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Epidemiological Significance of Repeated Infections with Homologous and Heterologous Strains and Species of *Plasmodium*

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During many years of experience with the use of induced malaria for the therapy of neurosyphilis, it has been possible to observe the course of malarial infections in patients with known previous history of infection by the same and different strains and species of Plasmodium. *This report presents the effect of prior infection on subsequent infection in regard to immune response of the host, clinical symptoms, parasitaemia, transmissibility and chemotherapeutic measures. These studies have shown that in no case was it impossible to induce additional infections in patients. However, in all homologous-species reinoculations, there were significant modifications of the infections, which were enhanced if both exposures were to the same strain. Variable results (ranging from no apparent effect to a fairly substantial modification) were seen after reinoculation of patients with a heterologous species of* Plasmodium.

The use of immunofluorescent methods may provide a clearer understanding of the mechanism of partial immunity as it affects reinfection. The relationship of this partial immunity to the epidemiology and chemotherapy of the disease in endemic areas is of importance, in that these infections in semi-immune persons provide a continuing and unapparent source of reinfection of the community.

Much of the experimental work on the biology and chemotherapy of human malaria has been done using infections induced in patients with no previous history of malaria. There have long been suggestions that these infections differ significantly from those seen in areas where malaria is endemic, where risk may begin at infancy and where subsequent infection patterns are complicated by the presence of pre-existing infection or partial immunity. While this assumed defect in no way detracts from the importance of studies in non-immune persons, it does suggest some difficulties in the extrapolation of these observations to immune populations.

During the years that induced malaria has been utilized in the therapy of neurosyphilis by our laboratories, there has been considerable opportunity to observe the course of malarial infections in patients with known previous history of the same and different strains and species of *Plasmodium*. These secondary infections appear to present problems in epidemiology and chemotherapy that may be of importance in efforts toward the control and eradication of malaria in areas where it is endemic. Included in this study are the effects of prior infections on the subsequent infection in regard to immune response of the host, clinical malaria, parasitaemia, the transmissibility of the infection and chemotherapeutic measures. While the first-mentioned characteristic, i.e., the immune response in the host, undoubtedly can be incriminated as the primary causal factor in the additional considerations, the others will be reported in some detail because of their peculiar importance to the practical consideration of endemic malaria and its control.

METHODS

All studies were done in adult mental patients undergoing treatment for neurosyphilis. The patients included members of both sexes and were about equally divided between Negroes and Caucasians.

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Malaria was induced through the bites of infected mosquitos or by the transfusion of small quantities of blood containing infective parasites. Observations on the recipients included daily blood films, with determination of asexual parasite and gametocyte densities, temperature readings at 4-hour intervals during the course of the infection (except during periods of fever, when readings were done hourly) and frequent feeding of mosquitos to determine infectivity of the parasitaemias. Suppressive and curative drugs were administered at appropriate times, and observations on the effect of the drugs on parasitaemias, symptoms and infectivity to mosquitos carefully made. Drugs used for the management of the symptomatic attack included quinine, bismuth thioglycollate and chlorguanide, in minor dosages.

In selected patients, serum or plasma was collected prior to and periodically during the infection for detection of antibody levels by immunofluorescent techniques. The indirect immunofluorescent method was used and has been described in some detail elsewhere (Collins, Jeffery & Skinner, 1964a).

The species and strains of *Plasmodium* used are listed in Table 1 and include 5 strains of *P. falciparum*, 4 of *P. vivax*, 2 of *P. ovale* and 2 of *P. malariae*. Mosquitos used in the infectivity studies included various strains of *Anopheles quadrimaculatus*, *A. albimanus* and *A. freeborni*.

TABLE 1
SPECIES AND STRAINS OF *PLASMODIUM* USED

Species and strains	Probable origin
P. vivax	
Chesson	New Guinea
Pait	New Guinea
St. Elizabeth	USA
Korean	Korea
P. falciparum	
Santee-Cooper	South Carolina, USA
McLendon	South Carolina, USA
Panama	Panama, Central America
Colombia	Colombia, South America
Thailand	Thailand
P. malariae	
USPHS	USA
Trinidad	Trinidad, West Indies
P. ovale	
Donaldson	Philippines
Liberian	Liberia, West Africa

Reinoculations of patients were done at variable periods following the termination or suppression of prior infections, the actual intervening time period varying from several weeks to several years. The effect of the prior inoculations on the subsequent ones will be discussed on the basis of the use of homologous or heterologous strains and species of parasite.

No differentiation is made between blood- and sporozoite-induced infections, since there was no apparent difference in the host response to the two types of infection.

OBSERVATIONS

Plasmodium vivax

A series of 23 patients received multiple inoculations with *P. vivax*; in 6 of these cases, several heterologous strains were used, while the remainder were homologous inoculations (Chesson strain). Two of the patients received 4 inoculations; 4 received 3 inoculations and the remainder 2 inoculations.

