fort chart to be applied for short periods of time, the temperature and humidity must vary with the outside conditions, so that the shock of the sudden change will not be too great when passing from one place to another.

IN CONCLUSION

Each individual patient we see will present entirely different requirements for the conditions of the air to be supplied. The results shown by the various investigators, whom I have quoted, show this to be the case. For example, the nursery for the premature infant should be maintained at a relative humidity of from 60 to 65 per cent, and a temperature of from 80 to 100 degrees Fahrenheit, depending upon the age and weight of the patient. The asthmatic, on the other hand, responds much more quickly if the relative humidity is kept at 40 per cent. The patient who has a stuffy nose in an atmosphere of high relative humidity responds nicely to a humidity of from 48 to 50 per cent, and a temperature of about 68 degrees Fahrenheit.

In the extremely dry climates of the interior of our western states it will be necessary, at times, to add water to the air circulated to increase the humidity. Patients in these regions frequently complain of the extreme dryness and irritation of the mucous membranes of the nose and throat.

Our hospitals should be conditioned by the unit system so that individual rooms, or groups of rooms, can be maintained at any desired state. They can accommodate, in this way, any type of patient that may require care.

I have endeavored to outline, briefly, the subject of air-conditioning as it has been developed by the leading ventilating engineers. Its value in the management of diseases of the upper respiratory tract has been shown by many investigators. I hope this paper will serve as a stimulus for all of us to become more conscious of the atmospheric environment of our patients in the future.

384 Post Street.

POLIOMYELITIS*

IN VITRO NEUTRALIZATION TESTS, USING NORMAL ADULT AND CONVALESCENT HUMAN SERUMS

I. INTRODUCTION

By BEATRICE F. HOWITT, M.A.
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DISCUSSION *by K. F. Meyer, Ph. D., San Francisco.*

DURING the outbreak of poliomyelitis in northern California in 1934, serum was collected both from patients who had recovered from the disease, and from so-called "normal" individuals. The pooled convalescent serums were given therapeutically to the active cases, four to twelve persons contributing toward each mixture. The pooled serums from adults having a negative history of poliomyelitis were used prophylactically for contact cases, eight to sixteen individuals contributing toward each of these.

The use of normal serum has been founded on the observations of numerous workers, who have estimated that between 70 and 80 per cent of the normal adult urban population possess neutralizing antibodies for the poliomyelitic virus. Based on these reports, and those of Davide [1] and of Flexner and Stewart,[2] for the prophylactic use of convalescent serum, Moro,[3] in Germany, suggested that "normal" adult serum, or the whole blood, should be administered prophylactically to young children in an epidemic. This type of passive immunization was used by Brebner [4] in the United States during 1932.

The normal serum collected in 1934 for the outbreak in northern California was divided into two classes, one obtained from the medical staff of the University of California Hospital and the other from outside volunteers. It was thought that members of the staff might have had greater opportunities for exposure to poliomyelitis and, therefore, possess more antiviral substances in their blood than the average adult. Serum was also collected from a group of professional donors who might be called on for direct transfusions if they proved suitable.

To determine the relative values of these several groups, *in vitro* neutralization tests were made on many of the normal and the convalescent pools, respectively, as well as on individual serums. The results will be presented, even though many workers have reported upon observations of a similar nature. All such data, however, may prove valuable as an aid in directing the course of public health policy during an epidemic or a time of sudden emergency.

II. METHODS

All neutralization tests were carried out with a standard dilution of 5 per cent cord suspension of a monkey passage virus kindly sent by Dr. M. Brodie of the New York University. Suspensions made from the same cord mixtures of one group of monkeys was used as the stock virus throughout all the different experiments over a period of several months. The 5 per cent suspension, made fresh about once a month, lightly centrifugated, maintained about the same degree of potency throughout this period, the M. L. D. being approximately a dilution of 1-800 or 0.00125 cubic centimeters. The standard amount used for the tests was a 1-25 dilution (0.04 cubic centimeters) or about 32 M. L. D. As many tests as possible were performed at one time for better purposes of comparison. Equal parts of the 5 per cent diluted virus, and the pools or single serums were mixed, placed at 37 degrees centigrade for two hours, and then kept overnight in the ice-box before injecting 1.5 cubic centimeters of each intracerebrally into monkeys, according to the method of Shaughnessy, Harmon and Gordon.[5] No preservative was put in the serums.

