

Chronology 1

CHRONOLOGICAL ACCOUNT OF PROCEDURES

Experiment 1

Date - May 13, 1947

Place - Asylum

With the decision to attempt human inoculation the choice had to be made whether to use human or animal passage *T. pallidum* for inoculation. Obviously the use of the rabbit testicular syphiloma would be the most advantageous if it were found to be virulent for man. From unpublished experimental data (17) there was reason to expect that the Nichols rabbit strain which was planned to be utilized would be virulent.

To study the effectiveness of various types of prophylaxis it was our feeling that the method of infection should be as similar as possible to the natural manner of infection. It was then our impression that infection in normal intercourse took place through an intact mucosal surface. Therefore, it was decided to use the closest approximation to the normal method of infection, i.e. local application to the intact penile mucosa. As a further test of the virulence of the Nichols rabbit strain the method of intracutaneous inoculation was planned to assure that spirochetes were actually introduced into the body. Thus the first experiment was set up to determine the following:

1. The virulence of Nichols strain *T. pallidum* obtained from a testicular syphiloma of a rabbit 32 days following inoculation.
 - a. When applied to the intact mucous membrane of the penis.
 - b. When injected intracutaneously.

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2. The effectiveness of the orvus-mapharsen prophylaxis following exposure of the penis in the event of virulence of the organism under the experimental conditions of local application to the penile mucous membrane of the control group.

Patients were selected who had long foreskins and the method of local application (q.v.) was used in experiments 0101 and 0103. The only cleansing preparation was to remove excess smegma by gently wiping with a cotton pledget.

Exposure of the control (Exp. 0103) and prophylaxis group (Exp. 0101) took 1 hour after which the inoculating pledgets were removed and the foreskin pulled back over the glans without drying. The controls were permitted to leave, while those treated with orvus-mapharsen prophylaxis received the application as described under prophylaxis methods (q.v.) and were then permitted to leave with no further supervision.

Another group (Exp. 0102) received intracutaneous inoculation in addition to the local application method of above. These were injected with 0.1 cc of the material into the anterior surface of the forearm.

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syphilis in most of the patients. It began to appear that the reactivity shown only by the flocculation tests utilizing the crude lipoidal antigen was probably an expression of false seropositivity and that the procedures utilizing the cardiolipin antigen and the complement fixation procedures probably gave a more nearly accurate indication of the syphilitic status of the individual. (9,14,15)

In view of the importance of gaining information as rapidly as possible on this matter so as to be able to pick subjects for future experiments it was necessary quickly to gain some answers to this question. We had reason to believe from earlier experience (17) that the rabbit-passed *T. pallidum* would prove virulent following intracutaneous inoculation, and so decided to begin the study of the possible significance of the discrepant serologic findings without waiting to determine the outcome of experiments of 5-13-47.

Two groups of patients were selected. The first group was completely seronegative by the VDRL cardiolipin and Kolmer tests but reactive by others and had no previous history of syphilis or antisyphilitic treatment. These comprise experiment 0201. The second group consisted of patients with positive VDRL cardiolipin Kolmer, Kahn and Mazzini tests and are in experiment 0202.

Three of the second group had a history of inadequate antisyphilitic treatment from 1 to 3 years earlier, 4 had either a history of a penile lesion or a scar thereof, and 3 had no history or scar of penile ulcers.

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Results: In some of the patients transient papules lasting from a few hours to a few days were noted. The immediate reaction to injection of the material was the formation of a pallid papule which later assumed the form of a wheal surrounded by an erythematous area. This reaction which appeared to be in the nature of a sensitivity reaction or an allergic reaction faded rather rapidly within 12 to 24 hours after injection and was followed in only a few cases by the persistence of a small papule. In no patient was the papule thus seen of more than 1 weeks duration. None of the individuals showed any serologic reactions subsequent to injection.

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If the Kahn and Mazzini tests, alone positive in the face of negative VDRL slide and Kolmer tests for syphilis, signified that the patient was syphilitic then this group of untreated patients should not react to the injection of virulent material. If, though, the serologic pattern mentioned in the former statement indicated false positivity and if, as was our hypothesis, positive VDRL slide and Kolmer tests gave firm evidence of the presence of syphilis under conditions tending to cause false seropositivity and thought to exist in the population under study, then those patients with positive VDRL and Kolmer tests in addition to positive Kahn and Mazzini would not show evidence of infection following inoculation, while those with only positive Kahn and/or Mazzini, supposedly false positive, and with negative VDRL slide and Kolmer tests would show evidence of infection.

Both groups were also to be given an injection of the heat-inactivated material in order to further study the possible non-specific effects of the heat-killed *T. pallidum* in syphilitic testicular tissue.

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Experiment 5
Date - August 10, 1947
Place - Asylum

By this time it was realized that no infections had resulted from the technic of local application alone in experiment 1. It was thought that this failure to infect might be attributed to either or both of two causes: (1) The organism taken from an infection of 32 days duration in the rabbit might have lost ability to penetrate the mucous membrane as a result of age of infection in the animal or, (2) The animal passage material might have become so attenuated or altered that it had lost the ability to penetrate the human mucous membrane.

To determine whether virulence was lost due to the length of infection in the rabbit donor, a rabbit with an infection of 14 days was used. The local application technic was again used in experiments 0501 and 0502 with all patients receiving a total of 3.69×10^6 organisms and the latter group receiving prophylactic protection of orvus-mapharsen.

In an attempt to increase the virulence and to recover human passage material for subsequent attempts to infect through the mucous membrane it was decided to pass the material through man (experiment 0503). Patients with long foreskins were selected and received inoculation by injection of .1 cc of the emulsion containing 1.23×10^6 organisms into the distal border of the foreskin in addition to intracutaneous inoculation of a similar amount of inoculum into the anterior aspect of the right forearm. This was done with the expectation that the lesions would be excised and used for preparation of inoculating emulsions.

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Certain studies in natural and artificial transmission of gonorrhoea were being concurrently carried on by the authors as reported in another document (27). In these, volunteers were permitted to have intercourse with prostitutes, some infected with gonorrhoea and others uninfected. The genitalia of these men were inspected immediately following completion of contact as they left the bedroom and came into the anteroom where the physicians were carrying out the prophylactic procedures. It had been observed that an appreciable number of the men showed alteration in the state of the penile mucosa immediately after sexual exposure. The changes ranged from moderate reddening and engorgement of the penile mucosa, through mucosal abrasion with grossly visible bleeding points, to profuse bleeding from lacerations or tears in the frenum. In this group, it may be noted, circumcision was rare. It was our impression that the severity of mucosal change seemed to be proportional to the total time elapsed for one or more contacts during the course of an evening. Since contact was not under medical observation there is no way of knowing whether vigor of coitus could be related to extent of abrasion of mucosa.

