Special Article

A CLINICAL EVALUATION OF VACCINATION AGAINST INFLUENZA

PRELIMINARY REPORT

By Members of the Commission on Influenza, Board for the Investigation and Control of Influenza and Other Epidemic Diseases in the Army, Preventive Medicine Service, Office of the Surgeon General, United States Army

In the autumn of 1943 members of the Commission on Influenza, and associates, Board for the Investigation and Control of Influenza and other Epidemic Diseases in the Army, Preventive Medicine Service, Surgeon General's Office, United States Army, undertook with Dr. Thomas Francis Jr., as director, to carry out a controlled clinical trial of the prophylactic efficacy against epidemic influenza of a concentrated, inactivated vaccine containing the viruses of influenza types A and B. Preceding studies had shown that a vaccine similarly prepared was capable of furnishing definite protection against experimental induction of influenza A or B.[1] The present account constitutes a preliminary clinical evaluation of the influence of vaccination on the incidence of influenza during the epidemic of influenza A which occurred in November and December 1943.

VACCINE

The vaccine was prepared in the laboratories of biologic firms according to specifications furnished by the commission and purchased at minimal cost with commission funds. Virus was obtained from the allantoic fluid of embryonated hen's eggs inoculated forty-eight hours earlier. The virus was concentrated approximately ten times in isotonic solution of sodium chloride following adsorption to, and elution from, the embryonic erythrocytes.[2] The infectious capacity was inactivated by solution of formaldehyde in a concentration of 1 : 5,000. Phenyl mercuric nitrate 1 : 100,000, or borate 1 : 50,000, was then added for bacteriostatic purposes. The material was bottled in 50 cc. amounts in liquid form. The standard requirements for sterility of bulk and bottled biologic products were met.

Each 1.0 cc. of the vaccine was made up of 0.5 cc. representing type A virus recovered from 5.0 cc. of allantoic fluid and 0.5 cc. representing the type B virus recovered from 5.0 cc. of allantoic fluid. The type A component represented equal parts of the PR8 strain and of the Weiss strain, isolated in May 1943.[3] The type B component contained only the Lee strain.

The vaccine was tested by inoculation of mice and eggs to demonstrate that no infectious capacity remained. Its capacities to agglutinate chicken erythrocytes [4] and to induce immunity in mice after intraperitoneal inoculation were also determined as indicative of antigenic activity.

Control material consisting of isotonic solution of sodium chloride to which solution of formaldehyde 1 : 5,000 and phenyl mercuric nitrate 1 : 100,000 were added was prepared, bottled and subjected to the same tests for sterility.

THE PLAN OF STUDY

With approval of appropriate authorities, the study was carried out in Army Specialized Training Program units of eight universities in different parts of the United States and in a ninth group comprising the members of Army Specialized Training Program units of five New York medical and dental colleges. Approximately 12,500 men were involved. The populations were highly stable, so that the proportion of men lost from the study was extremely low. In most instances the men were housed as large groups in dormitories.

Vaccine prepared by two different firms was employed in all locations. Except in one unit equal volumes of the two preparations were mixed just before inoculation, so that no selection occurred on this basis. Each

Table 1.—*Results in Group 1: Cornell University, Ithaca, N. Y., and New York Medical and Dental Colleges*

Major Norman Plummer, M. C., A. U. S., and Wilson G. Smillie, M.D., Cornell University Medical College, New York. Dr. Jocelyn Woodman participated in the clinical studies at Ithaca.

Epidemic period: Cornell, 11/23-12/18; New York Medical Colleges, 11/23-12/18/43.
Diagnosis: Patients reporting with temperature of 100 F. or greater. Cases of obviously different origin excluded.

Unit	Date Vaccinated	Number in Study		Cases by Weeks Ending 11/27	12/4	12/11	12/18	Total Cases	Incidence, per Cent	Percentage of Total Cases
Cornell University *.....	11/10/43	Vaccinated	498	1	0	1	13	15	3.01	26
		Control	484	2	4	9	28	43	8.88	74
		Total	982					58		
N. Y. Medical and Dental Colls.†..	10/21-11/4/43	Vaccinated	976	1	0	6	7	14	1.43	30
		Control	977	2	6	12	13	33	3.37	70
		Total	1,953					47		

* The incidence 11/28 to 12/6/43 is based to a large extent on questioning, since unit was on furlough during this period.
† These data are considered incomplete. The low incidence is probably related to the difficulty encountered in obtaining proper reporting among the high percentage of men living in private homes.

