ATTEMPTS TO TRANSMIT RHEUMATIC FEVER TO RABBITS AND GUINEA PIGS.

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(Received for publication, June 16, 1924.)

INTRODUCTION.

The reticence of investigators to report their failures has probably retarded the solution of many problems. Much labor might be spared if a new worker in a subject had at his disposal a detailed record of the unsuccessful as well as the successful endeavors of his predecessors, a record so complete that he could avoid the repetition of details in technique that impress him as being possible causes of previous failures. It is such a record that we desire to report. In doing so, it is fully appreciated that unsuccessful transmission experiments never prove conclusively the non-existence of a virus in the inocula employed.

The search for the etiological agent of rheumatic fever has occupied the attention of many investigators. Of the several microorganisms ascribed to this rôle the *Streptococcus viridans* has taken the foremost position. But a number of unexplained objections have restrained the majority of workers from accepting this bacterium as the cause of the disease. Perhaps the most important objection is the failure of laboratory animals inoculated with the *Streptococcus viridans* to develop the characteristic pathological lesion of rheumatic myocarditis, the so called submiliary nodule, or Aschoff body.

The production of experimental rheumatic myocarditis by the intravenous inoculation of rabbits with *Streptococcus viridans* was first attempted by Bracht and Wächter (1). They described small foci of parenchymatous necrosis which became surrounded by lymphocytes, polymorphonuclear leucocytes, and fibroblasts; and which tended to heal with scar formation. These lesions, now quite generally known as the Bracht-Wächter bodies, did not resemble Aschoff bodies, but rather the focal embolic myocarditis of subacute bacterial endocarditis.

Thalhimer and Rothschild (2) and Cecil (3) found no Aschoff bodies in the hearts of rabbits inoculated with *Streptococcus viridans*, but only lesions similar to those of Bracht and Wächter.

Jackson (4), Coombs, Miller, and Kettle (5), and Rosenow and Coombs (6) all report the production in rabbits injected with *Streptococcus viridans* of myocardial lesions closely resembling Aschoff bodies. Their descriptions and illustrations, however, are not convincing. The chief points of dissimilarity are the necrotic changes, the presence of streptococci, often in considerable numbers, the tendency to radiating configuration, and the absence of the characteristic large, multinucleated type cell of the Aschoff body. The myocarditis described by Herry (7) in rabbits injected with "*endotoxine rhumatismale*" prepared from streptococci will be discussed later.

Besides these studies by the adherents of the streptococcal etiology of rheumatic fever there have been two investigations based on the hypothesis that the disease is caused by a filterable virus. De Vecchi (8), in 1912, reported the production of a myocarditis, which he regarded as very closely approximating that of rheumatic fever, by the inoculation into rabbits, dogs, rats, and mice of bacteria-free blood from patients with rheumatic fever. At the time of inoculation his animals were also injected with repeated small doses of a 1:5,000 solution of epinephrine. This latter procedure was intended to increase the susceptibility of the heart muscle to the action of the virus. Of the several species employed the rabbits showed the most marked myocardial lesions. Uninoculated control rabbits and those injected only with epinephrine sometimes showed lesions but not so extensive as those receiving serum from patients with rheumatic fever. No control experiments with epinephrine and normal human serum were made. Further reference to this work will be made in the discussion. More recently Natali (9), using only rabbits, has repeated de Vecchi's observations.

EXPERIMENTAL.

We have attempted to transmit rheumatic fever to rabbits and guinea pigs by injecting them with various materials obtained from patients suffering with this disease. The materials selected were whole blood, serum, joint fluid, pleural fluid, throat washings, and tonsil tissue. Thus were included those body fluids easily available, which might rationally be assumed to contain the virus of the disease during its acute stage. The frequent association of tonsillitis with rheumatic fever explains the use of throat washings and of tonsil tissue.

Practically all of these substances were obtained either before the patients had received antirheumatic medication, or after a sufficient time following the discontinuance of the treatment for its effect

to have disappeared. The clinical data on the patients are given in Table I. In each protocol the relation to such medication has been noted.