III. NORMAL ADULT SERUMS

Because of the high incidence of poliomyelitis among the older age groups (35.41 per cent) in

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California, the endeavor was to accept blood of normal volunteers only from those individuals over thirty or thirty-five years, presuming that immunity increased with age.

As shown in Table 1, individual serums were tested from twenty-six normal professional donors varying in years from twenty-three to forty-six, averaging thirty-one. The undiluted serum of seventeen (65.3 per cent) neutralized the virus, while nine, or 34.7 per cent, failed to do so. Nine of ten serums proved potent in a dilution of 1-10, and six out of nine in a 1-40. The one serum tested at 1-80 failed to neutralize.

These figures are fairly in agreement with those given by Schultz and Gebhardt [6] for San Francisco (56.2 per cent), and are nearly the same as those presented by Weyer, Park and Banzhaf [7] for New York City (66.6 per cent), although Jungeblut [8] gives a lower value (56.6 per cent) for the latter. Most of the other reports show higher percentages for urban populations.

Of the normal pooled serums, five were mixtures from the medical staff and six from adult outside volunteers. In the first group, four pools contained four individual serums each and one had seven, while in the second, three pools contained eight serums each, two had thirteen, and one had sixteen. The average age incidence was thirty-five years for the first group, and thirty-nine for the second. All serums neutralized when undiluted. Four of each group were also positive when diluted 1-10. Upon further dilution the pools from volunteers seemed to be slightly weaker, although one of each went to 1-60, while none was positive at 1-80. These figures ran slightly higher than those given by Jungeblut [8] for normal pools. It also showed that there was practically no difference in potency between the supposedly better grade pools of serum than those chosen at random from the adult city population. However, there was a larger average number of individuals in each of the latter pools.

IV. POOLED CONVALESCENT SERUMS

Comparison was made with eighteen pools of convalescent serums chosen at random from adults and children having had the disease at varying periods. These contained from seven to eleven different serums, but several had only two or three. All except one neutralized when undiluted. The negative one was composed of only two serums, so that it could hardly be classed as a real pool. Eight serums were titrated further (Table 1), but failed to show quite the degree of potency as given by the normal serums. Of eight diluted 1-40, five (62.5 per cent) failed to neutralize, while of nine normal pools only two (22.2 per cent) were negative at this dilution.

This somewhat decreased strength shown for the convalescent serums is also in agreement with the recent work of Jungeblut,[9] but contrary to that of Brodie,[10] who found the normal serums weaker in New York. It would conform, however, with reports given by Howitt,[11] and by Paul and Trask,[12] for poorly neutralizing individual convalescent serums.

TABLE 1.—*Neutralization Tests on Normal and Convalescent Serums**

Type of Serum	Undiluted Serums			Dilutions of Selected Serums or Pools																	
				1-5			1-10			1-20			1-40			1-60			1-80		
	Total Number	+	0	Total Number	+	0	Total Number	+	0	Total Number	+	0	Total Number	+	0	Total Number	+	0	Total Number	+	0
Individual normal	26	17 (65.3%)	9 (34.7%)	..	..	..	10	9	1	1	0	1	9	6	3	..	..	..	1	0	1
Individual convalescent	18	12 (66.6%)	6 (33.3%)	5	3	2	12	7	5	5	3	2	12	5	7	..	..	..	..	..	..
Pooled normal — House staff 5		5 (100%)	0	..	..	..	5	5	0	..	..	..	4	4	0	1	1	0	1	0	1
Pooled normal — Outside volunteers 6		6 (100%)	0	1	1	0	5	5	0	..	..	..	5	3	2	1	1	0	2	0	2
Pooled convalescent	18	17 (94.4%)	1 (5.6%)	1	1	0	8	5	3	..	..	..	8	3	5	5	3	2	2	0	2

* +=Neutralization.
0=No neutralization.

