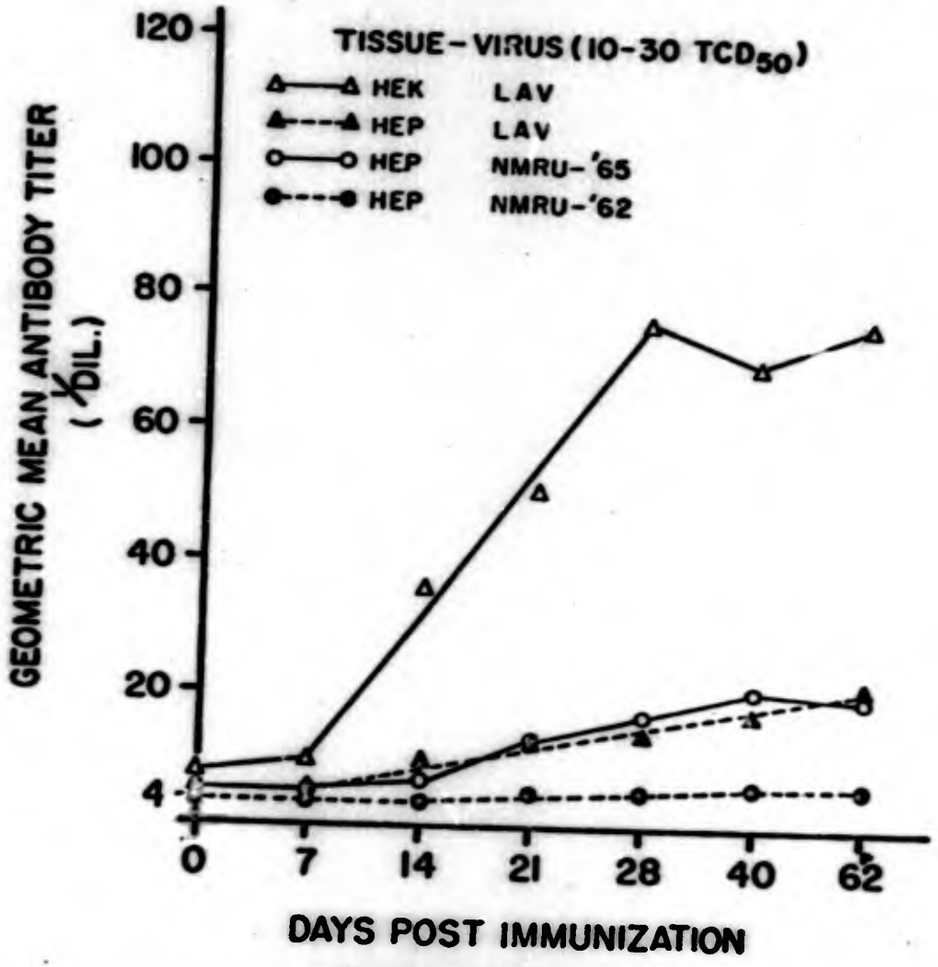


FIG. 4

ADENOVIRUS TYPE 4 ANTIBODY RESPONSES IN VARIOUS NEUTRALIZATION TESTS. LAV COMPANY STUDY, MONOVALENT GROUP GREAT LAKES, ILLINOIS 1965

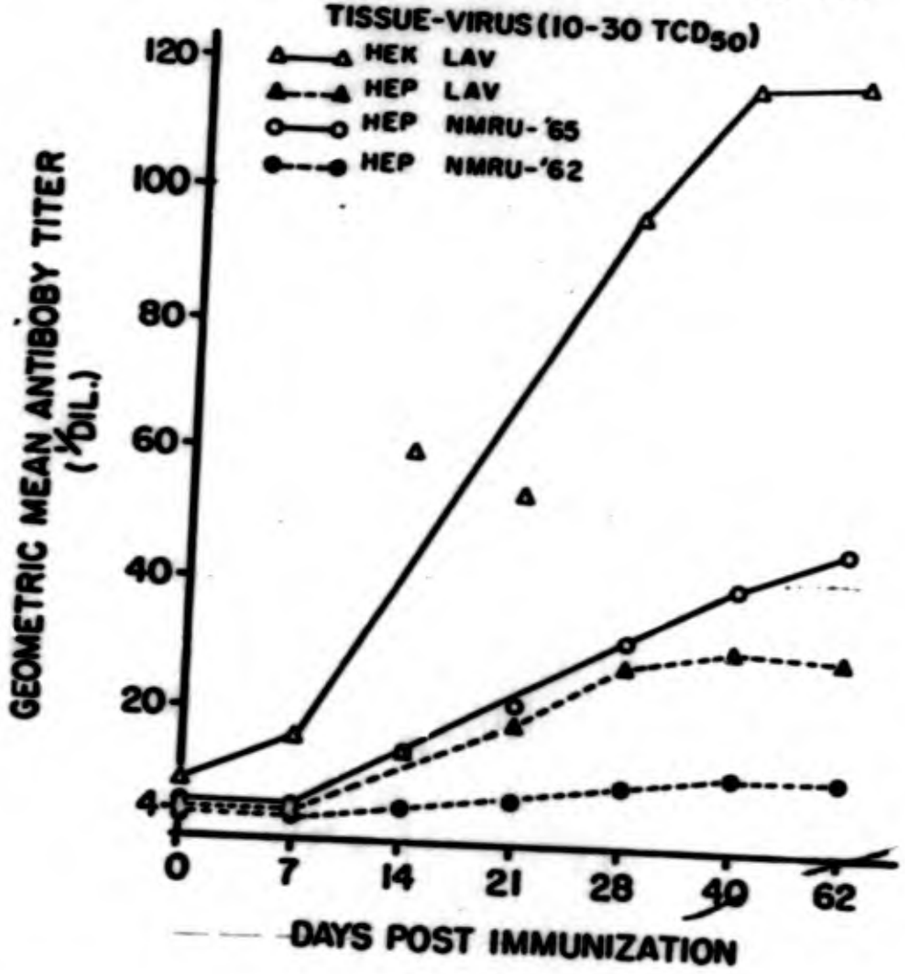


FIG. 5

ADENOVIRUS TYPE 4 ANTIBODY RESPONSES IN VARIOUS NEUTRALIZATION TESTS. LAV COMPANY STUDY, BIVALENT GROUP GREAT LAKES, ILLINOIS, 1965