It was our practice to take samples of cervical and urethral material to culture for gonococci both before and at the completion of contact with the experimental male group, thus affording opportunity for inspection of the condition of the vaginal and pudendal mucous membrane.

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In order to determine whether the Frew strain had sufficient infecting qualities a group of 4 patients were selected. Each patient in this experiment (0601) received .1 cc intracutaneously in the flexor aspect of the right forearm and .1 cc into the foreskin of the penis making a total of .2 cc or 2.46×10^6 organisms.

At the same time another group of patients were to receive a 2-hour exposure of the same emulsion to the intact mucous membrane (Experiment 0602). Each patient was given six local applications of virulent emulsion for a total of 4.61×10^6 organisms.

To test the significance of finding of abrasion following normal coitus it was decided to scarify lightly the mucous membrane of the glans penis and to allow a 2-hour exposure of the abraded area to the inoculum. Thus each patient in experiment 0603 received a total of 4.61×10^6 organisms.

It was realized at the outset that the mechanical abrasion would probably be more severe than that occurring naturally and might permit more ready penetration of the organisms. But it was felt that under such circumstances any agent to be tested for prophylactic value would be subjected to a more severe test condition than that occurring naturally.

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In experiments 0605, 0606, and 0607, all patients were inoculated by this method and all receiving 2.7×10^5 organisms in the drop of .0122 ccs. that was placed on the arm. Those in experiments 0605 and 0606 remained under observation for $1\frac{1}{2}$ hours during which time the drop dried with those in the latter experiment receiving orvus-mapharsen prophylaxis for 2 minutes immediately after exposure and the 0605 group remaining as a control group. Those patients in 0607 had the virulent emulsion in contact for two hours and fifteen minutes after which time they were also given the orvus-mapharsen prophylaxis.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	0601	4	3	0
	0602	7	1	2
	0603	6	5	1
	0604	6	2	0
	0605	6	5	0
	0606	7	0	0
	0607	7	0	0

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Exp. 0701.

Seven male patients with long foreskins were selected and exposed to this human chancre material. The foreskin was retracted and the penis gently washed with a normal saline solution. This was followed by 6 applications of virulent emulsion over a 2-hour period. Total number of organisms was 3.36×10^6 .

Exp. 0702.

Earlier experiments had indicated that a high degree of success might be expected to follow application of the infected material to an abrasion of the mucous membrane of the penis. Following the customary technic of scarification and that of local application as described in the previous experiment each patient received 3.36×10^6 organisms over a 2-hour period.

Exp. 0703.

It was desired to prepare possible potential donors of chancre material for later use so that 6 individuals received inoculation of 6.85×10^5 organisms into the dorsum of the penis at the coronal sulcus and a similar amount into the flexor aspect of the right forearm.

Exp. 0704 and 0705.

In these two experiments it was desired to study the results of intravenous inoculation of the material. Patients in experiment 0704 received inoculation of 1.31×10^6 organisms into the right arm. The one patient in 0705 was given 6.85×10^5 organisms intravenously in the flexor aspect of the right forearm and the same amount intracutaneously in the dorsum of the penis at the coronal sulcus for a total of 1.37×10^6 organisms.

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Experiment 8
Date - Sept. 12, 1947
Place - Penitentiary

Field studies within the United States at this time were being carried out to determine the possibility of administration of abortive penicillin therapy to those exposed to infection as reported by Alexander et al (29) and Plotke et al (30). In the following experiments it was felt desirable to attempt such abortive therapy under rigidly controlled experimental conditions.

Exp's. 0801, 0802, 0803, 0804.

All patients in these experiments were inoculated intracutaneously in each forearm receiving an inoculum total of 2.24×10^6 organisms. The first group received an injection of 300,000 units of Wyeth penicillin in oil and beeswax 24 hours after intracutaneous inoculation. The second group received an injection of 600,000 units of the same Wyeth preparation at 48 hours after inoculation. Further to investigate the significance of the positive and negative VDRL and Kolmer tests, as described under experiments 0401-0404 two more groups with negative (0803) and positive (0804) tests were inoculated.

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Experiment 9
Date - September 21, 1947
Place - Asylum

As yet there was doubt as to the advisability of utilizing a method of inoculation involving damage to mucous membrane in testing prophylaxis, and attempts were continued to establish infection through intact mucosa. Knowledge of methods of apparently increasing virulence of certain organisms by suspension in mucin had been available (31) and had in our experience been tried in experimental gonococcal infection (16). In view of the great increase in mucin production in the female genital tract during intercourse it was felt that mucin might play a role in enhancement of invasive potentialities of *T. pallidum* and that it would be well to study possible effects of such a mixture on intact mucous membrane.

It was becoming evident, though, that the method of scarification-local application was probably going to prove a highly effective means of setting up infection, so that a group of patients was subjected to prophylactic study following this type of inoculation.

It was also decided at this time to determine the effectiveness of the blood-spinal-fluid barrier in prevention of passage of *T. pallidum* between the systems.

The source of spirochetes for experiments 0901 thru 0907 was from a human chancre experimentally produced in experiment 0503. The number of spirochetes in 1 cc. of emulsion was 4.9×10^6 .

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Exp. 0904.

The one patient in this experiment was originally scheduled to be in experiment 0903. However, after scarification and the first application of emulsion the patient fled the room and was not found until 2 hours later with the pledget still in place. No further emulsion was added to the pledget and the patient received only .2 cc or 9.8×10^5 organisms as compared to .3 cc or 1.47×10^6 organisms received by the patients in experiment 0903.

Exp. 0905.

It was desired to test the orvus-mapharsen prophylaxis against the human material applied to the scarified penis. Inoculation and size of inoculum are the same as described in 0903. At the end of a two hour exposure period a solution of the orvus-mapharsen prophylaxis containing .15% mapharsen and 1% orvus was prepared with approximately 60 ccs. of the solution being used for each patient. Prophylaxis was applied by the physician. Following the routine procedure described earlier the time of application was approximately 2 minutes.

Exp. 0906.