Support and assistance in arranging the studies were furnished by Col. Charles M. Walson, Col. Don C. Hilldrup, Col. Herbert C. Gibner and Col. Howard C. Moore, respectively, surgeons of the 2d, 6th, 7th and 9th service commands.

Continued aid and cooperation were furnished by the commanding officers of the different A. S. T. P. units among which the investigations were made, namely Col. Edwin R. Van Deusen, Cornell University; Col. Arthur E. Fox, Princeton University; Lieut. Col. J. D. Cope, Rutgers University; Col. Raymond P. Cook, C. C. N. Y.; Col. Frederick C. Rogers, University of Michigan; Col. Harry King, University of Minnesota; Col. Luke D. Zech, University of Iowa; Col. Francis R. Hunter, University of California; Lieut. Col. Mark R. N. Zwilliam, Columbia Medical and Dental College; Lieut. Col. Phillip B. Connelly, Cornell Medical College; Capt. Robert Geiss, Long Island Medical College; Major Albert C. Dorat, New York Medical College, and Capt. George F. Dyson, New York University College of Medicine and Dentistry.

1. Francis, T., Jr.; Salk, J. E.; Pearson, H. E., and Brown, P. N.: Protective Effect of Vaccination Against Induced Influenza A, Proc. Soc. Exper. Biol. & Med. **55**: 104 (Feb.) 1944. Salk, J. E.; Pearson, H. E.; Brown, P. N., and Francis, T., Jr.: Protective Effect of Vaccination Against Induced Influenza B, ibid. **55**: 106 (Feb.) 1944.

2. Francis, T., Jr., and Salk, J. E.: A Simplified Procedure for the Concentration and Purification of Influenza Virus, Science **96**: 499-500 (Nov. 27) 1942.

3. Salk, J. E.; Menke, W. J., and Francis, T., Jr.: Identification of Influenza Virus Type A in Current Outbreak of Respiratory Disease, J. A. M. A. **124**: 93 (Jan. 8) 1944.

4. Hirst, G. K.: The Quantitative Determination of Influenza Virus and Antibodies by Means of Red Cell Agglutination, J. Exper. Med. **75**: 47-64 (Jan.) 1942.

company or organization within a unit was divided in half, so that alternate individuals received, respectively, vaccine and control material. One dose of 1.0 cc. was given subcutaneously. After vaccination was completed the records containing this information were removed to other quarters, so that on subsequent visits the observer had no information as to whether a patient belonged to the vaccinated or the control group. Individuals who did not receive inoculation of control material were not considered controls. Vaccination was carried out at different times in the various units but in the main was completed by the middle of November. After the group had been vaccinated, new arrivals were not taken into the study. The time of vaccination in relation to the recognized onset of influenza is seen in the subsequent data.

Prior to vaccination and throughout the period thereafter, close observation of all individuals reporting to sick call was maintained by members of the investigating groups. The same type of record card was used throughout. An effort was made to gain uniformity in the designation of cases by accepting for the diagnosis of influenza those individuals who at the time of reporting to sick call had symptoms suggestive of influenza, i. e. rapid onset with mild upper respiratory complaints, chilliness, aches and prostration and were admitted to hospital with sublingual temperatures of 100 F. or more without obvious evidence of other disease. Fresh typical common colds, characteristic follicular tonsillitis and infectious mononucleosis were excluded from the diagnosis of influenza. Owing to local regulations or facilities, certain variations in the requirements for admission to hospital were encountered. In general, however, it appears that the criteria adopted would tend more to the inclusion in the series of cases which were not influenza than to the exclusion of cases which were influenza. While extensive collections of materials for virus and serologic investigation were made, the clinical impressions here stated have not been modified or corrected by any such data.

TABLE 2.—*Results in Group 2: Princeton University, Princeton, N. J., Rutgers University, New Brunswick, N. J., and College of City of New York*

George K. Hirst, M.D., Major Norman Plummer, M. C., A. U. S., and William F. Friedewald, M.D., Laboratories of International Health Division, Rockefeller Foundation, New York.