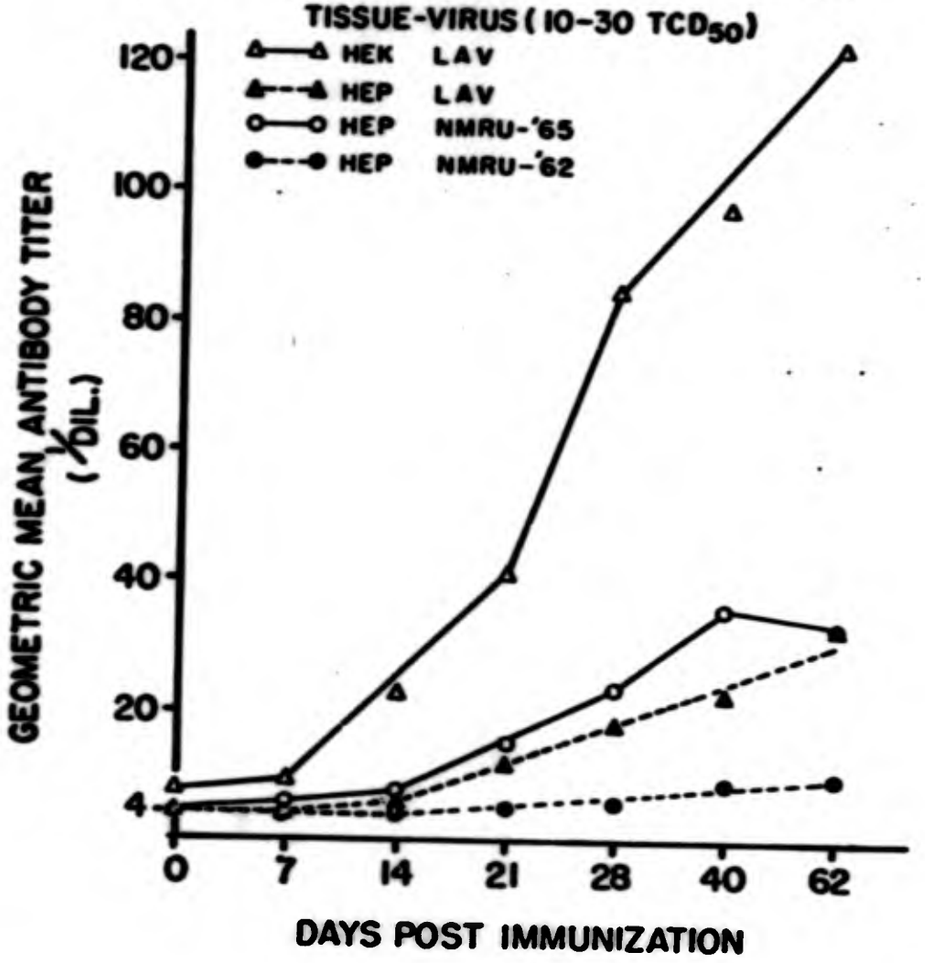


FIG. 6

ADENOVIRUS TYPE 4 ANTIBODY RESPONSES IN VARIOUS NEUTRALIZATION TESTS."LAV" COMPANY STUDY PLACEBO GROUP GREAT LAKES, ILLINOIS, 1965

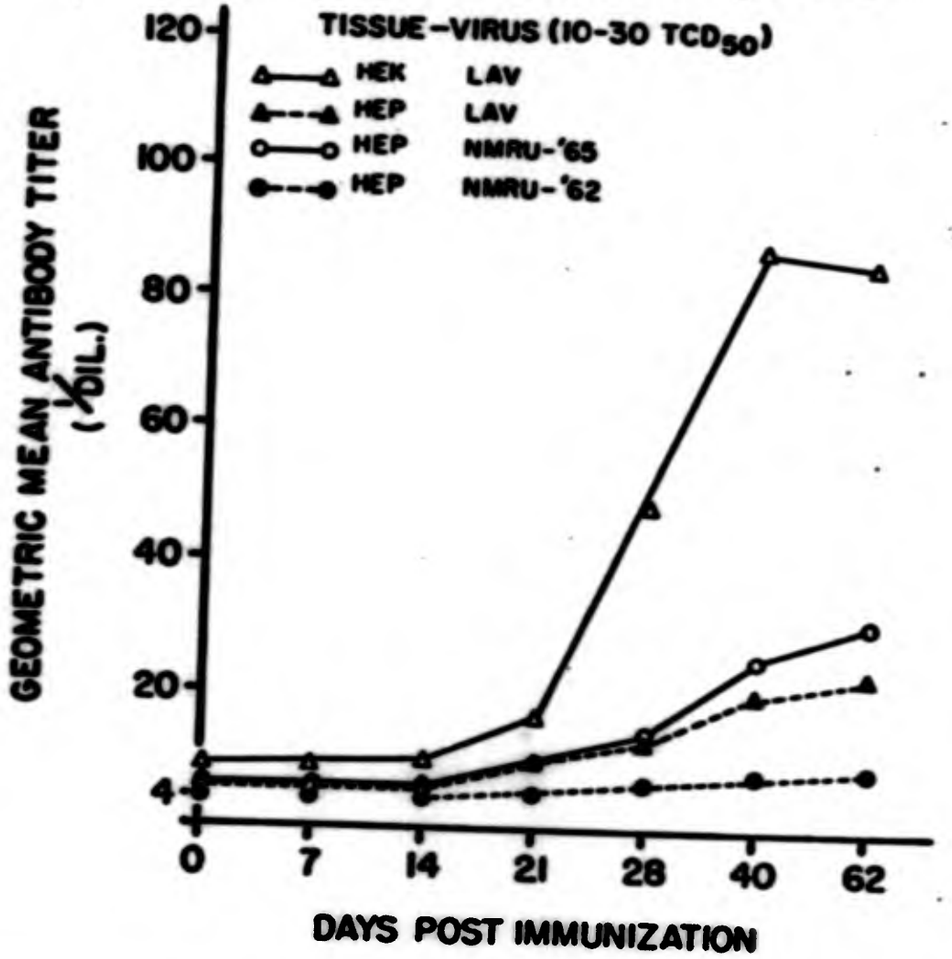


FIG. 7

ADENOVIRUS TYPE 4

NEUTRALIZING ANTIBODY RESPONSE TO LIVE AND INACTIVATED VACCINES LAV COMPANY

(MTC HEK; 10-30 TCD_{50})