Methods.

The following inocula were used.

1. Blood.—The blood was withdrawn by venipuncture during the acute febrile state while the polyarthritis was still developing. In some instances serum was used, but in most the whole uncoagulated blood, usually drawn into a syringe which had been previously washed with a 10 per cent solution of sodium citrate. In one experiment a 3 day blood culture in broth was used.

2. Joint Fluid.—The fluid was aspirated from acutely inflamed joints, sometimes within a few hours after the onset of symptoms in the joint punctured. The earliest possible exudate was thus obtained. Anaerobic cultures of joint fluid made according to various modifications of the Smith-Noguchi method were also used for inoculation. Such cultures were always free from any demonstrable bacteria.

3. Pleural Fluid.—This was withdrawn during an acute pleurisy with effusion in a patient with rheumatic fever. Culture of this fluid showed no growth.

4. Throat Washings.—The patient's throat was rinsed with about 15 cc. of Ringer's solution. The washings were then filtered through a Berkefeld N filter, with suction from the house vacuum system. Cultures of the filtrates were always sterile.

5. Tonsillar Material.—The tonsils were removed from patients under gas-ether-chloroform anesthesia. Within 1 hour the tonsils were minced until reduced to a pulp which was suspended in sterile Ringer's solution and centrifuged at slow speed for a few minutes to remove the coarser particles. Some inoculations were made with the suspension in this state and some with the filtrate passed through a Berkefeld N candle. The filtrates were invariably sterile in ordinary media.

Transfer Inoculations.—The materials used for transfer inoculation were blood and a suspension of heart muscle and spleen. These suspensions were made by mincing the tissues, mixing the pulp in sterile Ringer's solution, and centrifuging a few minutes at slow speed to throw down the coarser particles. In one instance anaerobic Smith-Noguchi cultures (ascitic fluid-kidney tissue) of joint fluid of Rabbit H-63 (Series III) were used. The same general plan was followed for transfer injections as in the initial inoculation.

Animals.—The animals employed were young, healthy guinea pigs and rabbits. The guinea pigs were always inoculated intraperitoneally, the rabbits intravenously into the ear veins with blood and joint fluid, but intraperitoneally with all other inoculants. This was necessary because it was found that the intravenous injection of such materials as tonsil suspension invariably proved fatal within a few hours.

Subcutaneous inoculations were also made into the abdominal wall of both rabbits and guinea pigs in order that any local reaction might be more easily observed and also with the hope that subcutaneous rheumatic nodules might be produced. A few intraarticular inoculations were made into the knee joints of rabbits.

Clinical Observations on the Animals.—The animals were fed on a diet of hay, oats, bread, greens, and water. They were weighed daily. Their rectal temperatures were taken twice a day. It was early found necessary to sterilize the thermometers in lysol solution and alcohol to prevent the spread of any epidemic gastrointestinal disease.

The animals were examined daily for evidence of arthritis, subcutaneous nodules, diarrhea, snuffles, or any other signs of illness. X-rays were made of any suspicious joints in order to avoid mistaking a bone fracture for joint involvement.

Routine Autopsy Procedure.—The animals were killed by a blow on the occiput. The chest was opened asceptically and blood collected into a large capillary pipette from the heart or great vessels. This blood was cultured routinely in dextrose agar plates and deep tubes, and sometimes also in broth and used for the inoculation of other animals as indicated in the tables. If inspection of the viscera suggested the possibility of pneumonia, pleuritis, pericarditis, or peritonitis, film preparations and cultures were made. If heart and spleen were to be used for the transfer inoculations a piece of each was removed and placed in a sterile dish to be treated as described elsewhere. Pieces of the following organs were fixed for histological examination: heart, lung, spleen, liver, and kidney.

The skin of the animal was then removed and the skeletal musculature and joints dissected and carefully examined. Joints showing such abnormalities as increased fluid content, congestion, or swelling of the periarticular tissues were cultured and then fixed for future histological examination.

