Repeated Immunization: Possible Adverse Effects

Reevaluation of Human Subjects at 25 Years

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A group of intensively immunized men, who had been subjected to detailed medical evaluations in 1956 and in 1962, was reexamined in 1971 and compared with a carefully matched control group. Clinical and laboratory studies were done to detect adverse effects induced by repeated parenteral inoculation with a variety of vaccines and toxoids. No clinical sequels attributable to long-term immunization were identified. Only one laboratory abnormality described in previous studies, elevated serum hexosamine, was observed. Few other abnormalities were detected in the immunized group; mean values were depressed for serum albumin and serum iron levels and were elevated for serum copper level and erythrocyte sedimentation rate. The findings do not exclude possible development of adverse consequences to individuals exposed to a similar course of inoculations with higher dosages or with an equally intensive course of injections with a single antigen or closely related antigens.

REPEATED IMMUNIZATION of experimental animals with large doses of antigen may result in a variety of adverse effects, including amyloidosis (1-7), multiple myeloma (8-10), and hypersensitivity (11). Similar consequences of long-term immunization have not been reported in man, possibly because subjects with analogous exposure to repeated inoculations are relatively rare. Further, vaccine dosages administered to humans are generally adjusted to contain a minimum effective antigenic content. We investigated the effects of intensive immunizations in a group of skilled laboratory workers undergoing long-term prophylactic immunization at Fort Detrick, Maryland.

In 1956, about 700 employees of Fort Detrick worked under conditions of possible exposure to virulent microorganisms and received a continuing schedule of multiple immunizations. From this group, 99 white males with the longest and most intensive history of immunization were selected for detailed medical evaluation by Peeler, Cluff, and Trevor (12); 89 of these individuals underwent a full evaluation in 1956, and 76 members of the same group were restudied in 1962 (13).*

In 1962, the average age of the subjects was 46.3 years, and the mean duration of immunization was 15.3 years. Despite multiple immunizations with a mean volume of 73.5 ml of antigen-containing fluids (13), members of the study group displayed no evidence of clinical illness attributable to repeated immunizations; however, certain abnormalities observed in the 1956 laboratory studies either persisted in some individuals or increased in incidence during the subsequent 6 years of immunization.

Abnormalities noted at the second evaluation in 1962 (13), without reference to a well-defined control population, were a relative lymphocytosis, altered values of renal and hepatic function, and weakly reactive antigammaglobulin factors. The mean value for serum hexosamine levels of the immunized group was significantly higher than that of a group of normal adults. Finally, 46 members of the study group showed merging of the a_2 and b globulins on serum protein electrophoresis on at least one occasion. Similar patterns had been reported for persons hyperimmunized with diphtheria toxin (14) and for persons with familial amyloidosis (15).

Concerning these abnormalities, Peeler, Kadull, and Cluff (13) concluded, "Whether they represent the prodromata of anatomical changes to follow or are simply interesting temporary laboratory changes of no prognostic significance will be answered only with continued observation."

Since the 1962 study, two employees who had received multiple immunizations but were not included in the original study group developed illnesses that could be related to hyperimmunization. A 52-year-old white man, immunized throughout a 12-year period, was diagnosed in 1970 as having lymphosarcoma; more recently, the diagnosis of leukemic reticuloendotheliosis was established (16). In 1971, a 34-year-old black woman, who had received immunizations for 9 years, developed an aggressive, multiple myeloma and died 3 months after diagnosis.

Also since 1962, at another institution where laboratory workers were immunized solely with

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* On both occasions, a complete history, physical examination, chest X-ray, electrocardiogram, and laboratory tests on blood and urine were evaluated for each subject.

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cholera vaccine, two men were found to have a monoclonal serum protein configuration. One had received multiple immunizations for 8 years when the gammopathy was diagnosed; 6 years later, at age 59,his status was unchanged. The other patient, age 52 at the time of diagnosis, was lost to follow-up*.

The unique opportunity afforded by the population at Fort Detrick for examination of a possible association of intensive immunization with immunologic illnesses or laboratory abnormalities prompted further study of the original group. This investigation was conducted 25 years after initiation of the intensive immunization schedule.