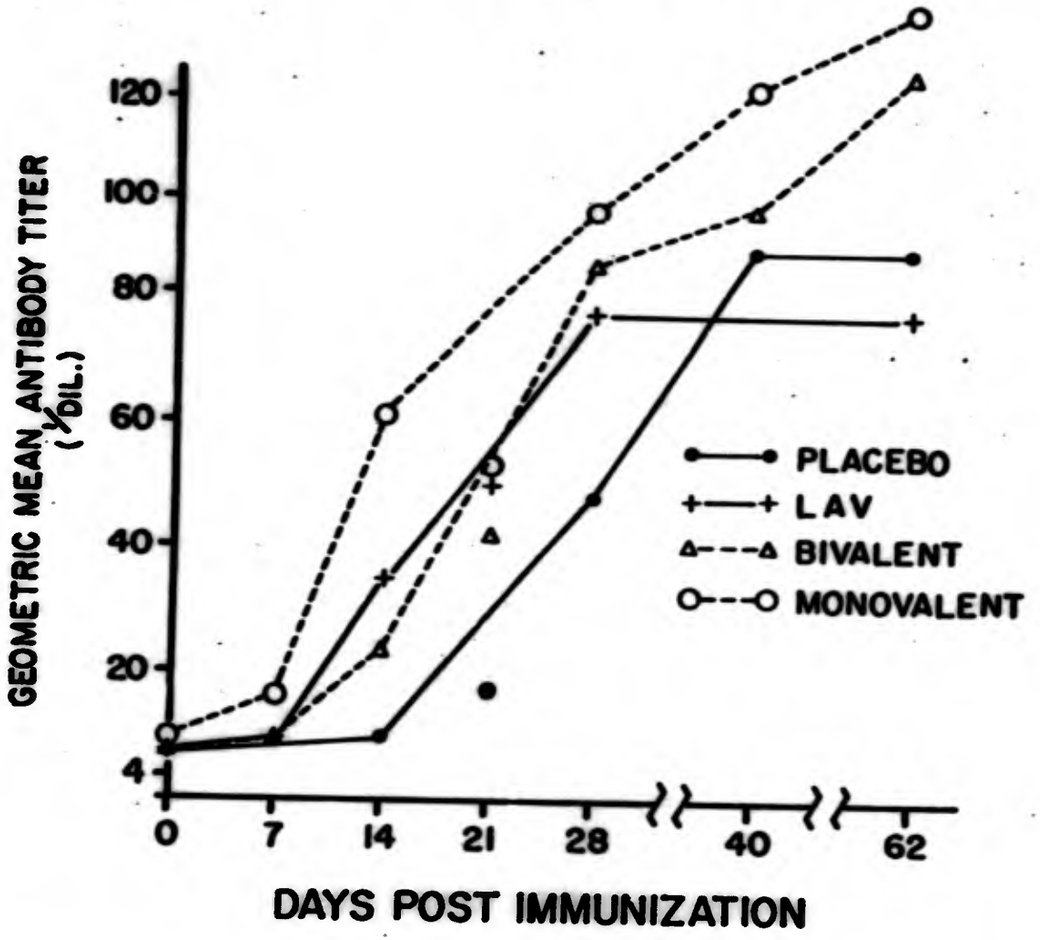
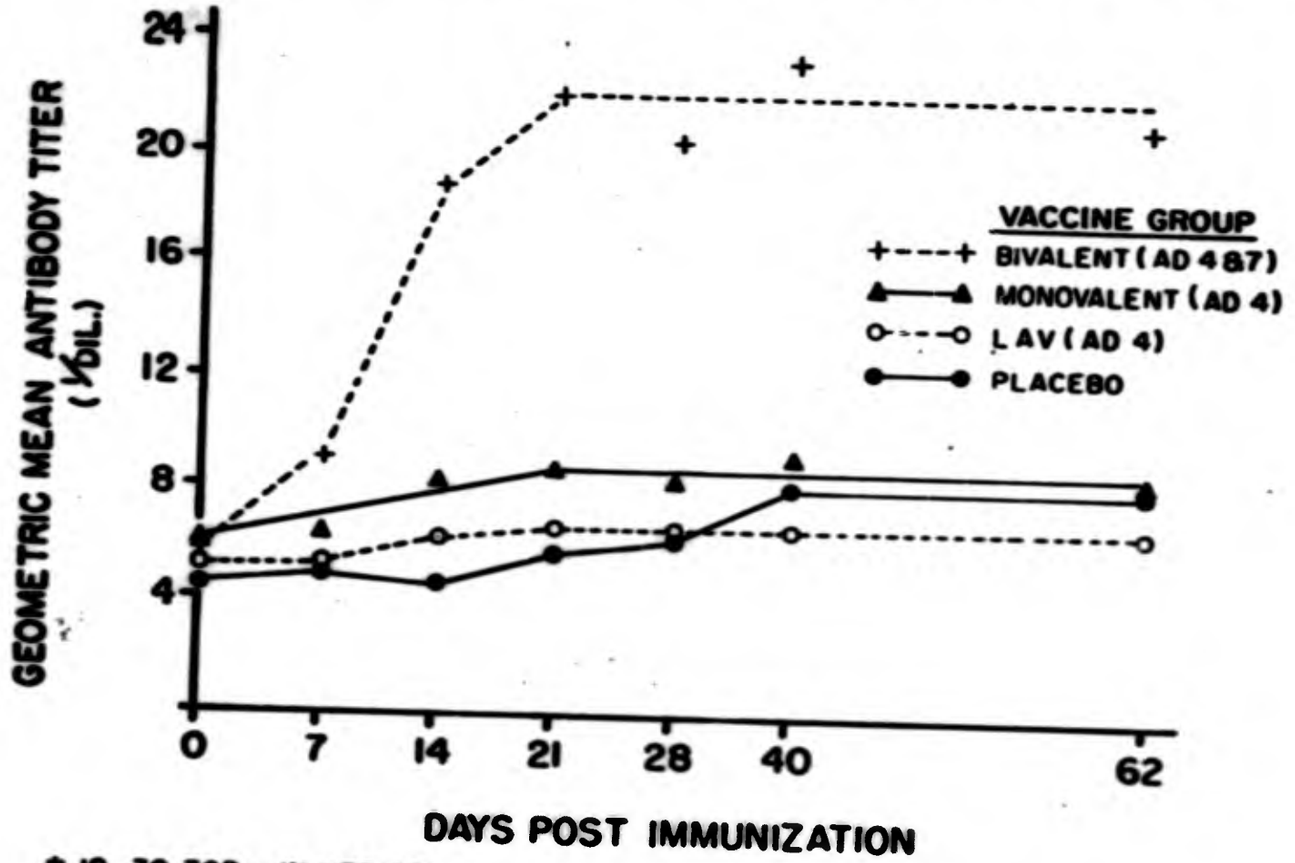


FIG. 8

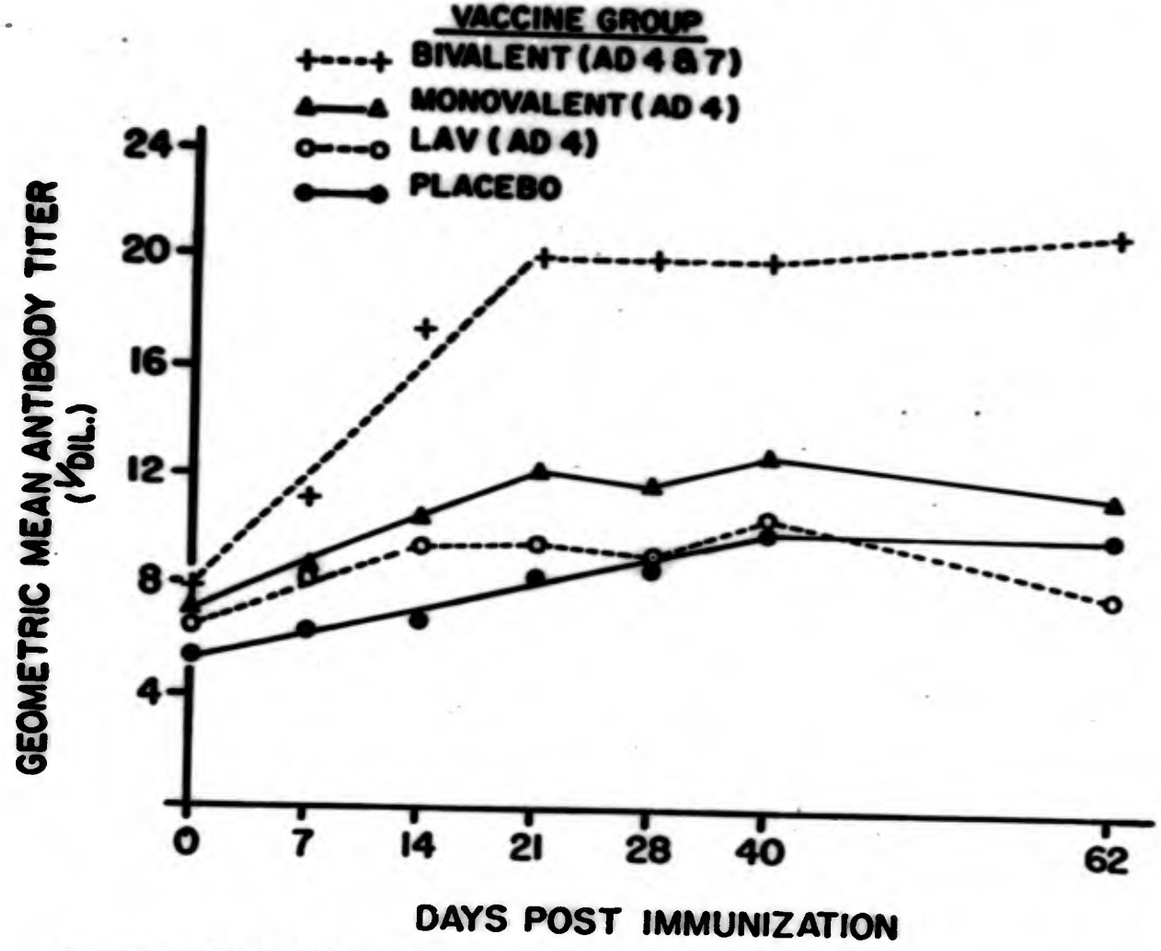
ADENOVIRUS TYPE 7 HOMOLOGOUS AND HETEROLOGOUS ANTIBODY RESPONSES* TO VARIOUS VACCINES. LAV COMPANY STUDY, GREAT LAKES, ILLINOIS, 1965