Histological Examination.—Tissues to be examined microscopically were fixed in either Zenker or Müller-formol, and a piece of the heart muscle was always fixed in absolute alcohol as well. Sections were cut in paraffin and stained with eosin-methylene blue or with hematoxylin and eosin (depending on the fixation). Sections of the heart were stained also with the Unna-Pappenheim methyl green-pyronine stain.

Controls.—Control animals, both rabbits and guinea pigs, were inoculated with normal human serum and whole blood, joint fluid from a case of arthritis deformans, control culture media, normal rabbit blood, and suspension of normal rabbit heart and spleen. These animals as well as uninoculated ones were kept under identical conditions and were autopsied and examined with the same care.

RESULTS.

Twenty-seven rabbits and fourteen guinea pigs were inoculated according to the methods described with body fluids and tissues of

patients suffering from rheumatic fever. Transfer inoculations, *i.e.* rabbit to rabbit or guinea pig to guinea pig, brought the total number of animals used in the experiments to 67 rabbits and 40 guinea pigs. These transfer inoculations were systematically carried out in a number of series in spite of the absence of symptoms of disease, because such negative evidence does not prove the non-existence of a virus in the body of the animal inoculated. It is well known that several passages may be necessary to adapt a virus to a new environment, and to render it parasitic and pathogenic for its new host species.

A definite arthritis occurred in only two animals, Rabbit H-63 (Series III) and Guinea Pig B-86 (Series xiii).

Rabbit H-63 had received the blood and suspension of heart muscle from a rabbit inoculated with whole blood from a patient with rheumatic fever. This rabbit had a mild fever, 104–104.5°, for a number of days. 26 days after inoculation the left ankle became swollen and hot. An x-ray picture showed the bony structures to be uninjured. On the 28th day the animal was sacrificed. The periarticular tissues were congested and edematous. Smears of this edema fluid showed mononuclear and polymorphonuclear leucocytes, but no bacteria. The joint cavity contained an increased amount of fluid, smears of which showed no cells and no bacteria. Cultures of the joint fluid and of the periarticular tissue juice in ordinary media and in Smith-Noguchi kidney-ascitic fluid showed no growth.

Efforts to repeat this occurrence by the inoculation into other rabbits of the blood of Rabbit H-63 and of the anaerobic cultures of the joint fluid were unsuccessful.

Guinea Pig B-86 (Series xiii) was injected intraperitoneally with 3 cc. of filtered throat washings from a patient with rheumatic fever during a relapse accompanied by severe pharyngitis. Cultures of the filtrate in blood agar showed no growth. The animal had fever (103°) from the 3rd to 7th days after inoculation and again for 2 days before death. It developed swelling of the right ankle on the 15th day without other evidence of disease and was sacrificed the following day. A complete autopsy revealed no pathological changes except swelling about the left ankle and an excess of fluid in the joint. No evidence of trauma was found. Anaerobic culture of the joint fluid in ascitic broth was sterile. Cultures of heart muscle and kidney were sterile. Two out of four cultures of heart's blood were contaminated by staphylococci; the others showed no growth. Transfer inoculations from this animal were negative.

Although these observations were unique in our entire series of experiments they are of considerable interest. Non-bacterial arth-

I.

Sources of Material and Occurrence of Myocardial Les

Patients furnishing material.				Material and	
Case No.	Age. Sex. Hospital. No.	No. of previous attacks of rheumatic fever.	Clinical features.	Material.[1]	y ase ied.
I	13 yrs. M. 4269	0 (4)[3]	Fever; slight arthritis disappearing spontaneously; moderately severe pancarditis. Mitral stenosis and regurgitation; subcutaneous rheumatic nodules. Six blood cultures negative. One attack of heart failure relieved by digitalis. At the end of 4 mos. he left the hospital against advice. Died of heart failure 7 wks. later. No autopsy.	Blood.	1
II	35 yrs. M. 4233	0	Fever; severe migratory polyarthritis; mitral endocarditis. Two blood cultures negative. Typical response to salicylate therapy. Recovery.	"	
III	38 yrs. F. 4255	2	Fever; migratory polyarthritis. Severe pancarditis and pleurisy. Four blood cultures negative. Mitral regurgitation. Salicylate medication unsatisfactory because of drug idiosyncrasy; spontaneous disappearance of arthritis. Left hospital against advice. Returned 2½ mos. later in advanced heart failure and died 11 days thereafter. No autopsy.	"	
IV	14 yrs. M. (Wm. E., out patient.)	2	Fever; mild polyarthritis, subcutaneous nodules. Chronic and acute myocarditis; aortic insufficiency, mitral insufficiency. Recovery. 3 yrs. later, a fourth attack in which he died. No autopsy.	Serum.[5] 3 day dextrose[6] culture of blo	