Epidemic periods: Princeton, 11/28-12/18/43; Rutgers, 11/22-12/18/43; C. C. N. Y., 11/7-12/18/43.

Diagnosis: Patients reporting with respiratory infection and temperature of 100 F. in whom diagnosis of some other disease could not definitely be made.

Unit	Date Vaccinated	Number in Study		Cases by Weeks Ending 11/27	12/4	12/11	12/18	Total Cases	Incidence, per Cent	Percentage of Total Cases
Princeton	11/2/43	Vaccinated	590	0	8	6	3	17	2.88	27
		Control	560	0	21	17	7	45	8.04	73
		Total	1,150					62		
Rutgers	11/1/43	Vaccinated	606	2	0	0	5	7	1.16	14
		Control	606	4	8	20	9	41	6.77	86
		Total	1,212					48		
C. C. N. Y.	11/19/43	Vaccinated	1,050	33	8	6	0	14*	1.33	16
		Control	1,055	27	52	17	6	75*	7.11	84
		Total	2,105					89*		

* Because influenza began about the time vaccination was done, figures represent only those cases which occurred on or after the ninth post-vaccination day. The number of cases indicated for the week ended November 27 include all occurring during the period from November 7 to November 27.

TABLE 3.—*Results in Group 3: University of Michigan, Ann Arbor*

Jonas E. Salk, M.D., and Wilbur J. Menke, M.D., Department of Epidemiology, School of Public Health, University of Michigan.

Epidemic periods: First case 11/12/43. Epidemic peak 11/20-12/4/43.

Diagnosis: Clinical diagnosis of influenza at time of reporting on sick call, temperature of 100 F. or more and admitted to hospital.

Unit	Date Vaccinated	Number in Study		Cases by Weeks Ending 11/20	11/27	12/4	12/11	12/18	Total Cases	Incidence, per Cent	Percentage of Total Cases
University of Michigan	10/26-11/2/43	Vaccinated	888	0	5	2	6	7	20	2.29	21
		Control	888	8	17	36	5	8	74	8.51	79
		Total	1,776						94		

First case on 11/12/43 was not in study group.

TABLE 4.—*Results in Group 4: University of Minnesota, Minneapolis*

E. R. Rickard, M.D., Minnie Thigpen, B.S., and James H. Crowley, B.A.. Influenza Laboratory, Division of Preventable Diseases, Minnesota Department of Health, Minneapolis. This study was aided by a grant from the International Health Division of the Rockefeller Foundation.

Epidemic period: 11/21/43-12/13/43.

Diagnosis: Reporting to sick call with respiratory illness and admitted with temperature of 99 F. or more.

Unit	Date Vaccinated	Number in Study		Cases by Weeks Ending 11/27	12/4	12/11	12/18	Total Cases	Incidence, per Cent	Percentage of Total Cases
University of Minnesota	11/5-11/13/43	Vaccinated	599	7	4	4	1	16	2.69	22.5
		Control	607	35	10	7	3	55	9.06	77.5
		Total	1,206					71		

Cases of influenza were not noted in any dormitory housing inoculated students until at least eleven days after vaccination of the group housed in that dormitory.

An epidemic of influenza A was first identified in the Middle West about the second week in November. The disease was subsequently recognized in other localities within a short time thereafter. The epidemic period in the posts under observation was three to four weeks. The disease was, in general, mild, of three to four days' duration and with a low incidence of complications.

The accompanying data represent tabulations of cases called influenza at the time of illness. The designation cent, while in the 6,263 receiving vaccine there was an incidence of 2.22 per cent, a ratio of 3.2 to 1.

The significance of the results is heightened by the uniformity of trend in practically all instances. The two greatest deviations are noted in the medical school units and in California. In the former the low incidence of the disease is thought to be related to the lack of central reporting. In the latter instance there is no clear difference between control and vaccinated groups; various factors such as furlough, the increased interval since

TABLE 5.—*Results in Group 5: University of Iowa, Iowa City*

William M. Hale, M.D., with technical assistance of Mr. Earl J. Gifford. Department of Bacteriology, University of Iowa, Iowa City.

Epidemic period: 11/29-12/25/43.