* 10-30 TCD_{50} IN HEP CELLS

FIG. 9

ADENOVIRUS TYPE 3 HETEROLOGOUS RESPONSES* TO VARIOUS VACCINES. LAV COMPANY STUDY GREAT LAKES, ILLINOIS, 1965



* 10-30 TCD_{50} IN HEP CELLS

COMPARISON OF LIVE AND INACTIVATED ADENOVIRUS VACCINES IN THE PREVENTION OF RECRUIT ACUTE RESPIRATORY DISEASE

BACKGROUND:

The recruit population at the U.S. Naval Training Center, Great Lakes, Illinois is made up of young adult males, primarily 17 to 20 years of age, who originate principally from the eastern half of the United States. Recruits arrive daily in varying numbers from the various recruiting stations.

Normally about 4 days are required to administratively process, classify, examine, inoculate and outfit a recruit. More than 10% of the recruits are delayed in processing for various administrative or medical reasons. At the completion of processing, recruits are trained in companies of 75 to 80 men for 9 weeks. All recruits not in training are housed in the same area.

After processing, the training company is moved to another camp which is for the early phases of training. After spending 3 weeks in this area, recruits are assigned work details and are housed and work throughout the entire recruit training complex, including the processing area for 1 week. Upon completion of this work week, the recruit companies are again physically moved to a different camp for the final 5 weeks of advanced training.

Sick calls are held 3 times a day and a recruit may seek treatment at his own discretion at any of these times. If treated as an outpatient, an individual permanent record is made of each visit, noting such data as complaint, temperature, and disposition. Daily official records indicate those men admitted to the sick list for inpatient care as well as the admission diagnosis. Such illness data on each recruit in training is abstracted and transcribed to machine punched cards.

Throughout the calendar year of 1964, the recruit population at Great Lakes, Illinois experienced relatively low admission rates for acute respiratory disease (Fig. 1). All recruits received the standard polyvalent influenza vaccine prior to the commencement of training. Benzathine penicillin, 1.2 million units, was given to all recruits, not hypersensitive to penicillin, in the second week of training. Further, throughout 1964 until 28 January 1965, all recruits received bivalent (Types 4 and 7) inactivated, adenovirus vaccine.

Serological determinations were made on sera from recruits in specially designated surveillance companies. The above prophylactic procedures were withheld from these companies. All recruits in these companies were bled on arrival at Great Lakes in the middle and at the end of training.

The adenovirus infection rates were low during the summer months (Table I). However, a definite increase in adenovirus infections was noted in recruits in training from October 1964 to January 1965. The respiratory illness rate likewise rose sharply at this time.

OBJECTIVES:

The general objective of this study was to extend and confirm the results of a previous study on use of live adenovirus vaccine to the Great Lakes population that usually experiences a high attack rate of adenovirus-associated disease. Specifically, the objectives were: (a) to determine the effectiveness of the live adenovirus vaccine in the prevention of recruit acute respiratory disease, (b) to compare its effectiveness with an inactivated adenovirus vaccine, and (c) to determine if there were any adverse reactions to or spread of the vaccine virus.

MATERIALS AND METHODS:

Forty-five recruit companies (3500 men) were assigned randomly to be treated as shown in Figure 2. Half of the men in each of these study companies were treated with either an adenovirus vaccine or its appropriate placebo control. The vaccines used were an oral live monovalent adenovirus Type 4, and a parenteral inactivated bivalent adenovirus Types 4 and 7 vaccine. As each company contained both recruits who had received a specific vaccine or placebo, this study design allowed a comparison of the placebo control groups to detect abnormal incidence of disease in such individuals who were cohorts of men fed live adenovirus vaccine.

The vaccines were administered from 19 February to 16 April 1965. The period of observation on this study population was from 19 February to 20 June 1965.

All men in the study companies who received the live and the monovalent vaccine, and all men from 10 companies who received the bivalent vaccine were subjected to extensive studies. A pre-vaccination, 35- and 65-day post-vaccination blood specimen was collected from all of these men. Throat and rectal specimens were collected for virus isolation as well as an acute and 21-day convalescent blood specimen from all of these recruits who were admitted. Serological tests with these sera included complement-fixation and neutralization. All tests were carried out in microplates with 4 units of adenovirus group complement fixing antigen, and 10-30 infectious doses of adenovirus Types 3, 4 and 7 for neutralization tests.

Virus isolation was performed in H.Ep-2, WI-38 or monkey renal cell cultures. Identification of virus isolates was by neutralization test with type-specific rabbit antisera. Myxovirus isolates were identified by hemadsorption-inhibition tests with influenza A2 guinea pig antisera.

In addition, all men from these companies who were admitted for inpatient care were seen by a medical officer who obtained a history and conducted a physical examination and evaluated the illness. A 14 x 17 chest film was obtained on all of the admitted men. Further, all men in these companies were seen by a team of medical examiners, which was headed by a medical officer, before vaccination and each week thereafter for 3 weeks, and then every 2 weeks until their departure from the Naval Training Center. This team took and recorded oral temperatures and elicited subjective disease complaints.

As part of the processing physical examination, all recruits received a 70mm chest x-ray. Those recruits with lung infiltrates are referred to the Naval Hospital for further evaluation. In connection with this study, commencing with those recruits entering training 2 weeks prior to the start of this study, and continuing until 2 weeks after the last of the study population had left the station, all recruits again received a 70mm chest film in the third week of training.

To preclude giving the live adenovirus vaccine to a recruit who may be held in the processing area or returned to his home, all vaccines were administered on or about the 4th day after arrival at Great Lakes, after all the processing procedures had been completed.

Double-blind techniques were carefully followed so that neither the recruit and the personnel responsible for training, nor the medical personnel responsible for observations or treatment had any knowledge of the treatment group to which any recruit or company was assigned.

All recruits received the standard polyvalent influenza vaccine during the processing period. Further, all men not hypersensitive to penicillin received 1.2 million units of benzathine penicillin in their second week of training, for the prophylaxis of streptococcal disease. All oral poliovirus vaccine was withheld from all of the 45 study companies during their training.