TABLE 2.—*Neutralization tests on Selected Groups of Serums**

Series	Selected Pools or Serums	Number Tested	Dilutions of Pools or Serums 1-5	1-10	1-20	1-40
I Pools	House staff over 35 years	1	....	+	....	+
	House staff under 35 years	1	....	+	....	+
	Normal adults over 35 years	1	....	+	....	+
	Normal adults under 35 years	1	....	+	....	+
II Pools	Convalescent in 1934	1	+	....	+	....
	Convalescent for two years	1	+	....	+	....
	Convalescent for four years	1	+	....	+	....
	Convalescent for ten years	1	+	....	+	....
	Convalescent for twenty years	1	+	....	0	....
	Convalescent over twenty years	1	+	....	+	....
III Individual	Convalescent in 1934	2	One + One 0	....	One + One 0	....
	Convalescent for four years	1	+	....	+	....
	Convalescent over twenty years	2	One + One 0	....	+	....

* +=Neutralization.
0=No neutralization.

V. COMPARISON OF SELECTED POOLS OF NORMAL SERUMS

Comparison was made of selected groups of serums from normal adults. Six were pooled from normal outside volunteers, and six from members of the medical staff over thirty-five years of age. A similar number was also pooled from the two groups of those under thirty-five years, but not below twenty-five. As shown in Table 2, all neutralized the standard dose of virus in a dilution of 1-40, again demonstrating the comparable effectiveness of the normal pooled serums.

It was also interesting to note that of two laboratory workers having had intimate contact with the poliomyelitic virus over a period of years, the serum of one neutralized in a dilution of 1-40 but not 1-80, while that of the other failed to neutralize even undiluted.

VI. SELECTED GROUPS OF CONVALESCENT SERUMS

To determine the potency of convalescent serums when pooled from individuals after varying periods of recovery, selections were made from different groups convalescent for a few months and for 2, 4, 10, 20, and over 20 years, respectively, as shown in the table. All except the serum of twenty-year duration neutralized in 1-20 dilution, even after a forty-year recovery.

Five individual serums were tested in the same way, two from recent convalescents in 1934, one from a patient recovered four years and two over twenty years. One of the recent serums failed to neutralize, while one was potent in a 1-20 dilution. Of the serums from older cases, both neutralized in a 1-20, although one failed in a 1-5 dilution.

It was interesting that the nonpotent serum of 1934 also failed to neutralize a recently isolated strain of virus obtained during the late outbreak. It was likewise found that unpreserved serum from a convalescent case was still potent after storage in the ice-box for one year and six months, while a second one lost its efficacy only after storing two and one-half years.

Serums were selected from two groups of convalescent patients, one having been given serum treatment during the attack of poliomyelitis and the other not having received it. All the individuals chosen were over eighteen years of age and had acquired the disease from several months to twenty years previously. Neutralization tests were performed both on the undiluted serums, and when diluted to a maximum of 1-40. As shown by Table 3, the results were fairly comparable for each group. Four out of seven (57.1 per cent) neutralized in the series given convalescent serum, while six out of nine (66.6 per cent) were positive in the untreated series. The advantage was more in favor of the latter group, both when undiluted and in the different dilutions. In comparing the total number of individual convalescent serums tested in a 1-40 dilution with those of the normal group in like dilution, the advantage lies with the normal serums, six out of nine (66.6 per cent) showed positive neutralization when diluted 1-40, while only five out of fourteen, or 35.7 per cent, were positive in the recovered group.

VII. DISCUSSION

The results of this small series of neutralization tests again indicate that the use of normal pooled adult serum from an urban population is justified in an epidemic of poliomyelitis. Potency tests in monkeys show an equal if not greater average degree of neutralizing ability among the positive so-called "normal" groups than among those recovered from the disease. However, since only a certain percentage of the urban population possess antiviral substances, individual serums chosen at random should not be relied upon for potency without a preliminary test in monkeys. On the other hand, normal serums pooled in groups of not less than ten or twelve persons apparently can safely be used in the central Californian region without previous testing.