Materials and Subjects

Seventy-seven of the original study group were available for reevaluation in 1971. Each had a complete history and physical examination, electrocardiogram, chest roentgenogram, and the laboratory tests described below. Subjects with abnormal findings were followed at the U. S. Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Maryland, or referred to private physicians.

Available medical and postmortem records were reviewed to investigate 11 deaths in the original group. Four subjects declined participation in the present reevaluation, and five were unable to travel to Fort Detrick for examination; eight of these nine persons stated that they were in good health, and one reported gastrointestinal complaints. Only two persons from the original group of 99 were lost to follow-up.

After outpatient and immunization records and all data from the two previous studies were assembled, the entire medical record for each person was reviewed in detail. Long-term clinical outpatient records had been maintained for most subjects by a single Fort Detrick clinic throughout the period of repeated immunizations. An intense awareness of the risk of occupational infection led most subjects to seek immediate medical attention for any symptoms suggestive of infection or reaction to immunization. The control group for the present study consisted of 26 age-matched, long-term, civilian, male employees from Fort Detrick who bad never received special immunizations or been exposed to laboratory infections.

In 1971, the age range of the 77 immunized men was 43 to 79 years with a mean of 55 years; 3 persons were older than age 70. Individuals in the study group had received some number of the 21 immunogens (17-29) listed in Table 1 as well as periodic skin tests with antigens for tuberculosis (purified protein derivative), brucellosis (Brucellergen®†), tularemia (Foshay), diphtheria (Shick and Maloney), histoplasmosis, blastomycosis, coccidioidomycosis, or glanders. At least 60 of the 77 subjects received all of the immunogens listed, except poliomyelitis (41 persons) and diphtheria toxoids (37 persons). Although knowledge of the chemical composition of any antigen preparation was inadequate for an estimation of the total antigenic load, the mean volume administered before 1971 was 97 ml (range, 52.25 ml to 134.35 ml), and the mean number of skin tests was 55 (range. 6 to 93). The immunization program was discontinued 10 months before the present study when all research and development work in the Biological Laboratories at Fort Detrick was terminated.

The following laboratory tests were done on each individual in the study and control group: complete blood and platelet counts, erythrocyte sedimentation rate (Wintrobe), prothrombin time, partial thromboplastin time; rheumatoid arthritis screening test (RA Test‡), antinuclear antibody (LE Test), VDRL, and lupus erythematosus cell preparation (method of Zinkham and Conley) (30); serum

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† Merck, Sharpe & Dobme, West Point, Pennsylvannia.
‡ Hyland Laboratories, Los Angeles, California.

Table 1. Immunogens Used for Prophylaxis at Fort Detrick

Viral	Bacterial	Rickettsial
Killed Vaccines		
Eastern equine encephalitis (18,19)	Brucellosis (23)	Q fever (28)
Western equine encephalitis (19)	Plague*	Rocky Mountain spotted fever (29)
Venezuelan equine encephalomyelitis (19)	Tularemia (24)	Typhus*
Psittacosis (20)		
Rift Valley fever (21)		
Influenza*		
Live Vaccines		
Venezuelan equine encephalomyelitis (22)	Tularemia (24)	
Poliomyelitis*		
Vaccinia*		
Yellow fever*		
Other Products		
	Anthrax protective antigen (25)	
	Botulinum toxoids (25, 26)	
	Diphtheria toxoids (27)	
	Tetanus toxoid*	

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sodium, potassium, chloride, blood urea nitrogen, creatinine, phosphorus, uric acid, glutamic oxaloacetic transaminase, alkaline phosphatase, bilirubin, cholesterol, triglycerides, and free fatty acids.

Serum calcium, copper, zinc, and iron were analyzed by methods previously described (31, 32). A complete urinalysis and test for 24-hour urine protein and creatinine excretion were done. Electrophoretic analysis of serum proteins (33), serum hexosamine and glycoproteins (34), and serum lipoproteins (33) were done with the Model RB Analatrol®§.