RESULTS:

Although the treatment a company or recruit received was assigned randomly, an analysis of certain attributes was made to determine the comparability of the relatively small numbers of subjects assigned to each treatment group. The results of some of these analyses are displayed in Tables II, III and IV. At least by parameters of similar age, geographic origin, and swimming ability, the various treatment groups appear to be comparable.

The extent of adenovirus infections of placebo men in each study company, as determined by complement-fixation serology, is shown in Table V. It is apparent from these data that the placebo-treated men in each company experienced a very constant, high attack rate of adenovirus infection. This is further evidence of the comparability of the study companies. Further, it is comforting to observe no excess of adenovirus complement-fixation conversions in the live virus vaccine placebo control group.

The effect the vaccines had on recruit acute respiratory disease hospital admissions is displayed in Table VI. The reduction of disease associated with each of the vaccines, when compared to its placebo, is statistically significant. While the reduction of total illness is the least for the live vaccine group, this difference is not statistically significant.

A slight protective effect of the live adenovirus vaccine became apparent by the 8th day post-vaccination (Fig. 3). All vaccines were definitely effective by the 11th day post-vaccination.

The readmission rate for acute respiratory disease was less in each of the vaccine groups (Table VII). However, the numbers involved are too small to be significant.

The number of men from each of the vaccine groups admitted with a diagnosis of pneumonia were relatively few as shown in Table VIII. About 2% of all men x-rayed in the third week of training had lung infiltrates consistent with the diagnosis of pneumonia. Both the live vaccine and the bivalent vaccine recipients had less evidence of pneumonia than did their placebos. Again the numbers are too small to be significant.

The amount of non-respiratory disease admissions from the various vaccine groups and placebos are essentially the same (Table IX). The vaccines had little effect on the length of time spent on the sick list for acute respiratory disease (Table X). Generally each vaccine was associated with a shorter hospital stay. These differences are too slight to be statistically significant.

Each vaccine did reduce the number of men requiring treatment as an outpatient for febrile respiratory disease as well as reducing the number of visits (Table XI). However, more men from each of the various groups were treated as outpatients for afebrile respiratory disease.

No difference in the number of men with symptoms or number of men with individual symptoms, or number of men febrile could be detected in any vaccine group in the barracks surveys. Indeed, the number of symptoms elicited in each vaccine group in each company were remarkably similar (Table XII). The number of men who were asymptomatic on every interview were rare and were distributed throughout the study companies (Table XIII).

The amount of adenoviruses recovered from men admitted with acute respiratory disease from each treatment group is displayed in Table XIV. As would be expected, more of the live vaccine recipients shed virus in their rectal specimens. With this exception, fewer adenoviruses were recovered from the specimens of each of the vaccine recipients. Likewise, fewer 4-fold complement-fixation responses were detected in men admitted with acute respiratory disease from each vaccine group (Table XV). The same reduction of laboratory evidence of infection is apparent in each vaccine group when different criteria for infection is used (Table XVI).

At the very onset of the study, a sharp increase in admission rates from recruits early in training was observed. Many of these cases presented a clinical picture of influenza. Influenza A2 virus was recovered from nasal washings from several of these admissions. A surveillance company not vaccinated with influenza vaccine had a complement-fixation conversion rate to influenza A of 35%. By hemagglutination-inhibition, the conversion rate was 45% (Table I). As all of the men in this vaccine study had received the standard polyvalent influenza vaccine, the extent of infection with influenza could not be accurately ascertained. In 3 weeks the clinical cases of influenza were no longer occurring, and the illness admission rate pattern had returned to normal. Therefore, in an attempt to determine the effect that influenza could have had on this vaccine evaluation, the study population was divided in half according to time of arrival (Table XVII). The ARD admission rates were determined for each half of the study population in each

of the vaccine groups. It was reasoned that those recruits arriving at the onset of the study probably were in more intimate contact with influenza infection than were those recruits who arrived later. All admission rates were about 30% higher in the first half of the study, except for those of the live vaccine placebo group. The crude relative reduction associated with the inactivated vaccines were approximately equal in each time period. However, because of the increased placebo admission rates in the second half of the study, a greater vaccine effect associated with the live vaccine was observed in the second half of the study population. Although this difference is not significant, the reversal of the illness pattern in the live vaccine placebo is an unexplained inconsistency.

DISCUSSION AND SUMMARY:

All adenovirus vaccines, live or inactivated, were equally effective in reducing the number of respiratory disease admissions by about 50%. This protective effect was apparent by the 11th day following vaccination. The protection that the two inactivated preparations gave are quite consistent with the effect of other inactivated adenovirus vaccines that have been tested in this population. However, the live vaccine gave better protection when tested in marine recruits at Parris Island, South Carolina, and Camp Lejeune, North Carolina. This is not surprising, however, as the epidemiological conditions of these two trials were markedly different. The vaccine was administered to Marine recruits at Parris Island, South Carolina, which at that time, was free of natural adenoviral disease. More than 3 weeks elapsed before these men were transferred to Camp Lejeune, North Carolina. At the time of this move, practically all of the vaccinated men had developed antibodies, while the placebos remained susceptible. The respiratory disease that those Marine recruits were subjected to at Camp Lejeune, North Carolina, was due primarily to adenovirus. The Navy recruits in this report were vaccinated 4 days after arrival at Great Lakes. Two weeks after vaccination, one-half of the respiratory admissions had occurred. It is apparent from the data presented in Figure 3 that these men were being challenged to adenovirus infection at the time of or shortly after vaccination. This early challenge, before the development of protective antibody could be expected to occur, could in part explain the lesser effect of these vaccines.

However, if one excludes those respiratory disease admissions which occurred within the first 10 days after vaccination, only a slight increase in the protective effect of the vaccine is observed (Tables VI and XVIII). This effect would suggest the presence of other respiratory disease pathogens in this population. There indeed was serological evidence of many infectious agents in this population, at the time of the study, as can be seen in Table I. Further evidence of the multiple etiology of respiratory disease in this population is shown in Figure 4 and Table XI. While all vaccines reduced hospital admissions, little effect was observed on febrile illnesses that were treated as outpatients. There is a definite excess of the milder afebrile respiratory disease which required only outpatient care found in all vaccine groups. This

type of adenovirus vaccine effect has been repeatedly observed in this recruit population, and has been defined as an ameliorative effect. Further, the number of recruits who escaped all acute respiratory disease,as shown in Table XIII, was not increased by the use of the vaccines. Likewise, the number of men with symptoms at any time in training was the same for each vaccine or placebo group.

Various attempts have been made to determine the effect that vaccines have had on the "specific disease" that they were intended to prevent. The most conventional techniques are the specific reduction which is based on the difference of laboratory diagnosed "disease" between vaccine and control. The IVE method of Stille utilizes the ratio of laboratory diagnosed infection in the control and the over-all difference in the disease rates in controls and vaccinated. The critical point of each of these methods is the criteria that is selected to indicate infection. In this study, virus isolations both from rectal and throat specimens, and complement-fixation serology is available on all of the patients hospitalized with acute respiratory disease (Table XVI). The specific reductions and IVE values have been calculated using these estimates of infection (Table XIX). It is apparent that varying degrees of effectiveness can be obtained depending on one's own criteria of infection.