Although there has been discussion, upheld by experimental data, to show that the strain of virus employed in the *in vitro* neutralization test may influence the outcome, yet if the same virus is similarly employed throughout the different series, comparative results should be of value, even

TABLE 3.—*Neutralization Tests on Serums of Convalescent Patients**

Serum Treatment	Patient	Serum Dilutions				
		Undiluted	1-5	1-10	1-20	1-40
Given	G. F.	0	0	....	0	....
	J. N.	+	+	....	0	....
	R. N.	+	0	....	0	....
	A. H.	0	0	....	0	....
	R. B.	+	....	+	....	+
	G. L.	+	....	+	....	+
	M. A.	0	....	....	....	....
Not given	W. H.	+	0	....	....	....
	M. C.	+	....	....	....	+
	H. B.	+	....	+	....	0
	R. G.	+	....	+	....	+
	H. M.	+	....	+	....	0
	A. F.	0	....	0	....	0
	J. O'R.	0	....	0	....	0
	M. H.	+	....	+	....	+
	A. J.	0	....	0	....	0

* +=Neutralization.
0=No neutralization.

though the percentage of positives may have differed when using a recently isolated or a "passage" strain of virus. This question of the desirability of using various strains has recently been ably discussed by Paul and Trask.[12]

Because the *in vitro* neutralization test has been the usual criterion for judging the potency of a serum, one would feel justified from the results here given in recommending the administration of normal adult pooled serums collected at random from urban dwellers over thirty-five years of age. It would, of course, be of more advantage to have on hand a list of tested donors whose serums could be pooled in periods of emergency, or who could be used individually for direct transfusions. From the data available one would have more confidence in the potency of normal adult serums selected from an urban population than in those collected from an average group of convalescent cases, mostly children. These observations are in accord with those of Jensen [13] in regard to the therapeutic use of serums in poliomyelitis. This author reports that, during the Danish outbreak in 1934, pooled serums obtained from abortive cases of poliomyelitis were found to have higher neutralizing ability than those from frankly paralytic cases.

The question of the efficacy of all these serums after administration to the patient is not being discussed here, but merely the theoretical value as demonstrated by the *in vitro* neutralization test. Undoubtedly, from this standpoint, many serums are effective, although varying in degree of potency, while from the clinical point of view the the subject is apparently a matter of controversy.[18]

VIII. SUMMARY

Seventeen (65.3 per cent) of twenty-six undiluted serums from normal professional donors neutralized a standard amount of poliomyelitic virus, while nine (34.7 per cent) lacked potency.

Pooled undiluted serum from members of the medical staff, as well as from normal adult outside volunteers, all neutralized the same amount of virus. Ten of them neutralized in a 1-10 dilution, seven out of nine in a 1-40, while none was positive after diluting 1-80. Eighteen pools of convalescent serum were tested, and all but one neutralized when undiluted. Titrations of the latter showed a lower degree of potency than did the normal serums.

Selected lots of serums from staff members over and under thirty-five years of age, respectively, as well as from normal adult outside volunteers of the same age groups, all neutralized the virus in a 1-40 dilution.

Titration of convalescent serums in lots from cases having had poliomyelitis, respectively, in 1934, and 2, 4, 10, 20, and over 20 years previously, showed a positive neutralization in a 1-10 dilution. Several individual serums from recently recovered patients showed less potency than those from convalescents of longer duration, even over forty years. There was very little difference in the average degree of potency between two groups of serums from convalescents, one having received serum treatment during the disease and the other not being treated.

From a comparison of the data presented, it would seem justifiable to use pools of normal adult serums from an urban district in the event of a sudden outbreak of poliomyelitis, even though the confirming neutralization test was unavailable at the time.

Hooper Foundation for Medical Research.

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DISCUSSION

K. F. MEYER, Ph. D. (Medical Center, Third and Parnassus Avenues, San Francisco.)—The paper by Miss B. Howitt does not consider the therapeutic or prophylactic use of antiviral serums in poliomyelitis. Two statements summarize the position taken by competent clinicians who feel that the case for or against the use of serum has not been made. Thus Ledingham (Bulletin, Johns Hopkins Hospital, June, 1935, Vol. 56, p. 346), says:

"Some physicians are prepared to accept the view that convalescent poliomyelitis serum is of value therapeutically in the preparalytic stage and act accordingly, but probably a majority believe that no evidence so far has been forthcoming that such serum exerts any therapeutic effect whatever. In any case accurate controlled statistics in support of the procedure are not yet available. I believe, however, that there is a field for antipoliomyelitis serum as a prophylactic for contacts; though here again it will be difficult to get the necessary statistical controls.