Serum immunoglobulins were quantitated by the radial immunodiffusion method (35) within 6 hours of collection; commercially available kits for C3Î and for IgG, IgA, IgM, and IgD** were used. Experimental values represented the mean of three replicate determinations, each of which was within 10% of the mean value. The Ouchterlony immunodiffusion technique was used to evaluate reactions with specific antiserum for kappa and lambda immunoglobulin light chains**. Immunoelectrophoresis was done by the method of Scheidegger (36) on glass slides coated with 1% agarose, and patterns were developed with specific antiserums.

To study the urinary proteins, a 250 ml aliquot from a 24-hour urine specimen was dialyzed against tap water for 48 hours at 5° C, filtered, freeze dried, and reconstituted to 1/60 original volume. The radial immunodiffusion method was used to detect IgG and IgA immunoglobulin fragments. The Ouchterlony technique was used to detect IgM, kappa, and lambda antigens, and immunoelectrophoresis to detect other serum protein antigens.

Absolute lymphocyte counts, lymphocyte morphology, and the effect of stimulation of peripheral blood lymphocyte cultures with phytohemagglutinin were studied.

Lymphocyte cultures were initiated from heparinized samples of peripheral blood. Erythrocytes were sedimented with 6% dextran as previously described (37). The lymphocytes were collected and prepared for culture (37), and each culture was initiated with 2×10^5 lymphocytes in 0.5 ml of medium. At least six replicate cultures were initiated for both the control, nonstimulated samples and the phytohemagglutinin-stimulated samples. The medium was RPMI-1640†† with 25 mM HEPES* buffer, 10% heated 56°C for 30 min, fetal calf serum, penicillin 100 units per ml, and streptomycin 100 mg/ml. Phytohemagglutinin †, 1 microlitre per culture, was added at the initiation of the cultures. At the end of 5 days, 1 mg tritiated thymidine‡ (specific activity 1.9 Ci/mM or 1mCi per 2 microlitres) was added to each culture, and 24 hours later the DNA was precipitated with 5% trichloroacetic acid, and the precipitates were prepared for scintillation counting as previously described (37). Results were expressed as the difference in the mean counts per minute in the phytohemagglutinin-stimulated cultures less the mean counts per minute in the control cultures.

Unpaired *t* tests were used for statistical analysis of group means when values for both immunized and age-matched control subjects had normal

§ Beckman Instruments, Inc., Palo Alto, California.
Î Travenol Laboratories, Inc., Costa Mesa, California.
** Kallestad Laboratories, Inc., Minneapolis, Minnesota.
†† Grand island Biological Co., Grand Island, New York.

Table 2. Nonoccupational Illnesses During 25-Year Surveillance of 77 Immunized Persons*

Cardiovascular	Pulmonary
Hypertension (28)	Pneumonia (5)
Arteriosclerotic heart disease (13)	Chronic obstructive lung disease (1)
Intermittent claudication (3)	Anaerobic lung abscess (1)
Congestive heart failure (3)	Joint disease
Varicose veins (3)	Degenerative joint disease (3)
Probable rheumatic heart disease (2)	Healed osteomyelitis (1)
Thrombophlebitis (1)	Metabolic
Gastrointestinal	Hyperlipoproteinemia (13)
Peptic disease (13)	Diabetes mellitus (4)
Hemorrhoidectomy (4)	Gout (1)
Cholecystectomy (4)	Cancer (*see* text)
Hiatal hernia (3)	Skin carcinoma (2)
Rectal cryptitis (2)	Adenocarcinoma of colon (1)
Diverticulitis (1)	Transitional cell carcinoma of bladder (1)
Pilonidalcyst (1)	Miscellaneous
Genitourinary	Inguinal hernia (20)
Renal stone (4)	Obesity (12)
Urinary tract infection (2)	Herniated nucleus pulposus (3)
Pyelonephritis (2)	Cataracts (2)
Hematuria, etiology unknown (2)	Ulnar neuritis (1)
Varicocele (2)	Glaucoma (1)
Ear, nose, throat	Familial clubbing (1)
Sinusitis (7)	Colloid degeneration of macula (1)
Meniere's disease (2)	Meningoencephalitis (1)
Deviated nasal septum (1)	
Perforated nasal septum (1)	
Otitis (1)	

* Numbers within parentheses are the number of individuals affected.

distributions and essentially equal variances. When data did not conform to this criteria, appropriate modifications of the *t* test were used. Median values for immunized and control groups were compared by chi-square analysis, and the Wald-Wolfowitz runs test (38) was used to compare sets of values. In addition, when frequency distributions for both groups were normal, chi-square analysis was used to compare the incidence of values that were more than 2 standard deviations from the control group mean (95% confidence limits).