R. indicates rabbit; G. P., guinea pig; +, myocardial lesions found on microscopic e nation (see text); −, no myocardial lesions.

[1] Unless otherwise noted, cultures, of these materials were sterile.

[2] Animal transfers are numbered so that the original inoculation is called the first gener

[3] Attacks of chorea.

[4] Rabbit H-63. Acute non-bacterial arthritis (see text).

[5] Culture grew a *Streptococcus viridans*.

[6] No organisms demonstrable at this time; subsequently grew a *Streptococcus viridans*

I.

l Lesions in the Inoculated Animals.

imal inoculation.		Animals inoculated.					
y ease ned.	Previous salicylate medication.	Generation.[3]					
		1st	2nd	3rd	4th	5th	6th
d	None.	R. −					
	"	" +					
		G. P. −	G. P. +				
			" " −	G. P. +			
	"	R. +	R.[4] +	R. +	R. −		
				" −			
		G. P. −		" +			
				G. P. +			
		" " +	G. P. +	" " −			
			" " +	" " +			
		R. +	R. +	R. +			
	?	" −	" +		" −		
				" −	" −		
		" −	" −	" +	" −	R. −	R. −
				G. P. +	G. P. +	" −	" −
			" −	R. −	R. −		
					" +		
			" +	" −			
				" −	" +		

TABLE 1

Patients furnishing material.				Material used
Case No.	Age. Sex. Hospital. No.	No. of previous attacks of rheumatic fever.	Clinical features.	Material.[1]
				Serum.[7]
				" [7]
V	36 yrs. M. 4193	3	Fever; migratory polyarthritis. Prompt response to sodium salicylate. Recovery.	Blood.
				Joint fluid, left knee (within 24 hrs. after onset of symptoms).
VI	12 yrs. M. 4277	0 (3)[8]	Fever; migratory polyarthritis, promptly relieved by sodium salicylate medication. Mitral regurgitation. Two relapses with fever, precordial pain, and lengthening of P-R interval in electrocardiogram. Three blood cultures negative. Tonsillectomy at end of 2nd mo. Uneventful convalescence.	Blood. Suspension of tonsil (filtered).
VII	31 yrs. F. 4312	0	Fever; severe migratory polyarthritis; severe pancarditis; tardy response to full doses of sodium salicylate. Relapse with pleurisy, marked disturbance of cardiac rhythm—auricular fibrillation and flutter. Six blood cultures negative. Prolonged convalescence.	Blood. Throat washings (filtered).
VIII	20 yrs. F. 46477 (Presbyterian Hosp.)	1	Fever; migratory polyarthritis; pericarditis with effusion; pleurisy with effusion. Mitral disease. Prolonged illness. Recovery.	Blood. Pleural fluid.

[7] Of three subsequent blood cultures, only one grew a *Streptococcus viridans*.

[8] During first relapse.