Using the human chancre material a group of 4 patients was inoculated with 8.5×10^4 organisms by the multiple pressure technic in each of two places, the upper arm and the penis just proximal to the coronal sulcus so that the inoculation took place into the inner surface of the foreskin. This procedure was practiced inasmuch as it was desired to prepare a few patients as possible donors for subsequent inoculations and it was desired to secure good penile and cutaneous chancres if at all possible.

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Exp. 0911.

As the control group for the spirochetal emulsion prepared from the rabbit testicular material six women were injected intracutaneously in the flexor aspect of the right forearm. Each individual received .1 cc of the suspension, or 7.7×10^5 organisms.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	0901	7	6	1
	0902	4	4	0
	0903	6	6	0
	0904	1	1	0
	0905	6	0	0
	0906	4	3	1
	0907	3	3	0
	0908	7	1	0
	0909	7	1	0
	0910	4	4	0
	0911	6	6	0

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In experiment 0704 the intravenous inoculation had been performed by removing the needle immediately after injection of the emulsion and by injecting through the same needle into the next patient, etc. It was noted that patients so inoculated developed darkfield positive papules at the site of injection so that it was not yet certain whether or not asymptomatic infection, or even infection, could be established by placement of the inoculum directly into the blood stream without contamination of the tissues overlying the blood stream. To permit inoculation into the blood stream without contamination of the overlying tissue the procedure described under technics was followed.

The Nichols-strain animal had been inoculated intratesticularly on the same day as the Frew-strain animal used, between 3 and 4 weeks before removal of the testes for inoculation.

Exp. 1001. Reinfection in the Male.

Each individual was injected with .15 ccs. intracutaneously at each of two sites in the upper arm making a total of .3 ccs of inoculum containing 4.92×10^6 organisms of Frew rabbit-passage material.

Exp. 1002. Frew Strain Rabbit Passage.

Each patient was inoculated intracutaneously in the flexor aspect of the right forearm with .022 ccs. containing 2.42×10^5 organisms.

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Exp. 1007, 1008, 1009.

In order to prepare possible donors for a future date 6 males were inoculated by the following technics:

.2 ccs. of the material or 4.94×10^5 organisms prepared from chancres of the Frew strain removed from humans was inoculated submucosally into the foreskin of the penis. The site of inoculation varied slightly in the three experiments. At the same site a drop of the material was placed on the foreskin which was lightly scarified through the drop.

Exp. 1010.

In a previous experiment to test intravenous inoculation it was noted that lesions developed at the site of the needle puncture. It was felt that this might have resulted from contamination of the skin overlying the vein so that the experiment was repeated in the following manner. The vein was entered and a quantity of the patient's blood was removed. The syringe was withdrawn with the needle in place in the vein; 0.2 cc. of the suspension was injected or 4.94×10^5 spirochetes. The patient's blood was then replaced thru the needle, washing in the spirochetes.

Experiment 11
Date - Jan. 11, 1948
Place - Asylum

Repetition of previous experiments to test the value of the orvus-mapharsen prophylaxis following the technic of scarification-local application for 2 hours was called for in order to increase the size of the sample subjected to this procedure.

Previous experiments had begun to indicate that reinfection could be attained following adequate penicillin therapy for primary and secondary syphilis. It was thought that local tissue-immunity to reinfection might not be found. If not found that fact would prove the inapplicability of the criterion formerly demanded clinically to substantiate the diagnosis of reinfection--i.e. the occurrence of a chancre at a new site. In order to test this, a group of patients having a well-defined scar of the previous experimental infection and having had adequate penicillin therapy was picked and subjected to reinoculation. Since the group was of necessity selected from a number of previous experiments, it was not practicable to attempt reinoculation with either the identical strain or dose of organisms originally utilized. Therefore, in the following experiments the strain used will necessarily vary from one experiment to another.

Exp. 1101 and 1102.

All of the patients used in these experiments had had previous successful inoculation and treatment. Five patients were inoculated intracutaneously into the scar of the previous infection with 1.31×10^5 (Exp. 1101) and five with 1.64×10^5 spirochetes (Exp. 1102). The volume of inoculum given to each patient was .1 cc.

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Experiment 12
Date - January 31, 1948
Place - Asylum

In this experiment it was desired to test a number of different factors. First it was necessary to compare the orvus-mapharsen prophylaxis with the standard Army pro-kit consisting of 30% calomel ointment against infection by the method of scarification and local application. Along with this it was desired to carry out further studies of orvus-mapharsen prophylaxis in females inoculated by the methods of multiple pressure vaccination. And finally, it was desired to carry out studies of reinfection in two groups of patients, the first of which had been fully treated with penicillin yet still had a high serologic titre, the second of which had been inadequately treated for an earlier infection by means of 150,000 units of aqueous penicillin solution.

In view of the fact that we were still not sure of whether there was any significant difference in the response to human and rabbit strains of *T. pallidum* and because the ultimate value of a prophylactic agent depended upon the ability to protect man against the infection in man, it was felt desirable to use human passage material for inoculation at this time.

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The inoculation was made by the usual technic of scarification and local application. Exposure took place for two hours with applications being made at one half hour intervals as in the control. The total suspension used .3 ccs containing 1.56×10^6 organisms. At the end of the two hour exposure the pledgets were removed and application of the prophylaxis was made by the physician. Alternate patients, as they stood in the row for inoculation, were given applications of either the Parke Davis orvus-mapharsen preparation or the "Pro Kit." The application was made by one of the physicians. For the orvus-mapharsen prophylaxis (Exp. 1205) the application was made in the usual fashion with the preparation being poured over the physician's hand as needed. Approximately 30 ccs. of the preparation was used for each patient and the application to each patient took two minutes. For a test of the Army Pro kit, (Exp. 1206), one tube of ointment was used for each three men. The time of the application for each man was 2 minutes and the application was given by the doctor in accordance with instructions on the tube. A small amount of the material was placed in the meatus and the rest of the material was put onto the glans and foreskin and was thoroughly rubbed into the glans, foreskin, shaft of the penis and onto the pubic hair by the physician giving the treatment. At the completion of prophylaxis by either technic the patient was allowed to leave the room without washing or without drying.

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Exp. 1210. Control Inoculations by Multiple Pressure.

The standard technic for multiple pressure was used. .022 ccs. of material containing 1.15×10^4 organisms was dropped onto the skin of the upper part of the deltoid region of the arm and multiple pressure vaccination was performed.

Exp. 1211.