In no case was an infection completely prevented by the occurrence of single or multiple prior experiences with the same or a different strain of *P. vivax*. In all cases there was a degree of modification of the second infection. Second inoculations were done at intervals ranging from 6 to 37 months following the initial exposure, and there seemed to be little difference in the general immune response between those done after shorter periods and those after longer ones. It was observed that in most secondary infections the parasitaemia and symptoms developed as early and as rapidly as might be expected in a primary infection, but spontaneous termination of symptoms occurred within a short time and parasite densities decreased prior to the time the usual peak might be expected in non-immune persons.

Table 2 summarizes the differences in parasitaemia and symptomatic response in the first and second infections with *P. vivax* (Chesson strain). In addition to the obvious difference in the number of paroxysms experienced by various patients, it should be noted that, during the initial infection, it was almost always necessary to intervene with minor amounts of drug to keep the symptomatic infection within manageable limits and often to terminate the infection prior to

TABLE 2

CHARACTERISTICS OF FIRST AND SECOND HOMOLOGOUS INFECTIONS WITH *P. VIVAX* (CHESSON STRAIN): 20 CASES

Infection	No. of fevers >100° F (37.8° C)		Maximum blood parasites per mm³	
	Range	Mean	Range	Mean
First	3-22	14.6	5 664-99 360	37 820
Second	1-15	6.1	40-41 400	10 587

the completion of the symptomatic attack, thereby effectively reducing the number of febrile episodes and the maximum parasite density. It was seldom necessary to resort to such partial suppression during the second infection, and the number of paroxysms represented approximately the total course of the attack prior to self-limitation. Presumably, the more than threefold difference in average maximum parasitaemia between the two infections would have been even greater had not the early drug intervention been so commonly resorted to.

Modification of heterologous second infections was less pronounced than that seen in the homologous reinfections. A third exposure to the homologous strain demonstrated a greater resistance of the host to the infection than did the second one. When a heterologous strain was induced following two or three previous *P. vivax* infections, the pattern of resistance was essentially the same as if there had been only a single previous exposure.

Parasitic relapses in sporozoite-induced infections appeared to occur as often in cases with prior experience with the homologous strain as in first exposures. However, none o the former were symptomatic, in contrast to thos associated with first infections where symptoms were the rule. Where a heterologous strain was used as a second, third, or fourth infection, symptomatic relapses were usual.

Infectivity to mosquitos of the initial infection generally continued at a high level until the attack was terminated by drug. Such drug intervention was not usually necessary during the subsequent infections and, in a number of these cases, infective asymptomatic parasitaemias continued for considerable lengths of time. In one instance these cases were infective to mosquitos until the 49th day of infection, at which time it was terminated. Gametocyte densities in secondary infections were on the whole somewhat lower than in the primary infection, but these lower gametocytaemias often appeared to be more highly infective, as determined from comparable mosquito feedings.

Parasitaemia and symptoms in secondary infections were more easily terminated by comparable dosages of antimalarial drugs than in primary cases. In the few cases where subcurative dosages of drugs were used to curb the symptomatic attack during the second infection, the response was complete, with no subsequent return of the symptoms. In contrast, such treatments during the primary infection were usually followed by a renewed clinical attack. The clearance of parasites by means of "curative" regimens was usually much more rapid in the second infection than in the first, but this was probably a function of the lower parasite densities at the time of treatment rather than an indication of greater drug sensitivity.

Plasmodium falciparum

Eleven patients received dual inoculations with *P. falciparum*; in 4 of these cases the inoculations were with a homologous strain (Panama) and the remainder with strains of widely separated geographical origins. Three additional patients received 3 or 4 successive inoculations with heterologous strains. In none of the trials did the second or subsequent infections fail to develop, although, as in *P. vivax* infections, there was a considerable modification in each case. Also as in *P. vivax* infections, the general observation was made that the secondary infection tended to develop normally during its early stages, with clinical symptoms occurring as early as usual and fairly high parasite densities being reached within the first few days. However, the peak parasitaemias were lower and occurred somewhat earlier than in non-immune subjects, after which there was a relatively rapid decrease in parasitaemias and an abrupt cessation of the symptomatic attack. In general, the 4 homologous reinfections were considerably milder than those where heterologous strains had been employed. In the 3 cases receiving 3 or 4 heterologous inoculations, the pattern was usually much the same; each infection progressed normally during the first few days, but spontaneous termination of the clinical attack and decrease of parasitaemias to lower but persistent levels occurred within a comparatively short time. In the single exception to this, the third infection reached a maximum parasitaemia of only 420 parasites per mm³, experienced no clinical attack and maintained intermittent low parasitaemias for only 50 days.