One point that is apparent from this type of analysis is that the pattern of estimated vaccine effectiveness between the various vaccines is the same as the crude reduction. That is, that the most effective vaccine is the inactivated monovalent Type 4, while the least effective is the live preparation. However, the size of the study would not allow one to attach significance to this trend.

In this recruit population where there are multiple etiologies of acute respiratory disease, it is difficult to determine the specific effect of a single vaccine. Combinations of vaccines for various agents tested simultaneously, alone and in combination, may give a better insight as to the effectiveness of vaccines and the role in the disease that each agent may have.

TABLE I

Incidence of Infection as Determined Serologically Navy Recruits Not Receiving Respiratory Disease Vaccines or Penicillin, NTC, Great Lakes, Ill., 1964-1965

Agent - Test	Percent men infected during inclusive dates: July 16-Sept. 21, '64	Oct 20, '64-Jan 20, '65	Jan 22-April 7, '65	April 9-June 24, '65
Adenovirus - CF*	4	63	85	60
Adenovirus Type 3 - Ntr**		10		
Adenovirus Type 4 - Ntr		76		
Adenovirus Type 7 - Ntr		10		
ASO	4	2	10	8
M. pneumoniae - CF			13	4
Parainfluenza 1 - HI†		0	6	12
Parainfluenza 3 - HI		1	3	3
Influenza A2(Japan 305) - CF			31	3
Influenza B - CF			5	4
Influenza A2(Japan 170) - HI			45	
Influenza A2 (Taiwan) - HI			43	
Influenza B (Taiwan) - HI			0	
2060 - Ntr		7	12	9
2211 (HGP) - Ntr		7	3	12
779 (B632) - Ntr		4	13	9

*Complement-fixation

**Neutralization

† Hemagglutination-inhibition

TABLE II

Age Distribution of Recruits Receiving Various Adenovirus Vaccines or Placebos, NTC, Great Lakes, Ill., 1965

	Percent in Age Group					
Age	Live	Placebo	Monovalent	Placebo	Bivalent	Placebo
17	44.1	40.1	38.9	40.8	39.2	39.6
18	26.1	28.1	25.9	29.0	26.9	28.5
19	14.0	14.0	15.6	13.8	14.6	12.4
20	8.7	9.0	10.3	11.4	9.9	10.8
21	4.7	5.7	7.2	3.0	5.7	6.3
22	1.6	2.7	.9	1.0	2.0	1.1
23	.3	.3	.6	.6	1.1	.8
>23	.6	.0	.6	.4	.6	.5

P = >.20

TABLE III

Home Area of Recruits Receiving Various Adenovirus Vaccines or Placebos, NTC, Great Lakes, Ill., 1965

Area	Percent from area					
	Live	Placebo	Monovalent	Placebo	Bivalent	Placebo
New England	16.7	17.6	20	19.8	18.6	19.1
Mid-Atlantic	33.3	36.6	34	32.9	32.8	31
N.E. Central	24.2	19.3	22.2	20.8	21.7	20.5
N.W. Central	4.3	4.4	4.4	6.5	7.0	6.3
South Atlantic	11.8	12.2	9.1	11.9	11.0	12.4
S.E. Central	5.8	5.1	5.3	4.2	4.5	4.3
S.W. Central	1.9	1.7	3.2	3.0	3.3	3.6
Mountain	.6	.3	1.0	0	.8	.6
Pacific	1.2	.7	.8	1.2	.4	2.2

P = >.20

TABLE IV

Percent of Non-qualified Swimmers in Various Adenovirus Vaccine or Placebo Vaccine Groups, Naval Recruits, Great Lakes, Ill., 1965

	% Non-qualified swimmers
Live	21.0
Placebo	18.5
Monovalent	24.0
Placebo	25.1
Bivalent	21.2
Placebo	20.0

P = ~ .20

TABLE V

Distribution of Percent of Placebo-treated Recruits in Different Study Companies Showing A Four-fold Complement Fixation Rise to Adenovirus, NTC, Great Lakes, Ill., 1965

% Four-fold response	Type Company			
	Live	Monovalent	Bivalent	Total
56-60	2	1		3
61-65	1	2	2	5
66-70	2	3	3	8
71-75	3	4	2	9
76-80	2		1	3
81-85			2	2
Mean	68.5	68.2	71.6	69.4
SD	7.8	6.0	7.5	7.2

TABLE VI

Acute Respiratory Disease Admissions in Various Adenovirus Vaccine and Placebo-treated Recruits
NTC, Great Lakes, Ill., 1965

	Live	Placebo	Monovalent	Placebo	Bivalent	Placebo
Population	386	386	375	391	963	986
No. ARD admissions	75	139	61	134	160	332
Rate/1000	194	360	163	343	166	337
% Relative reduction	46		53		51	

TABLE VII

ADMISSIONS AND READMISSIONS FOR ACUTE RESPIRATORY DISEASE,
VARIOUS ADENOVIRUS VACCINES AND PLACEBOS,
NAVY RECRUITS, GREAT LAKES, ILL., 1965

	Live	Placebo	Monovalent	Placebo	Bivalent	Placebo
Population	386	386	375	391	963	986
No. men admitted	70	122	59	129	142	300
No. admissions	77	141	62	137	165	357
No. readmissions	7	19	3	8	23	57
Readmission rate/1000	100	156	51	62	162	190

TABLE VIII

EVIDENCE OF PNEUMONIA, VARIOUS ADENOVIRUS VACCINES AND PLACEBOS, NAVY RECRUITS, GREAT LAKES, ILL., 1965

	Live	Placebo	Monovalent	Placebo	Bivalent	Placebo
Population	386	386	375	391	963	986
Admitted with pneumonia	1	6*	4	2	7	12
Infiltrates 3rd week	5	10	7	8	19	27**
Total	6	16	11	10	26	39

*2 men admitted 24 hrs. post-vaccination.

**3 men admitted with diagnosis of pneumonia subsequent to X-ray survey.

TABLE IX

NON-RESPIRATORY DISEASE ADMISSIONS, VARIOUS ADENOVIRUS VACCINES AND PLACEBOS, NAVY RECRUITS, GREAT LAKES, ILL., 1965

	Live	Placebo	Monovalent	Placebo	Bivalent	Placebo
Population	386	386	375	391	963	986
No. non-resp. disease admissions	27	27	22	19	54	53
Rate/1000	70	70	59	49	56	54

TABLE X

Average Number of Hospital Days for Acute Respiratory Disease, Naval Recruits Receiving Various Adenovirus Vaccines or Placebo, NTC, Great Lakes, Ill., 1965

	Live	Placebo	Monovalent	Placebo	Bivalent	Placebo
Mean number hospital days	3.3	3.7	3.4	3.7	3.3	3.7

TABLE XI

Outpatient Acute Respiratory Disease Navy Recruits Receiving Various Adenovirus Vaccines or Placebos, NTC, Great Lakes, Ill., 1965

	Treatment group					
	Live		Inactivated			
	Monovalent (Adenovirus Type 4)	Placebo	Monovalent	Placebo	Bivalent (Adenovirus Types 4 & 7)	Placebo
Population	386	386	375	391	963	986
No. Febrile episodes	50	56	39	45	133	129
No. Febrile visits	114	134	79	104	318	329
No. Afebrile episodes	88	63	103	88	207	164
No. Afebrile visits	167	123	203	181	400	285