In this experiment it was desired to determine the local effect of the orvus mapharsen prophylaxis, using the other arm as control, and to find out whether or not there were any systemic effects from the application. Each individual was vaccinated as described in the preceeding section, only vaccination was performed upon both the right and left upper arm by multiple pressure technic. Eighty minutes after vaccination the orvus mapharsen prophylaxis was applied by the physician to the inoculation site of the left arm only.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	1201	1	0	0
	1202	1	1	0
	1203	1	0	1
	1204	10	5	0
	1205	12	1	0
	1206	13	1	1
	1207	9	0	0
	1208	2	2	0
	1209	11	10	1
	1210	8	8	0
	1211	10	10	0

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Exp. 1305 and 1306.

Further studies in prophylaxis using penicillin in peanut oil and beeswax were carried out giving 600,000 units of POB preparation 12 hours after inoculation in experiment 1305 and 1.2 million units of the POB preparation at 24 hours after inoculation in experiment 1306. The patients thus treated prophylactically were inoculated in the flexor aspect of the left forearm.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	1301	6	6	0
	1302	7	7	0
	1303	7	5	0
	1304	8	3	0
	1305	6	0	0
	1306	6	0	1

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In addition to the two groups of patients considered latent another group with negative VDRL and Kolmer tests was selected as controls for experiment 1401.

In order to have a control emulsion, 3 ccs. of the supernate was removed following centrifugation and was inactivated for 1 hour in a water bath at a temperature of 62° centigrade for 50 minutes and 56° centigrade for 10 minutes.

All of the experimental patients received an intracutaneous injection of .1 cc of this inactivated material into the flexor aspect of the right forearm in addition to .1 cc of active material containing 7.95×10^5 organisms into the flexor aspect of the left forearm.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	1401	8	7	0
	1402	7	5	0
	1403	11	5	2

Experiment 15
Date - March 19, 1948
Place - Asylum

Further studies were indicated at this time both in immunity or susceptibility to reinfection and in prophylaxis using both penicillin and oral bismuth which has been found effective in animal studies.

Exp. 1501.

A control group of patients was inoculated by injection of .1 cc of the Nichols suspension containing 7.95×10^5 organisms into the mucous membrane of the glans penis just distal to the coronal sulcus.

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Exp. 1505.

It was desired to determine the effect of oral penicillin as prophylaxis following local scarification and local application. Inoculation was performed by the technic described in experiment 1503 with inoculum similar to experiment 0504. At the end of exposure, each patient was given 15 tablets of oral penicillin containing 100,000 units each per tablet, a total of $1\frac{1}{2}$ million units of buffered oral penicillin G.

Exp. 1506.

This experiment was performed to test the effect of sobismonol given 1 hour after final application by the technic of 1505. At 1 hour after final application 4 pulvules of sobismonol mass was given a total of 0.75 grams containing 150 mg. of bismuth.

Exp. 1507.

It was desired to determine the effect of aqueous penicillin applied locally as a prophylaxis. The technic for inoculation was that described above in 1504, scarification and local application for 1 hour. At the end of that time the pledget was removed and the physician washed the penis of each patient with 15 ccs of a penicillin solution containing a total of 500,000 units of solution of sodium salt of crystalline penicillin G. The time of the application was 2 minutes and the technic of application was that used for the orvus-mapharsen prophylaxis.

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Exp. 1603.

It was desired to study the further effect of local application of aqueous penicillin solution as prophylaxis. Following the technic of Exp. 1601 each patient was administered prophylaxis 6 hours after scarification by the physician following the technic used for application of the orvus-mapharsen prophylaxis. Each patient was washed with 15 ccs. of a solution of aqueous penicillin G. sodium salt containing 500,000 units in distilled water.

Exp. 1604.

The patient was inoculated by the method of scarification and local application. But during the procedure a urethral discharge was noted as the 2-hour exposure progressed. Smear disclosed the presence of gram negative intra cellular diplococci. It was then found that the patient was an active homosexual and that it would be undesirable to permit development of a penile chancre. At the end of the 2-hour exposure the patient was given orvus-mapharsen prophylaxis. As he represented a control patient from group 1605 inoculation was then performed by multiple pressure technic on the left arm.

No penile chancre developed, but a lesion did develop at the site of vaccination.

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A group of patients were inoculated by the technic of multiple pressure on the outer aspect of the left upper arm with .022 ccs. of suspension containing approximately 1.59×10^5 organisms. Two hours after the vaccination each patient in the group was given 3 tablets of buffered crystalline penicillin G potassium containing 100,000 units per tablet or a total of 300,000 units of oral penicillin.

Exp. 1609.

If an individual who had been working with clinical or experimental syphilis pricked himself with a needle or otherwise contaminated himself in some manner the recommended practice was that he be given immediately an injection of 40 or 60 mg. of mapharsen or an equivalent amount of arsphenamine or nearsphenamine. It was desired to test the validity of that practice. A group of patients was given intracutaneous inoculation into the left forearm. Each patient received .1 cc of the emulsion containing 7.95×10^5 organism. Two hours after inoculation each patient was given an intravenous injection of 40 mg. of mapharsen.

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<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	1601	13	3	4
	1602	8	7	1
	1603	6	0	0
	1604	1	1 (arm only)	0
	1605	7	6	0
	1606	1	1	0
	1607	9	9	0
	1608	13	0	0
	1609	7	1	0
	*1610	8	1	0
	1611	1	1	0

*All patients in this experiment that were exposed became infected. However, all but 1 recovered after treatment of 300,000 units of crystalline penicillin G potassium in oil and beeswax 11 days after inoculation.

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Exp. 1704.

A control group of patients for intracutaneous inoculation was used, each patient receiving by intracutaneous technic into the flexor surface of the left forearm .1 cc of the Nichols emulsion containing 6.71×10^5 organisms.

Exp. 1705.

This group who had previously been protected by prophylaxis was inoculated by the technic of scarification and local application for 2 hours. The technic used was that described earlier. Each patient received a total of .3 ccs. of the Nichols emulsion containing 2.01×10^6 organisms.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	1701	1	1	0
	1702	12	9	1
	1703	2	0	1
	1704	2	2	0
	1705	31	29	1

PART III

Chronology

Table III - Summarizing - All experiments should go here

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In view of the French experience that superinfection resulting in chancre formation following auto inoculation could be observed almost invariably during the first few weeks of infection, it was our thought that simultaneous inoculation of two sites should result in two chancres if infection were to take place at both. Thus at this stage of our experiments one patient could be expected to answer several questions if exposed both to parenteral as well as local application of the virulent material.