Results

CLINICAL EVALUATIONS

Review of all available information showed that six individuals had a history of an infectious illness categorized as "occupational"; their clinical symptoms were accompanied by a fourfold rise in specific antibody titer. Two subjects had a history of tularemia, two of brucellosis, one of Q fever pneumonitis, and one of paratyphoid B enteritis. Asymptomatic fourfold increases in serologic titer for tularemia or brucella occurred in five additional subjects. Frequency of nonoccupational illness is shown in Table 2. These illnesses included four reports of carcinoma: two of the skin; one of the colon, with apparently successful resection in 1965; and one transitional cell carcinoma of the bladder that was successfully removed by surgery. Thus, no clinical illness that could be attributed to repeated immunization was found in any of the 77 subjects.

LABORATORY EVALUATIONS

Laboratory determinations were designed to explore thoroughly those areas that were abnormal in the previous studies and to examine the current status of the 77 individuals.

There were no significant differences between the mean values of study and age-matched control groups for hemo globin, hematocrit, and prothrombin time, or for leukocyte, eosinophil, or platelet counts (Table 3). Absolute lymphocyte counts were within normal limits for all persons, and mean values for the two groups were not significantly different. Although, median chi-square analysis of erythrocyte sedimentation rates showed no significant between-group differences, the frequency distribution of values for individuals in the immunized group was asymmetric, and analysis of group means by *t* test and of sets by Wald-Wolfowitz runs test indicated that the sedimentation rate was significantly increased in the immunized group. Values for partial thromboplastin time were prolonged for 19 immunized persons but were within normal limits for all control subjects. ($P < 0.01$).

Tests for lupus erythematosus, rheumatoid arthritis, and antinuclear antibody were negative for all immunized subjects.

Renal function in the immunized group was at least equivalent to that of the age-matched control group (Table 4). The mean value for 24-hour urine protein excretion of immunized subjects was lower, and the mean value for creatinine clearance was higher than in control subjects. Likewise, there was no evidence that liver function was impaired by immunization. Whereas the incidence of elevated levels of serum alkaline phosphatase activity, bilirubin, and serum glutamic-oxalacetic transaminase (SGOT) was higher than in the earlier studies, it was essentially the same for both groups, and mean values did not differ significantly.

As in 1962, the mean value (±SE) for the serum hexosamine in the immunized group (87.4 mg/100 ml ±1.5) was significantly higher ($P < 0.001$) than the value for the control group (77.2 mg/100 ml ± 2.1).

Serum electrophoretic studies indicated that there were slight but statistically significant differences by unpaired *t* test in group mean values for albumin, a_2 globulin, and *b* globulin, as shown in Table 5. In contrast, when these data were examined by chi-square analysis for incidence of values more than 2 standard deviations from the control mean, the two groups differed only with respect to albumin ($P < 0.001$). Electrophoretic patterns showed no evidence for the previously reported abnormality of a_2 and *b* globulins (13).

* Grand Island Biological Co., Grand Island, New York.
† PHA-P, Difco, Detroit, Michigan.
‡ Schwartz-Mann Biochemicals, Orangeburg, New York.

Table 3. Hematologic Values for Immunized and Control Groups

Comparison	Test	Value	
		Immunized Group (77 subjects)	Control Group (26 subjects)
Group mean	Hemoglobin, *g/100 ml*	15.7	15.8
	Hematocrit, %	47.7	48.5
	Total leukocytes per mm^3	7 400	7 460
	Platelets per mm^3	285 000	281 000
	Sedimentation rate, *mm/h*	12.6*	6.7
Number	Prolonged prothrombin time, *s*	14	8
	Prolonged partial thromboplastin time. *s*	25†	0

* $P < 0.001$. † $P < 0.01$.