Table 3 summarizes the differences in maximum parasitaemia and symptomatic response to the first and second infections with homologous and heterologous strains of *P. falciparum*. It should be noted that frequent intervention with minor amounts of drug was necessary in the primary infections, which effectively reduced the number of febrile episodes which might normally have been expected. Such intervention was uncommon during the second infection. The decrease of asexual parasite densities in the homologous immunes was considerably more pronounced than in the heterologous reinoculations, and this phenomenon could also be seen in the maximum gametocyte densities. Considering all cases, there was more than a fourfold decrease in asexual maximum densities and nearly a fivefold decrease in the gametocytes. Early or minor drug intervention undoubtedly decreased the maximum asexual parasite densities in the first infection, but the gametocyte maximum was probably affected to a lesser degree. The total span of parasitaemia in cases allowed to continue to spontaneous termination varied little between immunes and non-immune cases; in 6 cases of the former, the parasitaemia continued for an average of 207 days, while in 8 of the latter the duration averaged 237 days.

As with *P. vivax*, there appeared to be little difference in the infectivity to mosquitos of comparable gametocyte densities in first and second infections. Although the gametocytaemias in the immunes were lower, they persisted as long as in the non-immunes and were usually above the minimum number necessary to produce infection in mosquitos; the resulting mosquito infections were proportionally lower in intensity.

TABLE 3

CHARACTERISTICS OF FIRST AND SECOND INFECTIONS WITH *P. FALCIPARUM*

Infection	No. of fevers >100° F (37.8° C)		Average maximum blood parasites per mm³	
	Range	Mean	Asexual	Gametocytes
Homologous strains (4 cases)				
First	4-16	11.5	144 400	2 778
Second	0-26	12.8	23 190	568
Heterologous strains (7 cases)				
First	2-24	12.0	49 493	1 200
Second	0-10	4.9	17 958	288
Total (11 cases)				
First	2-24	11.8	85 823	1 976
Second	0-26	7.7	19 861	428

There was little opportunity to observe any difference in response to drug in the first and second infections. The standard dose of drug for termination of an infection was 1.5 g of chloroquine (base); lower doses were not tested in the immune patients. In only a single case was modification of the second infection by a low dose of quinine (10 grains, or 0.65 g) deemed desirable. In this instance the patient had had only a brief but severe primary infection, which was terminated early. Although the use of this partial suppression eliminated the febrile attack for only 2 days, it effectively reduced the parasitaemia to manageable levels, and there appeared to be a greater effect than would have been seen in the absence of prior *P. falciparum* experience.

In three patients inoculated with heterologous strains of *P. falciparum*, it has been possible to determine the levels of specific antibody to each strain following the second inoculation, using fluorescent-antibody methods. These results have been reported elsewhere by Collins, Jeffery & Skinner (1964c) but will be briefly repeated here, since they may throw some light on the mechanism of partial immunity to different strains of the same species. All 3 subjects had been infected by the Colombia strain of *P. falciparum* approximately 18 months prior to inoculation with the Thailand strain. Antibody levels were determined weekly during the course of the second infection. At the time of inoculation, low levels were present for the prior strain (1 : 10 to 1 : 40); within a few days after the parasites became patent there was a significant increase in the specific antibody for the Colombia strain, accompanied by a slower response on the part of the Thailand antibody. Within 11, 15 and 32 days after the appearance of parasites, the Colombia antibody had risen to high titres, but it was not until much later in the course of infection that the antibody of the second strain approached the levels present for the initial infection, and generally the titres continued at lower levels during the entire infection. These studies suggest the possibility of the presence of strain-specific as well as species-specific antibody, with stimulation of the species-specific antibody by a heterologous strain early in the course of infection and somewhat of a lag in the production of the strain-specific antibody. Correlation of the antibody studies with the course

of events seen in the development of parasitaemia and symptoms in heterologous reinoculations may explain the usual rapid and normal development of the infection and the rather abrupt spontaneous termination. This termination apparently occurred at a time when the antibody to the initial strain had increased significantly and the antibody to the second strain had reached levels where an additive effect on the infection became operative.

Plasmodium ovale

Ten patients were inoculated with a heterologous strain of *P. ovale* (Liberian strain) following an initial course of infection with the Donaldson (Philippine) strain, and a single patient received 2 inoculations with the homologous strain (Donaldson). In none of these cases was the second infection prevented, although there was a considerable modification of its severity. Three of the 10 heterologous infections and the single homologous infection were asymptomatic, and the maximum parasitaemias ranged from 70 per mm^3 to 1130 per mm^3. The mean number of paroxysms in the second inoculations, considering only those subjects with symptoms, was 2.4, while the usual number in non-immunes is 9.4. The mean maximum parasitaemia was also significantly decreased from over 10 000 per mm^3 to about 2000 per mm^3 in immune persons. In general, there was a direct correlation between the degree of immunity to a second infection and the lapse of time between the 2 inoculations. The persistence of parasitaemias in spontaneously terminating cases often appeared to be as long with the second experience with *P. ovale* as with the first; in 1 immune subject, these parasitaemias persisted at low levels for a total of 132 days, although only 2 febrile episodes had occurred during the acute attack. Infectivity to mosquitos appeared to be considerably lower during second infections than during first infections, but this decrease was proportionate to the lower total parasite densities.