<u>Results:</u>	<u>Number of Patients</u>			
	<u>Experiment</u>	<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	0101	7	0	1
	0102	7	6	1
	0103	7	0	0

Experiment 2
Date - May 14, 1947
Place - Penitentiary

At this stage of our work, in epidemiologic surveys of various population groups, we had already been impressed by the high rate of occurrence of serologic activity as shown by the Kahn standard test and the Mazzini test, both with crude lipoidal antigens, as compared to the low rate of serologic activity as shown by the VDRL slide test and the Kolmer simplified test. It had been noted that many samples of blood tested showed serologic discrepancy with the former two procedures reactive in the face of negativity as shown by the latter two tests. It had been impossible for us to correlate the serologic findings with a history of, or signs and symptoms of

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The procedure used was that of intracutaneous inoculation with each patient receiving .2 cc of emulsion for a total of 1×10^5 organisms.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	0201	7	7	0
	0202	10	3	0

Of the 3 patients becoming infected in experiment 0202, 1 patient had received inadequate treatment while the remaining 2 had received no treatment and one had no history of a penile lesion and no scar.

Experiment 3

Date - June 28, 1947

Place - Penitentiary

In order to rule out clinical and serologic changes which might result from the injection of rabbit testicular tissue alone it was decided to take a group of individuals approximately half normal (Exp. 0301) and the other half with latent syphilis (Exp. 0302) to receive an injection of normal rabbit testicular material prepared as though for inoculation with the testicular syphiloma. The subcutaneous injection of .2 ccs of the testicular-spirochetal-broth mixture was performed and the patients were followed by clinical and serologic examinations.

Chronology 7

Experiment 4
Date - August 7, 1947
Place - Penitentiary

By this time it had become evident that intracutaneous inoculation of rabbit testicular syphilomatous material would probably result in a high degree of infection. At this time, though, we still did not understand the relationship between the high degree of serologic reactivity that we had found in the penitentiary, and the presence or absence of syphilis; although we felt that probably the more nearly true indicator of syphilis in the absence of clinical evidence was either the cardiolipin procedure or the complement fixation procedure. It seemed that the Mazzini and Kahn reactions had a high degree of tendency to false positivity. It was our opinion, on the basis of animal experience and on the basis of our own interpretation of the literature, that superinfection, i.e. establishment of a new infection by either the same or different strain in a patient having untreated syphilis with *T. pallidum* still present in the body, was not possible. Thus development of clinical or serologic evidence of infection following injection of virulent material would indicate the absence of infection, latent or active, at the time of inoculation. We decided to test the significance of the positive serologic tests for syphilis observed against this hypothesis.

Chronology 9

We were also concerned with the possible non-specific effect of the testicular syphilomatous tissue so that it was felt necessary to inject the heat-inactivated testicular syphilomatous material alone into groups of patients of both categories defined above to determine what the clinical and serologic reaction would be in both non-syphilitic individuals and those considered syphilitic on the basis of a positive VDRL slide test. The material for inoculation was prepared in the same manner as described in the section on technics. Both heat-killed material and virulent emulsion were given intracutaneously in the flexor aspect of the right and/or left forearms.

As can be seen from the Experiment Summary Table, twenty three inmates were divided into groups as to the presence or absence of a positive VDRL cardiolipin test for syphilis. Those in experiments 0401 (cardiolipin negative patients) and 0402 (cardiolipin positive patients) received 1.75×10^6 virulent organisms in one site and an equivalent dosage of heat-inactivated organisms in the other. Two other groups of VDRL cardiolipin positive and negative patients, numbers 0403 and 0404, received only the inactivated material.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	0401	3	3	0
	0402	8	4	0
	0403	(No serologic response was noted in these groups receiving inactive material. Only slight, transient clinical response noted.)		
	0404			

Chronology 11

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	0501	7	0	1
	0502	7	0	1
	0503	8	7	0

Experiment 6
 Date - August 24, 1947
 Place - Asylum

By this time it had become even more clearly evident that an application of rabbit-passage *T. pallidum* of Nichols strain to the intact mucous membrane of the penis for one hour could not produce infection; while the same material introduced submucosally was fully capable of producing syphilis which clinically and serologically was identical with that observed following sexual contact with human sources. In other words after injection into the penile mucosa, the Nichols strain originally isolated in 1912 from the spinal fluid of a patient with a neurorelapse following inadequate therapy produced, after 35 years of rabbit passage, (26) a clinical and serologic syndrome no different from that of the "street strains" of *T. pallidum* which is seen in clinical practice. It was not certain, though, whether the Nichols strain material differed from human "street strains" in the ability to penetrate the unbroken human mucous membrane. But at this stage sufficient human chancre material to test this factor further was not at hand.

Chronology 13

In contrast to the evidence of physical trauma to the penile mucosa it was noted that after contact with as many as 20 men in the course of 3 to 4 hours the genital mucosa of the prostitutes showed no physical evidence of trauma or even of hyperemia.

These findings suggested that in the course of coitus, infection with syphilis might be dependent, to some extent, upon the existence of breaks in the continuity of the mucous membrane, even though they were microscopic. It thus seemed that any method of inoculation which destroyed the continuity of the skin or mucous membrane might offer a more nearly physiological approach to the problem of bringing about experimental infection.

Two further possibilities suggested by these findings existed to explain the failure to obtain infection by application to the intact mucous membrane:

1. That the time of exposure to the infecting emulsion was not sufficiently long. This could be tested by increasing the exposure period to 2 hours.

2. That the Nichols strain had lost the power to penetrate the intact human mucous membrane, even though retaining that ability for the rabbit (28). This could be tested by utilizing a strain recently isolated in the rabbit from the human and having undergone few transfers.

Thus for this experiment the Frew strain was used, taken from an animal representing the 3rd rabbit passage, of 24 days duration when harvested.

Chronology 15

One of the questions still to be answered was the ability of the spirochete to penetrate the intact mucous membrane of the gastrointestinal system. It was felt that knowledge of this factor would assist in clarifying the problem of oral contagion through kissing and oro-genital sexual contacts. Each patient selected for this experiment (0604) was given to swallow under observations 20 ccs of distilled water containing 1cc of testicular material having 1.04×10^7 organisms per cc. The patients were not allowed to eat during an hour subsequent to swallowing the material.