TABLE XII

Distribution of Percent Men in Different Companies with Respiratory Disease Symptoms Found on Barracks Interview, Various Adenovirus Vaccines or Placebos, NTC, Great Lakes, Ill., 1965

% of men in company with symptoms	Treatment group					
	Live		Inactivated			
	Monovalent (Adenovirus Type 4)	Placebo	Monovalent	Placebo	Bivalent (Adenovirus Types 4 & 7)	Placebo
26-30	1		1			
31-35		1	3	3	1	1
36-40	3	3	1	4	1	3
41-45	4	5	3	2	7	3
46-50	1	1	2	1	1	3
51-55	1					
Mean	39.4	38.6	41.1	42.0	41.2	41.6
S.D.	9.3	5.7	7.0	4.2	5.0	4.2

TABLE XIII

Percent of Men Interviewed without Respiratory Disease Symptoms throughout Recruit Training, Various Adenovirus Vaccines and Placebos, NTC, Great Lakes, Ill., 1965

	Treatment group					
	Live		Inactivated			
	Monovalent (Adenovirus Type 4)	Placebo	Monovalent	Placebo	Bivalent (Adenovirus Types 4 & 7)	Placebo
% men without symptoms	6.2	4.6	5.3	5.1	6.7	6.4

TABLE XIV

Adenovirus Type 4 Isolations from Hospitalized (ARD) Recruits Treated with Various Vaccines, NTC, Great Lakes, Ill., 1965

Type specimen positive	Treatment group					
	Live		Inactivated			
	Monovalent (Adenovirus Type 4)	Placebo	Monovalent	Placebo	Bivalent (Adenovirus Types 4 & 7)	Placebo
	Percent specimens positive					
Throat and rectal	30	60	30	67	34	65
Throat only	18	12	13	14	14	9
Rectal only	22	4	4	0	5	3
Throat	48	72	43	81	48	74
Rectal	52	64	33	67	38	68
Throat or rectal	70	76	46	81	52	77

TABLE XV

Complement-fixation Antibody Response and Adenovirus Type 4 Isolation in Recruits Hospitalized with Acute Respiratory Disease and Treated with Various Adenovirus Vaccines or Placebos, NTC, Great Lakes, Ill., 1966

Adenovirus Type 4 recovered	Treatment group					
	Live		Inactivated			
	Monovalent (Adenovirus Type 4)	Placebo	Monovalent	Placebo	Bivalent (Adenovirus Types 4 & 7)	Placebo
	Percent with 4-fold or greater antibody titer increase					
All cases	58	82	43	81	60	88
None	33	66	27	67	41	72
Throat and rectal	72	86	88	86	77	94
Throat only	73	80	29	71	89	91
Rectal only	62	100	0	-	66	75
Throat	72	85	70	83	81	93
Rectal	68	87	78	86	76	93
Throat or rectal	69	86	64	83	79	93

TABLE XVI

Evidence of Adenovirus Infection in Recruits Hospitalized with Acute Respiratory Disease Who Received Various Adenovirus Vaccines or Placebos, NTC, Great Lakes, Ill., 1965

Infection criteria	Percent Infected Various Treatment Groups					
	Live		Inactivated			
	Monovalent (Adenovirus Type 4)	Placebo	Monovalent	Placebo	Bivalent (Adenovirus Types 4 & 7)	Placebo
Complement-fixation rise	58	82	43	81	60	88
Throat isolate	48	72	43	81	48	74
Throat isolate, with CF rise	35	61	30	67	39	69
Rectal isolate	52	64	33	67	38	68
Rectal isolate, with CF rise	35	56	26	58	29	63
Rectal or throat isolate	70	76	46	81	52	77
Rectal or throat isolate, with CF rise	48	65	29	67	41	72
Rectal, or throat isolate, or CF rise	80	93	65	94	74	94

TABLE XVII

ACUTE RESPIRATORY DISEASE ADMISSIONS, VARIOUS ADENOVIRUS VACCINES AND PLACEBOS, NAVY RECRUITS, GREAT LAKES, ILL., 1965
(STUDY DIVIDED IN HALF)

	Live	Placebo	Monovalent	Placebo	Bivalent	Placebo
FIRST HALF						
Population	197	192	187	190	450	470
No. ARD admissions	46	66	37	76	88	194
Rate/1000	234	344	198	400	196	413
% relative reduction	32		51		53	
SECOND HALF						
Population	189	194	188	201	513	516
No. ARD admissions	29	73	24	57	72	139
Rate/1000	153	376	128	284	140	269
% relative reduction	59		55		48	
% difference first half vs. second half	35	-9	35	29	29	35

TABLE XVIII

Reductions of Respiratory Disease Admissions Associated with Various Adenovirus Vaccines, Navy Recruits, Great Lakes, Ill., 1965
(First 10 Days Post-vaccination Excluded)

	Treatment group					
	Live		Inactivated			
	Monovalent (Adenovirus Type 4)	Placebo	Monovalent	Placebo	Bivalent (Adenovirus Types 4 & 7)	Placebo
Population	386	386	375	391	963	986
No. ARD admissions	51	105	38	116	95	265
Rate/1000	132	272	101	297	99	269
Crude % relative reduction	51		66		63	

TABLE XIX

Estimates of Vaccine Effectiveness Utilizing Specific Illness Reduction, and the IVE Method Based on Various Criteria of Infection

Criteria of infection	Live vs. Placebo		Monovalent vs. Placebo		Bivalent vs. Placebo	
	Specific reduction	IVE	Specific reduction	IVE	Specific reduction	IVE
Complement-fixation response	62	56	75	64	66	58
Throat isolate	64	64	75	64	68	69
Throat isolate + CF response	69	75	83	78	72	74
Rectal isolate	56	72	77	78	72	75
Rectal isolate + CF response	66	82	79	90	77	81
Rectal or throat isolate	50	61	73	64	67	66
Rectal or throat isolate + CF response	60	71	79	78	72	71
Rectal isolate or throat isolate or CF response	54	49	65	55	61	54