Diagnosis: Cases with diagnosis of influenza, most all with temperatures of 100 F. or more.

Unit	Date Vaccinated	Number in Study		Cases by Weeks Ending 12/11	12/18	12/25	1/1/44	Total Cases	Incidence, per Cent	Percentage of Total Cases
University of Iowa	12/2-12/4/43	Vaccinated	599	(9) 3	2	4	2	11	1.83	21
		Control	599	(11)12	16	11	1	40	6.67	79
		Total	1,198					51		

Five cases before vaccination completed. Summarized totals exclude the cases occurring in the first five days following vaccination. Numbers in parentheses indicate those occurring in the first five days. Hemolytic streptococcus pharyngitis occurred concurrently with the outbreak of influenza. Twenty per cent of throat cultures were positive for B. hemolytic streptococcus.

TABLE 6.—*Results in Group 6: University of California, Berkeley*

Monroe D. Eaton, M.D., and Gordon Meiklejohn, M.D. Research Laboratory of the California Department of Public Health, Berkeley, Calif. This study was aided by a grant from the International Health Division of the Rockefeller Foundation.

Epidemic period: 11/26/43-1/15/44.

Diagnosis: All cases hospitalized with acute febrile respiratory disease.

Unit	Date Vaccinated	Number in Study		Cases by Weeks Ending 12/3	12/10	12/17	12/24	12/31	1/7/44	1/15	Total Cases	Incidence, per Cent	% of Total
University of California	10/19-10/27/43	Vaccinated	457	1	1	5	8	4	1	4	24	5.25	41
		Control	435	3	1	10	5	3	4	8	34	7.81	59
		Total	892								58		

About 10 cases of streptococcic infection including 2 with scarlet fever occurred during the influenza epidemic.

The unit was on furlough 12/4 to 12/12/43. A few were away 12/22 to 12/28/43.

TABLE 7.—*Summary of Clinical Evaluation of Vaccination Against Influenza*

The combined totals of all results.

Group	Service Command	ASTP Unit	Dates of Vaccinated	Total Number	Number of Subjects: Vaccinated	Number of Subjects: Control	Number of Cases: Vaccinated	Number of Cases: Control	Incidence, per Cent: Vaccinated	Incidence, per Cent: Control	Percentage of Total Cases: Vaccinated	Percentage of Total Cases: Control
1	2d	Cornell	11/9	982	498	484	15	43	3.01	8.86	26	74
	2d	N. Y. Med. Schools	10/26-11/4	1,953	976	977	14	33	1.43	3.38	30	70
2	2d	Princeton	11/2	1,150	590	560	17	46	2.88	8.20	27	73
	2d	Rutgers	11/1	1,212	606	606	7	42	1.15	6.93	14	86
	2d	C. C. N. Y.	11/19	2,105	1,050	1,055	14	75	1.33	7.10	16	84
3	6th	Michigan	10/26-11/2	1,776	888	888	20	74	2.25	8.35	21	79
4	7th	Minnesota	11/5-11/13	1,206	599	607	16	55	2.68	9.06	22	78
5	7th	Iowa	12/2-12/4	1,198	599	599	11	40	1.83	6.67	21	79
6	9th	California	10/19-10/27	892	457	435	24	34	5.25	7.80	41	59
Totals				12,474	6,263	6,211	138	442	2.22	7.11	23.8	76.2

has been made purely on clinical grounds without reference to serologic or other virus studies for identification of individual cases. The division according to vaccinated or control was not done until the epidemic period was thought to have been passed. The results for the respective units were compiled by the investigating teams and, in all but 1 instance, a report was submitted to the Office of the Surgeon General of the Army before the evidence obtained in other locations was known.

It is seen that the incidence of clinical influenza in the 6,211 men receiving control material was 7.11 per

vaccination and the protracted incidence of disease may be involved, but no single explanation is offered at present. When these two pronounced deviations are excluded, the ratio of influenza in controls to influenza in vaccinated is 4 to 1. In some of the units, ratios of 5 or 6 to 1 were recorded.

It is of interest to note also that, in general, the difference between vaccinated and control individuals was greatest at the height of the epidemic curve and as the epidemic subsided the differential was less marked.