With limitations on the number of males available for use it was evident that it would be desirable to utilize females both in prophylactic studies, as controls, and in immunity studies. With the knowledge that the so-called multiple pressure method of smallpox vaccination depends upon deposition of the virus within the epidermis where it might be expected to be susceptible to action of locally applied medication it was decided to use this technic.

Chronology 17

Experiment 7
Date - August 31, 1947
Place - Asylum

Complete results of any given experimental procedure were not available for at least 4 months following inoculation. With limited time available for completion of the project it was thus not feasible to delay four months between each experimental run so as to plan successive experiments on the basis of knowledge gained from the predecessors. Thus it was necessary to anticipate results upon bases of early observations and to move ahead on the strength of incomplete experimental data with knowledge that final analysis would be made of the completed work so that any errors in the early hypotheses would be shown up.

Mention was made in experiment 6 of the possibility that human "street strain" of *T. pallidum* or human-passed Nichols strain might have greater ability to penetrate intact genital mucous membrane than the rabbit strain. In order to test this hypothesis and secure information necessary for further prophylactic studies it was determined to repeat some of the earlier technics utilizing human material taken from patients with chancres appearing in the venereal disease ward of one of the local hospitals. Since it was obvious that it would be impossible to secure enough material from one chancre to inoculate a large group it was decided to pool several chancres and to call the several strains thus mixed "street strain".

Chronology 19

Exp. 0706.

There was some question in our mind as to whether there would be any differences in clinical or serologic response, or incubation time of material taken from humans infected with "street strain" of *T. pallidum* as compared with the infections resulting from material secured from a rabbit strain particularly the Nichols strain. In order to determine this, 7 females were vaccinated by the technic of multiple pressure. For this, suspension No. 1 was used containing 6.85×10^5 organisms in .1 cc. .022 cc. of the material or 1.5×10^5 organisms remained in contact with the skin for 1 hour and 15 minutes before drying completely at which time the patients were dismissed from observation.

Exp. 0707.

Seven more patients were inoculated by the same technic and size of inoculum as in 0706 and were then given prophylactic treatment with orvus-napharsen solution. Approximately 15 ccs. of the prophylaxis was applied by the expedient of soaking a cotton pledget in the solution and rubbing the exposed area for 2 minutes.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	0701	7	4	0
	0702	7	7	0
	0703	6	6	0
	0704	6	6	0
	0705	1	1	0
	0706	7	7	0
	0707	7	5	0

Chronology 21

In this experiment it was also desired to carry out further the effects of intramuscular penicillin prophylaxis begun in the penitentiary on this same date.

Each patient was inoculated with .1 cc of the suspension in each forearm giving a total inoculum of 2.8×10^6 . The control group (Exp. 0805) received no further treatment. The second group (Exp. 0806) received an injection of 600,000 units of Wyeth penicillin in oil and beeswax 24 hours after inoculation. The third group (Exp. 0807) received 1.2 million units of the same preparation 48 hours after inoculation. The fourth group (Exp. 0808) receiving a total inoculum of 2.24×10^6 remained as a control group without further treatment.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	0801	9	6	2
	0802	7	2	0
	0803	2	2	0
	0804	5	1	2
	0805	5	4	1
	0806	6	4	0
	0807	7	3	2
	0808	3	3	0

Chronology 23

Exp. 0901.

To answer the latter problem seven patients, all of whom were deteriorated and debilitated epileptics, were given intracisternal inoculation of 4.9×10^5 organisms. It was hoped that by shock of inoculation it might be possible to influence favorably their epilepsy. This experiment was undertaken at the expressed desire of the clinical director in hopes that he might be able to do something for these women who had been completely resistant to all types of anticonvulsive therapy. All of these were so uncontrollable that they had inflicted serious injuries upon themselves such as burns leading to contractures, blindness, wounds, etc., as a result of the loss of consciousness and motor activity due to epileptic attacks.

Exp. 0902.

The patients used in this experiment were selected to serve as control patients for the human spirochetal emulsion. Each patient was inoculated intracutaneously with .1 cc. of emulsion or 4.9×10^5 organisms into the right forearm.

Exp. 0903.

There was also a question in our mind as to whether or not there might be a difference in the invasiveness of human-passage as compared with animal-passage organisms so that a group of patients was subjected to the technic of scarification and local application (q.v.). A total of 1.47×10^6 organisms were administered during 4 applications at intervals of 30 minutes each.

Chronology 25

Exp. 0907.

Three patients were used as control patients for the human spirochetal emulsion. Each patient was inoculated intracutaneously into the flexor aspect of the right forearm with 4.9×10^5 organisms.

Exp. 0908.

In this experiment and following experiments performed on this date the Frew strain rabbit testicular material was used. In this particular experiment it was desired to study the effect of a still longer exposure to the infected material when applied to the intact mucous membrane of the penis. Seven individuals with long foreskins were selected, without any preliminary cleaning of the penis and foreskin. Inoculation was practiced by the method of local application for 3 hours with each patient receiving 6 applications with a total of 2.85×10^6 organisms.

Exp. 0909.

To test the effect of mucin in promoting infection through the intact mucous membrane of the penis, the suspension of spirochetes was made up in a .15% fresh mucin solution. This preparation was applied by the technic of local application (q.v.)

Exp. 0910.

These men served as control for the virulence of the mucin solution being tested by local application in 0909. Injection of 3.85×10^5 organisms was made into the penile mucous membrane so as to secure a chancre which might be removed by circumcision and used for subsequent inoculations.

Chronology 27

Experiment 10
Date - November 30, 1947
Place - Asylum

It had by now become evident that the only practicable method of testing prophylactic substances for local application under conditions approximating normal sexual exposure was that called "scarification and local application". It was not yet certain, though, whether or not there was significant difference in serologic or clinical response to various strains of spirochetes, rabbit-carried strains or human-passed rabbit strains as contrasted to human or "street-virus" strains. To test these questions a group of patients was divided and inoculated with equal numbers of different strains of spirochetes administered in identical manner by intracutaneous inoculation in the forearm.

The infections which had already developed in previously inoculated patients had begun to be treated, and a group of patients having received penicillin therapy of 50,000 q 2 h x 85 injections, or 4,250,000 units, was available to challenge for immunity by reinoculation. To rule out nonspecific clinical manifestations such as local tissue reaction to foreign protein, possible allergic response to reexposure to rabbit tissue, etc., certain patients received control injections of identical volumes of the inoculum in which spirochetes had been killed by heating for two hours in a water bath at 56 degrees C. Inactivated material was given in one site and virulent material in the identical site on the other arm in the small group thus challenged.