FIG. 1

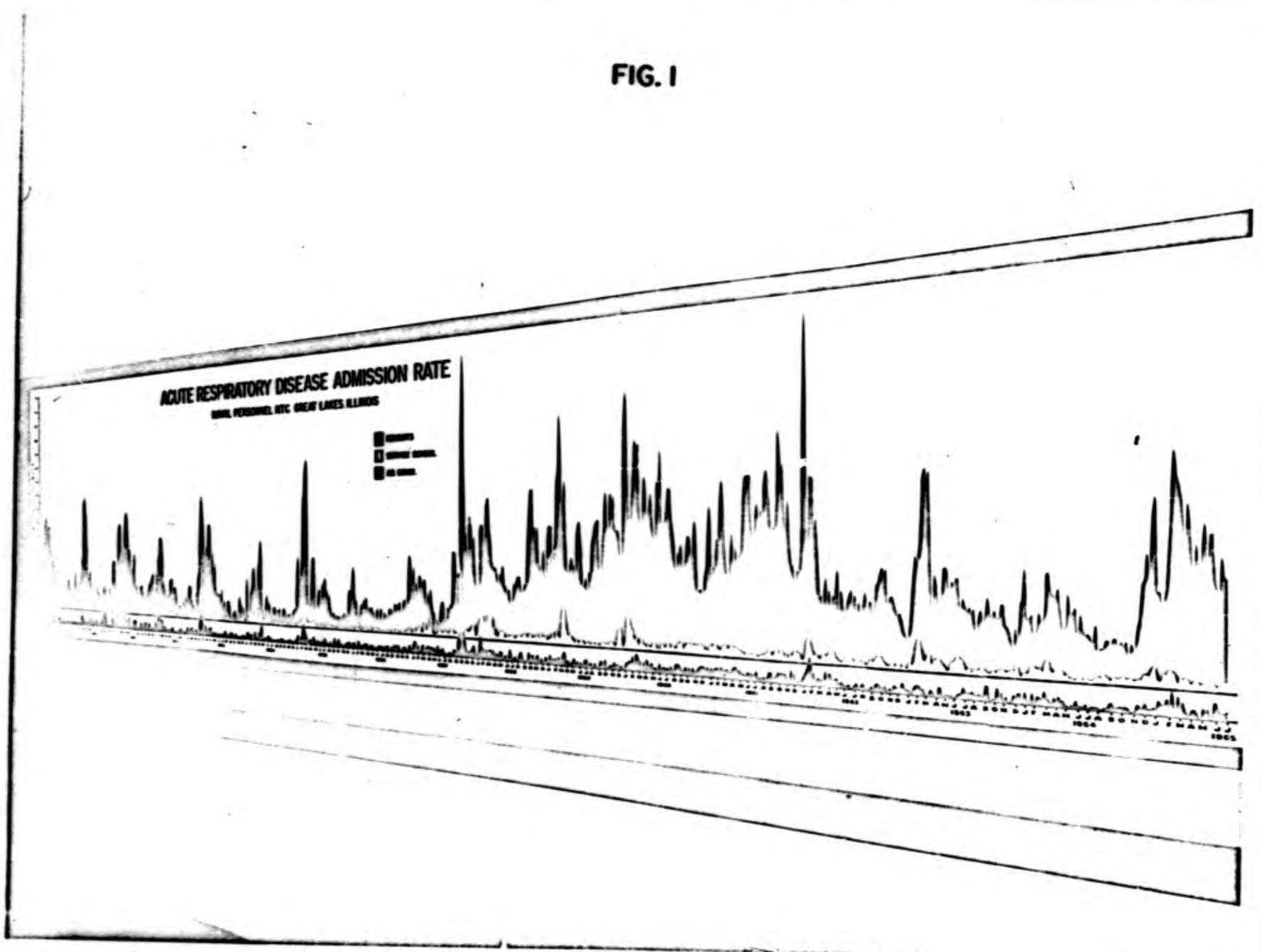


FIG. 2

STUDY POPULATION LIVE ADENOVIRUS VACCINE NAVAL RECRUITS, GREAT LAKES, ILLINOIS, 1965

NUMBER COMPANIES		NUMBER MEN
10	LIVE ORAL	386
	ORAL PLACEBO	386
10	INACTIVATED MONOVALENT	375
	SALINE PLACEBO	391
25	INACTIVATED BIVALENT	963
	SALINE PLACEBO	986

FIG. 3

ACUTE RESPIRATORY DISEASE ADMISSION RATES AFTER VACCINATION WITH ADENOVIRUS VACCINES OR PLACEBO
NAVY RECRUITS, GREAT LAKES, ILLINOIS, 1965

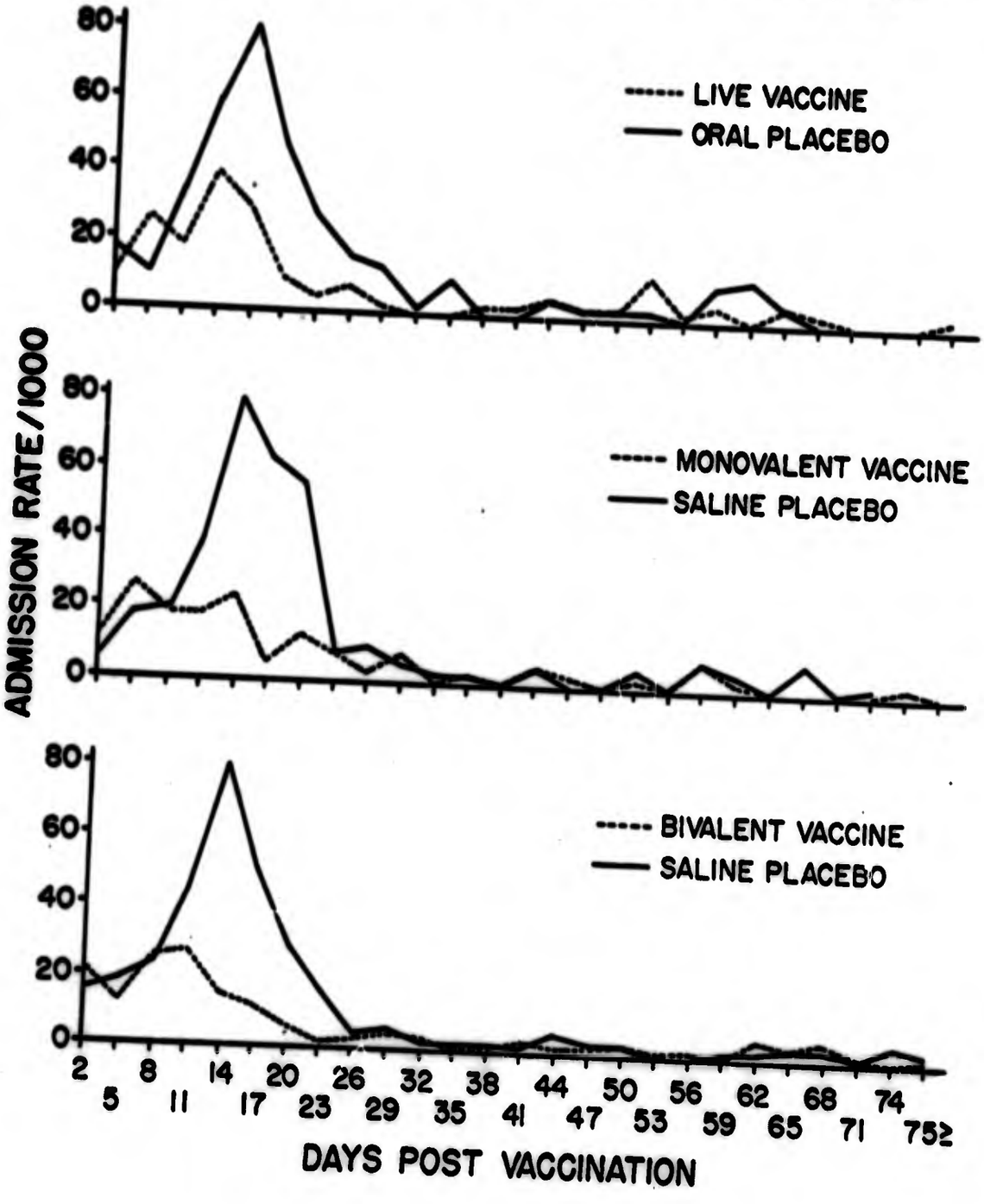


FIG. 4

MOST SEVERE RESPIRATORY DISEASE CONDITION REPORTED BY NAVAL RECRUITS DIFFERENT ADENOVIRUS VACCINES GREAT LAKES, ILLINOIS, 1965