The results at the College of the City of New York and at Iowa, where vaccination was begun after the

epidemic was in progress, indicate that the effect of vaccine becomes evident in about one week after inoculation. In these instances the attack rates in the vaccinated and controls were not especially different during the first week but then diverged sharply. The duration of the effect is not known.

In this brief report no consideration is given to the results of serologic and virus studies which are under way and which will be incorporated in a subsequent complete report.

SUMMARY

The influence of subcutaneous inoculation of a concentrated inactivated vaccine on the incidence of clinical influenza in a series of Army Specialized Training Program units comprising approximately 12,500 men was studied during the recent epidemic of influenza A. Vaccination done shortly before or even after the onset of the epidemic was found to exert a protective effect with a total attack rate of 2.22 per cent among the 6,263 vaccinated and 7.11 per cent among the 6,211 controls, a ratio of 1 to 3.2. The influence of vaccine was most clearly evident at the height of the epidemic prevalences. The duration of the effect has not been determined.

Office of the Influenza Commission, School of Public Health, University of Michigan, Ann Arbor, Mich.

Council on Foods and Nutrition

ACCEPTED FOODS

THE FOLLOWING ADDITIONAL FOODS HAVE BEEN ACCEPTED AS CONFORMING TO THE RULES OF THE COUNCIL ON FOODS AND NUTRITION OF THE AMERICAN MEDICAL ASSOCIATION FOR ADMISSION TO ACCEPTED FOODS.

GEORGE K. ANDERSON, M.D., Secretary.

PREPARATIONS USED IN THE FEEDING OF INFANTS (*See* Accepted Foods, 1939, p. 156).

Beech-Nut Packing Company, Inc., Canajoharie, N. Y.

BEECH-NUT BRAND STRAINED VEGETABLES AND BEEF, WITH RICE AND BARLEY.

Analysis (submitted by manufacturer).—Total solids 13.95%, moisture (by difference) 86.05%, ash 1.25%, fat (ether extract) 0.56%, protein (N × 6.25) 2.91%, crude fiber 0.48%, carbohydrates other than crude fiber (by difference) 8.75%, calcium (as Ca) 0.03%, phosphorus (as P) 0.04%, iron total 7.8 parts per million, iron total available 7.3 parts per million, copper 3.1 parts per million.

Calories.—0.52 per gram; 14.74 per ounce.

Libby, McNeill & Libby, Chicago.

LIBBY'S BRAND HOMOGENIZED APPLE SAUCE.

Analysis (submitted by manufacturer).—Total moisture 85.49%, total solids 14.51%, total ash 0.32%, nitrogen 0.02%, protein (N × 6.25) 0.12%, crude fiber 0.46%, fat (ether extract) 0.02%, salt (as NaCl) 0.18%, total carbohydrates (by difference) 13.59%, calcium 2.18 mg. per hundred grams, copper 0.218 mg. per hundred grams, iron 0.30 mg. per hundred grams, phosphorus 6.22 mg. per hundred grams, lead 0.64 part per million.

Calories.—0.55 per gram; 15.62 per ounce.

Libby, McNeill & Libby, Chicago.

LIBBY'S BRAND HOMOGENIZED BEETS.

Analysis (submitted by manufacturer).—Total solids 10.25%, total moisture 89.75%, total ash 1.29%, nitrogen 0.163%, protein 1.02%, crude fiber 0.478%, fat 0.004%, carbohydrates (by difference) 7.458%, calcium 17.36 mg. per hundred grams, copper 0.145 mg. per hundred grams, iron 1.065 mg. per hundred grams, phosphorus 34.76 mg. per hundred grams.

Calories.—0.34 per gram; 9.63 per ounce.

Libby, McNeill & Libby, Chicago.

LIBBY'S BRAND HOMOGENIZED PEACHES.

Analysis (submitted by manufacturer).—Total solids 15.22%, total ash 0.33%, total moisture 84.78%, nitrogen 0.07%, protein (N × 6.25) 0.44%, crude fiber 0.34%, fat 0.01%, carbohydrates (by difference) 13.17%, salt (as NaCl) 0.15%, calcium 5.5 mg. per hundred grams, copper, 0.20 mg. per hundred grams, iron 1.01 mg. per hundred grams, phosphorus 20.40 mg. per hundred grams.