Chronology 29

Exp. 1003 and 1004. Reinoculation Studies in the Female.

In this group it was desired to have a control of inactivated rabbit testicular material, so that a portion of the material was decanted and inactivated at 56° for 120 minutes. Those patients receiving this heat-killed material (Exp. 1004) were given .09 ccs. of the heat-inactivated Nichols strain material intracutaneously into the right upper arm in the same area as that receiving the virulent inoculum in the left arm. The supernate was not poured from the tissue after counting. Each individual was inoculated intracutaneously into the left upper arm. Some individuals received .2 ccs. others received .25 ccs. so that the total dosage was 2.2×10^6 organism or 2.75×10^6 organism depending upon the individual patient.

Exp. 1005 and 1006.

For these two experiments the rabbit Nichols strain used was taken from a rabbit with an infection of identical ages as the Frew strain infection. The material was prepared in the same manner as the emulsion of the Frew strain used in Exp. 1002. In experiment 1006 each patient was given an intracutaneous inoculation with .1 cc of virulent preparation containing 2.47×10^5 organisms. In experiment 1005 in addition to virulent material each patient received 2.47×10^5 organisms that had been heat killed by procedures described previously.

Chronology 31

Exp. 1011. Frew Strain Human Passage.

The patients were inoculated intracutaneously in the flexor aspect of the right forearm with .1 cc of this suspension, containing 2.47×10^5 organisms per 0.1 cc thus giving an opportunity to compare the human-passage Frew strain with the rabbit-Frew strain (1002) and the rabbit-Nichols strain (1005 and 1006) with respect to the clinical and serologic response to inoculation.

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	1001	10	10	0
	1002	8	8	0
	1003	7	7	0
	1004	8	8	0
	1005	6	6	0
	1006	1	1	0
	1007	4	4	0
	1008	1	1	0
	1009	1	1	0
	1010	6	6	0
	1011	7	7	0

Chronology 33

Exp. 1103.

Two patients were prepared as possible future donors by inoculation of .1 cc of the suspension containing 1.37×10^5 organisms into the mucous membrane of the foreskin just distal to the coronal sulcus.

Exp. 1104.

This group of patients was inoculated by the technic of scarification and local application. At the end of this two-hour period the patients were given a 2 minute prophylaxis application by the physician. The prophylaxis was 30 ccs. of a orvus-mapharsen preparation consisting of 1% orvus and 0.15% mapharsen in distilled water.

Exp. 1105.

Three patients were inoculated by scarification and local application with .15 ccs. in contact for 20 minutes. Then they were injected with .1 cc into mucous membrane of the penis. Total number of spirochetes was 2.80×10^6 . They were being prepared as possible future donors of strain material.

Exp. 1106.

This group of control patients for the prophylactic procedure was inoculated with the same material and technic as written under experiment 1104.

Results:	Experiment	Number of Patients		
		Exposed	Infected	No Data
	1101	5	5	0
	1102	5	5	0
	1103	2	2	0
	1104	12	0	0
	1105	3	3	0
	1106	10	10	0

Chronology 35

Exp. 1201, 1202, 1203.

In these experiments in reinfection three patients were used. The two patients in 1201 and 1202 had been treated for experimentally produced syphilis and had a high titre at the time of reinoculation. The other patient in experiment 1203 had latent syphilis from a naturally acquired infection. All patients were given an injection of .1 cc of the antigen emulsion consisting of 5.2×10^5 organisms. Patients in 1202 and 1203 were injected into the mucous membrane of the foreskin with the former patient having scarification 30 minutes prior. Total number of spirochetes in this case was 7.80×10^5 . In experiment 1201 the patient was inoculated by injection into the flexor aspect of the right forearm.

Exp. 1204.

The patients in this experiment were used as control patients for the technic of scarification and local application. The application was made at one half hour intervals. A total of .3 ccs. of the suspension was made containing 1.56×10^6 organisms over the two hour period.

Exp. 1205 and 1206.

In experiment 1205 the orvus-mapharsen aqueous prophylaxis of 1% orvus and .15% mapharsen was used. In experiment 1206 the Army pro kit, Lot No. V-48, Item No. 9118000 of the Comfort Manufacturing Company, Chicago, Illinois, containing 30% calomel was used.

Exp. 1207.

It was desired to determine whether or not superinfection could take place. Eighteen days before inoculation a group of 9 patients with primary syphilis resulting from a previous inoculation were given treatment consisting of aqueous penicillin 25,000 units every two hours for 6 injections, a total of 150,000 units. Each of these patients was given an intracutaneous injection consisting of .1 cc of the inoculum in the right and left upper arm. A heat killed control inoculum had been prepared in the usual way as described under Technic and was injected intracutaneously into the left forearm. A total of 1.04×10^6 living organisms was injected. Each of the patients had had only one infection before attempted reinoculation and each patient was seropositive at the time of reinoculation. (There was no reaction of any kind at the site of the control inoculation of the heat killed material, either immediate or delayed).

Exp. 1208 and 1209. Reinfection of Patients with High Titre.

All of the patients had previously received adequate treatment for experimental infection. For some patients the reinoculation represented the second exposure and in others the third exposure. All patients were serologically positive at the time of reinoculation and all had a relatively high titre. In both experiments all the patients were given an intracutaneous injection of .1 cc of the emulsion containing 5.21×10^5 organisms into the right upper arm. However, in experiment 1209 the patients were given an intracutaneous injection of .1 cc of the heat killed material described above into the left upper arm. (As before (Exp. 1207) none of the patients receiving the heat killed control showed evidence of any type of reaction following inoculation.)

Chronology 39

Experiment 13
Date - February 8, 1948
Place - Asylum

In this group of experiments questions to be explored were the effect of varying size of inoculum and the use of penicillin given parenterally as prophylaxis against infection.

Exp. 1301.

Six patients were inoculated with .1 cc of the suspension intracutaneously into the flexor aspect of the right forearm for controls.

Exp. 1302, 1303, and 1304.

In this experiment it was desired to do studies on the effect of varying the size of the inoculum on rates of infection and also the speed of development of clinical and serologic evidence of the disease. Dilutions of the inoculum used for the controls were made containing 50,000, 5,000 and 10 spirochetes per .1 cc. The diluent used was the 50% mixture of rabbit serum and saline which was used in the preparation of the original emulsion. Patients in experiment 1302 received 50,000 organisms, 1303 received 5,000, and 1304 received 10. All were inoculated by intracutaneous injection into the flexor aspect of the left forearm.