Table 4. Incidence of Renal and Hepatic Function Abnormalities in Immunized and Control Groups

	Immunized Group (77 subjects)		Control Group (26 subjects)	
	n	%	*n*	%
Blood urea nitrogen level elevated	12*	16	0	0
Creatinine level elevation >1.5 mg/100 ml	0	0	0	0
(Mean creatinine clearance, *ml/min*)	(113)		(95)	
Urine protein level > 200 mg/24h	5	6	0	0
(Mean 24-h urine protein level, mg)	(115)		(132)	
Alkaline phosphatase level elevated	12	16	3	12
Bilirubin level elevated	8	10	2	8
Serum glutamic oxalacetic transaminase level elevated	5	6	3	12

* All <30 mg/100 ml.

Evaluation of serum trace elements was done because of changes reported during acute infection in man (32). There were no significant between-group differences for levels of serum calcium, phosphorus, or zinc, but in the immunized group the mean value was depressed for serum iron concentration (98.7 ± 4.0 mg/100 ml, control 139 ± 9.5 mg/100 ml, $P<0.001$) and was elevated for serum copper (131 ± 34 mg/100 ml, control 98 ± 6.2 mg/100 ml, $P<0.001$).

Reports of increased concentrations of serum glycoprotein constituents and globulin fractions during febrile, acute, systemic infection and afebrile, sterile inflammation (39) prompted evaluation of serum lipids and globulin fractions of serum glycoproteins and lipoproteins. Again, no significant differences were found between groups.

Serum concentrations of IgG, IgA, IgM, or C3 were similar for both groups (Figure 1). Although mean IgD levels were not significantly different, the incidence of undetectable values (<2 mg/100 ml) was greater in the control group. No monoclonal or Bence Jones proteins were observed in serum or urine.

The mean lymphoproliferative response to phytohemagglutinin was not significantly different for the immunized subject group and age-matched control group (Figure 2). Seven individuals in the immunized group had stimulation values lower than 2 standard deviations from the mean of the normal, whereas two of the immunized group had values 2 standard deviations above the normal mean.

MORTALITY DATA

By 1971, 15.5 years after their selection for study, 11 of the 99 immunized persons had died, a mortality rate in agreement with the 10.76 deaths predicted by actuarial data*. Arteriosclerotic heart disease was the cause of death in four persons, cancer in three (oat-cell carcinoma of lung, colon adenocarcinoma, brain tumor†), and chronic lung disease in two. Two other persons died suddenly without postmortem examination; one, on insulin treatment, had shown low voltage on his last recorded electrocardiogram (ECG), and the other had shown left bundle-branch block and premature ventricular contractions on ECG.

Tissue sections obtained from four postmortem examinations and one biopsy showed no evidence of amyloidosis (Congo red stain and thioflavin-T). Reexamination of available necropsy tissue (Congo red stain) for amyloid deposition in spleen, liver, lymph nodes, heart, lungs, or bowel confirmed the original findings.

* Supplied by Metropolitan Life Insurance Company.
† Biopsy of the brain lesion showed only necrotic tissue. No other cancer was found before death; no postmortem examination was done.

Discussion

Chronic stimulation of the immunoglobulin-producing system in man is thought to be associated with amyloidosis, plasma cell dyscrasias, and autoimmune diseases (40). Amyloidosis may appear secondary to chronic inflammatory diseases, such as rheumatoid arthritis or glomerulonephritis, and has been reported in a man who received more than 600 blood transfusions (41). Three cases of multiple myeloma (42, 43) and one of Waldenstrom's macroglobulinemia (44) have been reported in patients who underwent desensitization for treatment of allergies.

The bulk of available information about possible adverse effects of intensive immunization, however, is derived from experimental animals in which diseases with human analogues have been induced. Many investigators have examined the response of rabbits to hyperimmunization with certain streptococcal vaccines (45). Formation of cryoprecipitating serum protein has been shown; induction of a uniform (monoclonal) antibody that seemed to be genetically controlled has been observed in some rabbits (46). The development of factors that mimic human rheumatoid factor or anti-

DNA antibody have been reported (47). Experimental production of amyloidosis (1-7) and of multiple myeloma (8-10) in animals has been studied intensively.