Continued.

for animal inoculation.		Animals inoculated.					
Day of disease obtained.	Previous salicylate medication.	Generation.[3]					
		1st	2nd	3rd	4th	5th	6th
13th	?	R. − " +	R. − " −				
		G. P. ? " " −	G. P. + " " − " " +				
23rd	?	R. + " +					
25th	?	" −					
28th	None.	" −	R. − " + " − " +				
19th	"	" − " + " −					
21st[8]	" for 10 days.	" +	" −				
60th	" for 8 wks.	G. P. +					
4th	"	R. −	" +				
5th	"	G. P. +	G. P. −				
14th	?	R. − " +	R. −				
17th	?	G. P. +					

TABLE

Patients furnishing material.				Material use
Case No.	Age. Sex. Hospital. No.	No. of previous attacks of rheumatic fever.	Clinical features.	Material.[1]
IX	23 yrs. M. 4313	0	Fever; migratory polyarthritis; mitral regurgitation; lengthening of P-R interval in electrocardiogram; improvement under sodium salicylate therapy. Seven blood cultures negative. Tonsillectomy. Recovery.	Suspension of ton sils.[9] Suspension of tonsi (filtered). 18 hr. serum brot culture of ton suspension.[10] 18 hr. serum brot culture (filtered). 18 hr. serum bro culture (filtere plus 1.0 cc. br culture *Stre coccus viridans.*
X	14 yrs. M. 4348	0	Fever; mild polyarthritis which disappeared without sodium salicylate medication; mitral and aortic regurgitations. Tonsillectomy followed by relapse. Five blood cultures negative. Slow convalescence. Death from heart failure 5 mos. after discharge. No autopsy.	Suspension of to sils.[11] Suspension of tonsi (filtered).
XI	17 yrs. F. 4126	1	Repeated bouts of fever; mild transitory polyarthritis. Evidence of pancarditis. Three blood cultures negative. Tonsillectomy in 11th mo. Prolonged illness; slow convalescence. Two subsequent admissions. Death. *Autopsy.*—Numerous typical Aschoff bodies in myocardium; chronic and acute mitral and aortic endocarditis, and acute verrucous tricuspid endocarditis.	Suspension of tonsil (filtered).
XII	29 yrs. F. 4111	0	Fever; migratory polyarthritis, lengthening of P-R interval in electrocardiogram. Mitral and aortic valvular disease. Typical response to sodium salicylate. Two blood cultures negative. During convalescence an acute pharyngitis. Tonsillectomy. Recovery.	Throat washings.[12] Suspension of tonsi (filtered).

[9] Cultures grew hemolytic and non-hemolytic streptococci and Gram-negative diplococci.

[10] Gram-positive and Gram-negative cocci and bacilli.

[11] Cultures grew *Staphylococcus albus* and non-hemolytic streptococci.

[12] Cultures grew *Streptococcus viridans*, indifferent streptococci, and diphtheroids.

—*Continued.*

for animal inoculation.		Animals inoculated.					
Day of disease obtained.	Previous salicylate medication.	Generation.[2]					
		1st	2nd	3rd	4th	5th	6th
51st	3 gm. sodium salicylate a day for several wks.	R. −					
51st	3 gm. sodium salicylate a day for several wks.	" −					
51st	3 gm. sodium salicylate a day for several wks.	" −					
51st	3 gm. sodium salicylate a day for several wks.	" +					
51st	3 gm. sodium salicylate a day for several wks.	" −					
30th	None.	G. P. −	G. P. +				
30th	"	" " +					
11th mo.	"	" " +					
85th	Sodium salicylate discontinued 1 mo. before.	R. +					
125th	Discontinued 2 mos. before.	G. P. −					
		" " +					

TABLE

Patients furnishing material.				Material used
Case No.	Age. Sex. Hospital. No.	No. of previous attacks of rheumatic fever.	Clinical features.	Material.[1]
XIII	19 yrs. F. 46033 (Presbyterian Hosp.)	0	Fever; migratory polyarthritis. Improvement with subsequent relapse accompanied by severe pharyngitis. Mitral disease (?). Recovery.	Throat washings (filtered).
XIV	34 yrs. M. 4350	0	Fever; migratory polyarthritis; mitral endocarditis(?). Improvement under sodium salicylate therapy. Tonsillectomy followed immediately by relapse. Improvement under sodium salicylate therapy. Severe relapse with pericarditis and pleurisy, with effusion. Seven blood cultures negative. Recovery.	Suspension of tonsils (filtered). Joint fluid, left ankle.
XV	18 yrs. M. 4367	2	Fever; severe migratory polyarthritis; mitral and aortic valvular disease. Relief of symptoms by sodium salicylate followed by relapse after discontinuance. Joint aspirated at this time. Three subsequent relapses. Prolonged convalescence. Recovery.	Joint fluids, both knees. (Aspirated within a few days after onset of symptoms.)
XVI	19 yrs. F. 4329	0	Fever; migratory polyarthritis; pancarditis; pleurisy. Subcutaneous rheumatic nodules. Four blood cultures negative. Tonsillectomy. Slow convalescence.	Smith-Noguchi cultures of joint fluid left knee. Right knee.