Calories.—0.58 per gram; 16.55 per ounce.

Council on Pharmacy and Chemistry

NEW AND NONOFFICIAL REMEDIES

THE FOLLOWING ADDITIONAL ARTICLES HAVE BEEN ACCEPTED AS CONFORMING TO THE RULES OF THE COUNCIL ON PHARMACY AND CHEMISTRY OF THE AMERICAN MEDICAL ASSOCIATION FOR ADMISSION TO NEW AND NONOFFICIAL REMEDIES. A COPY OF THE RULES ON WHICH THE COUNCIL BASES ITS ACTION WILL BE SENT ON APPLICATION.

AUSTIN E. SMITH, M.D., Secretary.

TYROTHRICIN.—An extract, first isolated by Dubos, obtained from Bacillus brevis, a gram-positive, aerobic, spore-forming soil organism. Tyrothricin possesses antibacterial action against several species of gram-positive organisms.

Actions and Uses.—Tyrothricin consists of at least two substances, gramicidin and tyrocidin, the former agent being by far the more active component. It seems not unlikely that some of the earlier reports which were claimed to be based on the use of gramicidin were actually concerned with the mixture. Included in the organisms that show some degree of susceptibility are species of pneumococci, streptococci and staphylococci. Its action on bacteria appears to consist, at least in part, of inhibiting enzymatic action, retarding growth and causing lysis of the bacteria against which it is effective. Its standardization is determined at present by the protection afforded mice infected with pneumococci administered intraperitoneally.

Tyrothricin should be applied locally. It is ineffective when administered orally and is ineffective *and dangerous* when given intravenously. It has been reported to be of value in the treatment of superficial indolent ulcers, the predominating organism of which is gram positive, mastoiditis, empyema and some other wound infections. Its field of usefulness is limited and it appears to exert no effect unless it can come in direct contact with the organisms. Thus it may not exert much effect in the presence of deep-seated infections. Body fluids such as saliva, urine and serum offer a slight inhibiting action, whereas substances from gram-negative organisms are decidedly inhibiting.

It may be used with caution in body cavities as long as there is no direct connection with the blood stream. But in no instance should proper surgical treatment be ignored when it is indicated. It should be remembered that, although tyrothricin appears to have a field of usefulness in medicine, its use is still in an experimental stage and much work remains to be done before its true status is established and final comparisons can be made with other antibiotics and anti-infective agents in general.

Dosage.—Tyrothricin must be applied locally, *not intravenously or by mouth.* It is administered after diluting with sterile distilled water to form an isotonic solution in a concentration which yields 500 micrograms of the drug per cubic centimeter. This concentration is usually effective against the infecting organism, although higher concentrations may be used if indicated. However, higher concentrations may be irritating to the tissues.

SHARP & DOHME, INC., PHILADELPHIA

Tyrothricin Concentrate: 1 cc. ampul of a solution of tyrothricin, 25 mg. per cubic centimeter, accompanied by a vial containing 49 cc. of sterile distilled water which contains phenylmercuric borate in a concentration of 1:50,000; 20 cc. ampul of a solution of tyrothricin, 25 mg. per cubic centimeter, not accompanied by a diluent.

ESTROGENIC SUBSTANCES (See New and Nonofficial Remedies, 1943, p. 401).

The following additional dosage form has been accepted:

THE SMITH-DORSEY COMPANY, LINCOLN, NEB.

Ampul Solution of Estrogenic Substances (in sesame oil) with Benzyl Alcohol 3%: 10 cc. Each cubic centimeter contains the equivalent of 20,000 international units of estrone. Three per cent benzyl alcohol added as a preservative.

THEOPHYLLINE ETHYLENEDIAMINE (See New and Nonofficial Remedies, 1943, p. 356).

The following dosage form has been accepted:

CHEPLIN BIOLOGICAL LABORATORIES, INC., SYRACUSE, N. Y.

Ampul Solution Aminophylline: 0.48 Gm. in 2 cc. and 0.24 Gm. in 10 cc.

VIOFORM (See New and Nonofficial Remedies, 1943, p. 121).

The following additional dosage form has been accepted:

CIBA PHARMACEUTICAL PRODUCTS, INC., SUMMIT, N. J.

Vioform Insufflate: 8 ounce bottles.