Table 5. Serum Protein Levels

Fraction	Immunized Group (n = 77)	Control Group (n = 26)	Significance
	g/100 ml (mean ± SD)		P
Albumin	4.01 ± 0.39	4.31 ± 0.33	<0.001
Alpha$_1$ globulin	0.18 ± 0.06	0.19 ± 0.07	—
Alpha$_2$ globulin	0.59 ± 0.13	0.68 ± 0.13	<0.01
Beta globulin	0.66 ± 0.12	0.73 ± 0.10	<0.01
Gamma globulin	0.94 ± 0.23	0.92 ± 0.22	—

The increasing number of antigens recommended for routine clinical use in man imposes a concomitant need for information on the possible adverse effects of repeated exposure to antigenic stimuli. Despite cautious extrapolation from animal findings to man, evaluation of these potentially adverse effects remains speculative because few intensively immunized human populations have been available for study. In one report, mortality analysis for 18,000 soldiers given influenza vaccine in an emulsified oil adjuvant showed that the incidence of malignant neoplasms, allergic disease, or collagen disease was essentially the same for vaccinates immunized with or without adjuvant (48).

In our study, reexamination of clinical or postmortem findings for 88 of 99 repeatedly immunized individuals failed to produce evidence that development of neoplasms, amyloidosis, or autoimmune diseases was associated with the vaccine dosages and frequencies used at Fort Detrick. It is especially noteworthy that persons selected in 1962 for gum or kidney biopsy on the basis of multiple laboratory abnormalities now show no important clinical or laboratory findings. It is doubtful that the "nonsecretory" plasma cell myeloma reported by Azar and associates (49) was overlooked, because clinical and laboratory findings showed no evidence of signs reported to be present in all such cases, such as osteolytic lesions, hypogammaglobulinemia, and mean survival time of 7.5 months.

The evaluation in 1962 (13) suggested that laboratory abnormalities might be transient because there was no continuing abnormality in some individuals and seven men who had not received an immunization within the preceding 2 years had no antigammaglobulin factors. Likewise, the absence of an abnormal serum protein electrophoretic pattern may be related to reduced schedules of immunization after 1962 or to the 10-month interval between the last antigenic exposure and our evaluation.

The only abnormality noted in both this and previous studies was the elevation in serum hexosamine levels. Hexosamine was initially measured in these subjects because it is found in tissue amyloid deposits (50) and is elevated in the serum from some humans with amyloidosis (5). No direct relation is apparent between elevated serum hexosamine level and amyloid; substances indistinguishable from amyloid can be created from material entirely free of carbohydrate constituents (51-56). Hence it is unclear whether the elevated hexosamine levels reflect inflammatory stimulation or some other characteristic of the immunized group.

Other unexplained differences between groups have been noted in these tests: erythrocyte sedimentation rate; serum iron and copper levels; serum albumin, a_2 globulin, and b globulin values; and partial thromboplastin times. The significance of findings for the a and b globulins is less impressive because most values for the immunized subjects fall within the 95% confidence limits of the control mean (mean ± 2 SD). Some chronic inflammatory effect of repeated immunization might be responsible for such abnormalities as elevated sedimentation rates, elevated serum copper levels, or depressed serum iron values, but no relation between immunization and the other abnormalities is suggested.

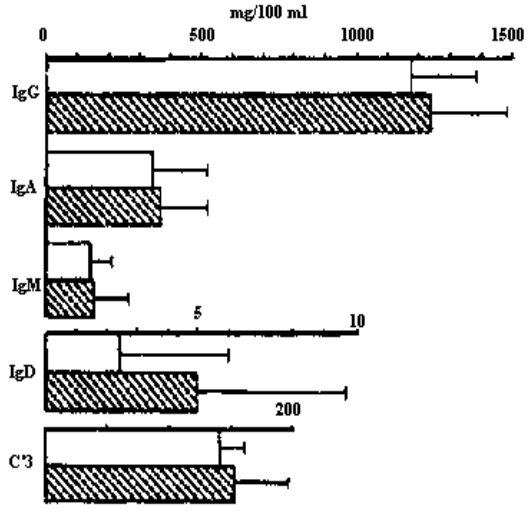

Figure 1. Group mean values ± 1 SD for serum immunoglobulins and C3 component of complement for the immunized subjects (shaded bars) and age-matched (clear bars).