[13] Guinea Pig B-86. Acute non-bacterial arthritis (see text).

ritis does not occur spontaneously in healthy rabbits or guinea pigs. The animals were in a good state of nutrition and had been gaining weight steadily until 2 or 3 days before the appearance of joint involvement. Trauma cannot be definitely ruled out, even though x-ray gave no evidence of injury to the hard parts.

A fairly common finding in both the rabbits and guinea pigs was the presence of bright red petechial hemorrhages into the synovial

Concluded.

For animal inoculation.		Animals inoculated.					
Day of disease obtained.	Previous salicylate medication.	Generation.[3]					
		1st	2nd	3rd	4th	5th	6th
40th	Sodium salicylate 4 gm. a day for 3 wks.	R. +	R. −	R. −	R. −		
			" −				
		G. P. +[13]	G. P. −				
			" " +				
21st	Sodium salicylate daily for 18 days. 4 gm. the day before tonsillectomy.	" " −	" " +				
		" " −					
6th	None for 6 days.	" " +	" " −				
17th	" " 5 "	" " +	" " +				
23rd	"	" " +					
		" " −					
30th	"	" " +					
		" " +					
		" " −					

membranes of the joints and into the periarticular tissues. As they were found in uninoculated control animals, and as microscopic examination showed them to be fresh and devoid of any inflammatory reaction, they were thought to be agonal in origin.

Myocardial Lesions.—The microscopic examination of the hearts of all of the rabbits and guinea pigs revealed the presence of myocardial lesions in a large percentage. Their presence or absence has been indicated by + and − marks in Table I.

The lesions most commonly consisted of groups of lymphocytes to which were sometimes added endothelial leucocytes in varying proportions. Polymorphonuclear eosinophils and more or less mature connective tissue cells were not uncommon. Plasma cells, mast cells, and polymorphonuclear neutrophils were rather rare. The

TABLE II.

The Relation of Age to the Incidence of the Myocardial Lesions. (Weight Has Been Used as an Approximate Index of Age.)

	Rabbits.			
	Less than 500 gm.	500–999 gm.	1,000–1,500 gm.	Over 1,500 gm.
No. of rabbits receiving initial inoculations	2	14	9	2
" " such rabbits showing myocardial lesions	1	3 (21%)	6 (66%)	2
No. of rabbits receiving transfer inoculations	5	26	7	1
" " such rabbits showing myocardial lesions	1 (20%)	9 (29%)	4 (57%)	1
Per cent of the total in each weight group showing myocardial lesions	28	30	62	

	Guinea pigs.		
	Less than 300 gm.	300–500 gm.	Over 500 gm.
No. of guinea pigs receiving initial inoculations	9	10	2
" " such guinea pigs showing myocardial lesions	6 (75%)	6 (60%)	2
No. of guinea pigs receiving transfer inoculations	10	5	4
" " such guinea pigs showing myocardial lesions	7 (70%)	4 (80%)	2
Per cent of the total in each weight group showing myocardial lesions	68	66	

arrangement of the cells was not strikingly constant. The various types of cells seemed to be mixed together without order. The lesions tended to assume an elongated, irregularly elliptical shape, their long diameters parallel to the adjacent muscle fibers. The more extensive lesions filled the spaces between a number of fibers. In

such instances the muscle cells sometimes appeared normal and sometimes showed loss of striation. But evidences of necrosis were uniformly absent except in the hearts of rabbits which had had a terminal bacteremia.