"A recent serious outbreak of poliomyelitis in Denmark, in which extensive prophylactic use has been made of antipoliomyelitis serum from the horse, may possibly afford some evidence for or against its value in such circumstances. This antipoliomyelitis serum prepared in the horse has high neutralizing value—considerably higher than that of human convalescent serum—when tested on monkeys."

Probably the majority will concur in the remark by C. Jensen (Proceedings of the Royal Society of Medicine, June, 1935, Vol. 27, p. 1024): "No real proof has hitherto been given that convalescent serum is without effect."

In view of the unfortunate and distressing accidents which have and will accompany the present methods of active immunization, it may indeed be advisable to reconsider the position taken with respect to the use of the antiviral serums. The observations made and the conclusions drawn by Stokes and associates (*American Journal of Diseases of Children*, 1935, Vol. 50, p. 581), deserve consideration in this connection.

THE RÔLE OF THE URETHRA IN FEMALE UROLOGY*

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DISCUSSION *by Edward W. Beach, M. D., Sacramento; F. S. Dillingham, M. D., Los Angeles; James Steinberg, M. D., Los Angeles.*

ALTHOUGH several excellent papers on the female urethra have appeared in the literature during the last few years, it will be necessary to emphasize the importance of this organ for a longer period of time before the frequency and significance of its lesions are recognized by the majority of the medical profession. My attention was first called to the importance of this organ about sixteen years ago because of the frequency with which the introduction of an examining cystoscope was followed by marked alleviation of symptoms, although these were often suggestive of upper urinary tract pathology.

In a recent study of 425 female patients with urinary disturbances, the urethra was entirely responsible in 67 and at least partly responsible for the symptoms in 328 cases. Lesions of the generative organs were responsible etiologic factors in 90, and other organs not connected with the urinary or generative tracts in seven. These figures correspond to those resulting from a former study of 234 other cases in which urethral lesions were apparently wholly responsible in 56, and partly responsible in 173 instances. Cases of urethritis in which the gonococcus could be demonstrated were not included, otherwise the numbers would have been larger.

The most common pathological conditions of the urethra were: strictures at the external meatus or in the canal, urethritis usually associated with trigonitis, polyps and polypoid formations, papillomata and caruncles. Three of the latter were associated with strictures. Carcinoma occurred in three, sarcoma in one, urethrovaginal fistula in one, and a calculus in one of these cases. I have seen but three urethrovaginal fistulas and one urethral calculus in several thousand women with urinary tract pathology coming under my observation.

Concomitant pathology was found in other portions of the urinary tract in 92 and in the generative organs, most often in the cervix, in 63 of the present series of cases.

EXAMINATION OF THE FEMALE URETHRA

It is often advisable to use three types of instruments in the examination of the female urethra—a skenoscope, a female urethroscope with the lamp at the distal extremity, and a near-vision cysto-urethroscope using water dilatation. If the latter is not used, those pedunculated papillomata and polyps, which lie against the wall of the urethra, may be overlooked. A properly performed, two-glass test is of value in the female as well as male patients, especially when infection is limited to the urethra. Palpation through the vagina will aid in the detection of tumors, foreign bodies, thickening and induration of the urethra, urethroceles and obstruction from extrinsic causes such as displacement of the uterus, fibroid tumors and other growths.

URETHRITIS

Urethritis was found in 150 of this series of 425 patients. Although apparently second in frequency to strictures, with which it is often associated, many cases of urethritis were probably not detected because of the impracticability of satisfactory urethroscopic examination in the presence of urethral obstruction.

Urethritis usually occurs as a diffuse more or less granular inflammation involving the entire urethra, or a limited area more commonly in the posterior portion of the canal. Infection with the gonococcus is the most common etiological factor, although the colon bacillus and other organisms are frequent offenders. Urethritis, due to infection by the latter, with or without infection of the bladder and kidneys, is more common in the female, regardless of age, than is generally appreciated.

Many of our urethritis cases were associated with infection of the cervix. Infection of the urethra from this source and ascending lymphatic infection of the kidneys from the urethra, are

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