Experiment 14
Place - Penitentiary
Date - February 14, 1948

There had been much question in our minds about resistance to reinfection following treatment of patients with early or late latent syphilis. In order to resolve that question more fully, the following experiment was set up. Eighteen patients all of whom had a positive Kolmer and VDRL test were selected. Some of the individuals gave a history of penile lesions. Others gave no history whatsoever of infection with gonorrhoea or syphilis. None of the individuals had had previous antisyphilitic treatment yet all of them had been found to be positive on more than one occasion by both of the tests mentioned. Penicillin therapy was instituted on seven of these patients which constitute experiment 1402. The complete course was given by two of the authors, S. L. & J. C. C. and the medical students who worked with them at the Asylum so that it could be certain that each patient received the full amount of therapy consisting of an injection of 50,000 units aqueous solution of penicillin G every 2 hours for 85 injections giving a total of 4.25 million units over a period of 7 days. Three days after termination of treatment the individuals were challenged by inoculation. These patients constitute experiment 1402 while the untreated group make up experiment 1403.

Chronology 43

Exp. 1502.

It was desired to determine whether or not reinfection would be regularly possible. A group of patients were selected all of whom had had previous experimental inoculations and who had serologic tests for syphilis ranging from complete negativity to rather strong sero-positivity at the time of reinoculation. Each one of these patients was reinoculated by the intracutaneous injection of .1 cc of the Nichols emulsion containing 7.95×10^5 organisms. Injection was intracutaneous into the flexor surface of the left forearm.

Exp. 1503.

It was desired to determine the value of sobismonol mass given 11 hours after final exposure. For this group of patients the method of inoculation consisted of scarification and local application to the mucous membrane of the penis for 1 hour's exposure.

Exp. 1504.

It was desired to test the effects of penicillin oil and beeswax (POB) as prophylaxis following technic of scarification and local application. The technic of application was exactly that reported in experiment 1503. However, the Frew strain was used and the number of spirochetes was 1.69×10^6 organisms. Eleven hours after a 1-hour application an intramuscular injection of 600,000 units of POB was given to each patient.

Chronology 45

<u>Results:</u>	<u>Experiment</u>	<u>Number of Patients</u>		
		<u>Exposed</u>	<u>Infected</u>	<u>No Data</u>
	1501	8	7	1
	1502	15	15	0
	1503	9	0	0
	1504	10	1	2
	1505	8	0	0
	1506	8	5	0
	1507	8	3	3

Experiment 16
 Date - May 9, 1948
 Place - Asylum

Various questions still remained to be answered, or required further study and are detailed below in the various sections of this experiment.

Exp. 1601.

It was desired to study the effect of administration of the orvus-mapharsen prophylaxis at a longer time interval after inoculation than in previous experiments. A group of patients was inoculated by the technic of scarification and local application for 2 hours with a total dosage of 3.18×10^6 spirochetes. Six hours after completion of the inoculating procedure each patient was treated by the standard technic of the orvus-mapharsen prophylaxis.

Exp. 1602.

For the control of experiment 1601 the technic was identical as that described above.

Chronology 47

Exp. 1605.

Using the technic of scarification and local application described earlier a group of patients was given $1\frac{1}{2}$ hours exposure to the organisms. The pledget was moistened every 20 minutes and a total of .25 ccs of the emulsion containing 1.98×10^6 organisms was used for each patient.

Exp. 1606.

This patient was inoculated by the technic of scarification followed by injection of 0.1 cc of emulsion into the dorsum of the penis in an attempt to produce a typical hunterian chancre for detailed photographic studies. A total of 1.98×10^6 organisms was used.

Exp. 1607.

In view of the fact that rabbit experiments indicated that there might be changes in immunity to the disease as time passed following successful treatment, a group of patients were reinoculated 1 year after adequate penicillin treatment. Each patient was given an intracutaneous injection of .1 cc of the emulsion containing 7.95×10^5 organisms into the flexor surface of the left forearm.

Exp. 1608.

Much speculation existed as to the possible value of oral penicillin as a prophylaxis against syphilis. In view of the evidence that oral penicillin in the dosage of approximately 300,000 units could be used very effectively as a prophylaxis against gonorrhoea, it was desired to study the effect of that dose on syphilis.

Chronology 49

Exp. 1610.

In studies of abortive therapy for syphilis up to this time, animal and human experimental work had indicated that there was an important relationship between the stage of incubation and the amount of penicillin required for abortive therapy. In previous experiments prophylaxis had been given up to 48 hours after exposure to the organisms. In view of the fact that patients might frequently be given penicillin for treatment of gonorrhea acquired during the incubation period of syphilis, so that the disease might be a week or more along in incubation at the time of treatment of gonorrhea, it was desired to test the effect of a small dose of penicillin such as that used for treatment of gonorrhea late in the incubation period of syphilis. For this purpose a group of patients was inoculated intracutaneously with .01 cc of a suspension containing 7.95×10^5 organisms. Eleven days after inoculation each of the patients was given 1 cc of crystalline penicillin G potassium in oil and beeswax, 300,000 units per patient.

Exp. 1611.

As was our practice whenever possible, each patient who had previously been protected by any sort of prophylactic procedure was at a later date reexposed to infection by the same technic without protection by prophylaxis in order to assure that the patient was susceptible to the infection. Thus this patient, previously protected by penicillin prophylaxis, was given intracutaneous inoculation of .01 cc of the suspension containing 7.95×10^5 organisms.

Chronology 51

Experiment 17
Date - July 4, 1948
Place - Asylum

This was the last group of experiments to be done. It was desired to inoculate individuals who had been protected by prophylaxis in previous experiments or who had failed to become infected following inoculation in previous experiments or who had failed to become infected following inoculation in previous experiments so that a number of different technics were used some involving only 1 patient previously protected prophylactically when exposed by the technic to which he was again subjected.

Exp. 1701.

One patient was inoculated by the technic of multiple pressure vaccination on the left upper arm, outer aspect through a drop of 0.022 ccs. of Frew strain emulsion containing 2.24×10^5 organisms.

Exp. 1702.

This group received intracutaneous inoculations of .1 cc of the Frew emulsion into the right forearm. Each patient received 1.02×10^6 organisms.

Exp. 1703.

Two patients with latent syphilis were inoculated. Each patient was given .3 ccs. of Frew emulsion by intracutaneous inoculation into the left forearm which made a total of 3.06×10^6 organisms per patient.