The lesions were most frequently found in the myocardium of the papillary muscles and septum, less often in the walls of the ventricles and only occasionally in the auricles. Some lesions occurred subendocardially and subepicardially. None was ever found in the valves.

In the rabbits the incidence of the myocardial lesions was roughly proportional to their ages, but this was not true for the guinea pigs (see Table II). As the exact ages of the animals were not known,

TABLE III.

Incidence of Myocardial Lesions in Animals Injected with Human and with Homologous Substances.

Of 27 rabbits inoculated with human material	12 (or 44 per cent) showed heart lesions.
Of 40 rabbits inoculated with transfer material from other rabbits	19 (" 47 " ") " " "
Of 21 guinea pigs inoculated with human material	14 (" 66 " ") " " "
Of 19 guinea pigs inoculated with transfer material from other guinea pigs	13 (" 68 " ") " " "

their weights were used as approximate indexes of age. In these tables the animals have been separated into two groups, those which had been injected with human material (initial inoculation) and those which had been injected with transfer material from other animals of their own species (transfer inoculations).

This grouping is necessitated by the suspicion which naturally comes to mind that the lesions might have been the result of inoculations with foreign proteins and be similar in nature to those described by Longcope (10). That this is not the case is shown by Table III.

The lesions occurred with equal frequency in animals receiving human material and in those receiving blood and tissue suspensions from animals of the same species, and cannot, therefore, be regarded as a response to the inoculation of foreign proteins.

Lesions in every way identical with those described were found in control animals, both inoculated and uninoculated.

DISCUSSION.

With the exception of the two instances of outspoken arthritis already discussed no evidence of successful transmission of rheumatic fever was encountered in these experiments. As has been mentioned, non-bacterial arthritis is not met with in healthy rabbits and guinea pigs, and its occurrence in animals injected with material from patients with rheumatic fever is interesting and suggestive. But the presence of arthritis in those two animals does not justify the conclusion that they were infected with rheumatic fever.

The transmission of rheumatic fever to laboratory animals is an important problem, one which must not be abandoned, for on its solution may rest the elucidation of the etiology of the disease. Our failure to accomplish it does not convince us that the active agent was not present in some of the materials with which we were working. A more reasonable explanation for our failure seems to be the inability of the agent to gain a foothold in the species of animal employed under the conditions of our experiments. And it is with the hope that other workers may be hereby stimulated to attack the problem that this report is published.

The microscopic lesions in the hearts of the inoculated animals are of particular interest for several reasons. They might have been mistaken for Aschoff bodies, for occasionally one was seen which on casual observation could have been passed as an Aschoff body, not typical, to be sure. But one might not expect the tissues of the rabbit or guinea pig to react exactly as human tissues do to a human virus.

As lesions apparently identical were found in uninoculated control animals, one is forced to conclude that they occur spontaneously in rabbits and guinea pigs. This point is dealt with in more detail in other communications (11). It is obvious, therefore, that when myocardial lesions are discovered in animals subjected to experimental inoculation great caution must be exercised in the interpretation of their significance. As a similar myocarditis was considered

by de Vecchi and Natali to be proof of the production of experimental rheumatism in their rabbits, and comparable foci were ascribed by Herry to the action of an "*endotoxine rhumatismale*" from streptococci, we feel that the interpretation of these authors of the etiology and nature of the lesions found by them cannot be accepted without further proof.

SUMMARY.

In a series of attempts to transmit a virus from patients in the acute stages of rheumatic fever, twenty-seven rabbits and fourteen guinea pigs were inoculated with one of the following materials: whole blood, serum, joint fluid, pleural fluid, throat washings, suspensions of tonsil tissue. Subsequent transfer inoculations from animal to animal brought the total number of animals employed in the experiments to 67 rabbits and 40 guinea pigs. Only two animals developed an acute non-bacterial arthritis. No other evidence of successful transmission of the disease was obtained.

In about one-half of the rabbits and two-thirds of the guinea pigs myocardial lesions were encountered which consisted of interstitial accumulations of lymphocytes and endothelial cells. Similar lesions were found in control animals.

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