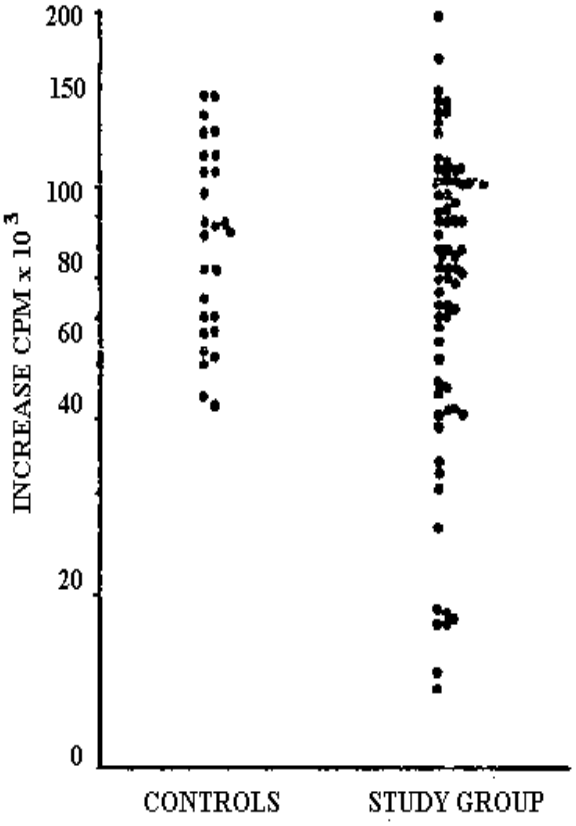

Figure 2. Phytohemagglutinin-induced increases in ^{2}H-thymidine incorporation in lymphocyte cultures initiated from peripheral blood lymphocytes of control and immunized subjects. Results are expressed as a count per minute (CPM) of a phytohemagglutinin-stimulated culture less the count per minute of a paired, nonstimulated culture from the same subject.

The ability to stimulate lymphoid cells with phytohemagglutinin has been reported to be a measure of thymic function in mice (57) and humans (58). Mice, either with tumors or repeatedly immunized, have a decreased ability within their thymic lymphoid cells to respond to phytohemagglutinin in culture (59). This decrease may result from shifts in population of cells in their lymphoreticular tissue with a relative decrease in the phytohemagglutinin-reactive population (60). Our study did not show any wholesale deficiency of phytohemagglutinin response in lymphocytes from the repeatedly immunized individuals. This may be caused by the lapse of time between the last immunization and the date of testing, since the depression of the phytohemagglutinin responsiveness of spleen cells is dependent on continual contact with tumor or other antigens (59-61); or it may be that in humans, immunization has little effect on a circulating phytohemagglutinin-responsive population.

These data and the accompanying evaluation of an intensively immunized population provide evidence that no obvious adverse effects result from repeated immunization. Although abnormal laboratory findings were observed in comprehensive studies of those subjects after 10, 16, and 25 years of immunization, the absence of documentation of corresponding clinical illness or consistency in laboratory abnormalities from study to study was notable. Thus, this group provides reassurance that schedules for routine immunization with a diversity of vaccines should not produce untoward effects merely because of frequency of inoculation. Nevertheless, the presence of two persons with neoplastic disease of lymphoid origin in the total immunized population (by 1970) of approximately 1500 individuals at Fort Detrick suggests that continued surveillance of the entire group of repeatedly immunized persons is warranted. Moreover, it is possible that continuing stimulation of a restricted population of immunoreceptor cells by repeated exposure to a single antigen or closely related group of antigens (such as multiple blood transfusion, allergy desensitization, chronic infection) might initiate immunologic abnormalities or constitute a predisposing condition for their development.

ACKNOWLEDGMENTS: The authors thank Mrs. Shirley Jewell for secretarial services, Karen Bostian for technical assistance, and Peter J. Bartelloni, LTC, MC, G. L. Cockerell, CPT, VC, John Warren, MAJ, MC, and William R. Beisel, M.D. for their advice and assistance.

Received 5 April 1974; revision accepted 15 July 1974.

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