P. ovale is characterized by its sensitivity to antimalarial drugs, and there was no opportunity to observe any differences between the first and second infections with this species in regard to drug effectiveness.

Plasmodium malariae

Our studies produced little information concerning the use of multiple inoculations with this species. In the single such case in our series, a USA-strain infection that terminated spontaneously was followed within a month by inoculation with a strain from Trinidad. The second infection was not prevented, but again the parasitaemia and symptoms were modified. There was a parasite maximum of only 1280 per mm^3, which was only about one-tenth that of the first infection, and the febrile attack consisted of only 7 mild episodes, as compared with 22 during the first infection.

Interspecific relationships

In a number of cases, heterologous-species inoculations were done, and the effect of the first on the second or subsequent infections was noted. The findings are summarized in Table 4 and will be discussed briefly.

P. vivax–P. falciparum. Neither of these species appeared to confer much protection against reinfection by the other. In a series of 15 cases where *P. vivax* infection was followed by *P. falciparum* infection, the latter was characterized by the following average values: maximum asexual parasitaemias of 155 727 per mm^3, maximum gametocytaemias of 3627 per mm^3 and 13.3 febrile episodes. All of these are at least as high as one would note in a primary infection by the same species. Further, partial suppression or early termination was necessary to the usual degree in the management of these cases, probably altering to some extent the normal course of the infection. It is also of interest that 9 of the 15 cases had experienced 2 or 3 previous *P. vivax* infections, with 6 having had only one prior exposure. The *P. falciparum* infections appeared to be normal in all respects, including response to antimalarial drugs and infectivity to mosquitos.

In the only case in which *P. vivax* (Chesson strain) infection was induced subsequent to a *P. falciparum* infection, the *P. vivax* infection did not appear to be significantly altered from the normal. This infection reached a maximum parasite density of about 45 000 per mm^3, and there was a total of 12 febrile episodes. The infection terminated spontaneously, and partial suppression was not needed during the clinical attack. It should be pointed out that the patient was a Negro, and that there is often full or partial immunity to this strain of *P. vivax* in Negroes.

P. falciparum-P. malariae. In 6 cases, *P. falciparum* infection was induced in patients who had already experienced *P. malariae* infection. The *P. falciparum* infections in these subjects did not appear to vary from those seen in non-immune subjects. The maximum asexual parasite densities averaged 165 000 per mm^3 and the gametocytes 2780 per mm^3, and there was

TABLE 4

CHARACTERISTICS OF SECONDARY INFECTIONS IN PATIENTS WITH PRIOR HETEROLOGOUS-SPECIES EXPERIENCE

Species		No. of cases	Average maximum number of blood parasites per mm³				Average number of paroxysms	
			Asexual		Gametocytes			
Initial	Secondary		Normal [a]	Secondary	Normal [a]	Secondary	Normal [a]	Secondary
P. vivax	*P. falciparum*	15	85 823	155 727	1 976	3 627	11.8	13.3
P. falciparum	*P. vivax*	1	37 820	45 000	—[b]	—[b]	14.6	12.0
P. falciparum	*P. malariae*	2	Second infection normal			—	—	—
P. malariae	*P. falciparum*	6	85 823	165 900	1 976	2 780	11.8	12.5
P. vivax	*P. ovale*	15	8 314	7 916	—[b]	—[b]	8.1	5.8
P. ovale	*P. vivax*	5	37 820	29 556	—[b]	—[b]	14.6	15.0
P. falciparum	*P. ovale*	11	9 000	7 073	—[b]	—[b]	9.0	7.3
P. ovale	*P. falciparum*	11	85 823	63 371	1 976	1 408	11.8	12.4
P. vivax	*P. malariae*	2	Second infection normal			—	—	—
P. malariae	*P. vivax*	3	Two of three second infections modified					—

[a] Normal values for parasitaemias and number of paroxysms are derived from observations on groups of comparable patients (ranging from 11 to 103 per group) who had not experienced prior malaria.

[b] Because of inherent inaccuracies in gametocyte densities of *P. vivax* and *P. ovale*, these values were not considered.

an average of 12.5 paroxysms. In all other respects, including response to antimalarial drugs, need for partial suppression in management of the infection and infectivity to mosquitos, the *P. falciparum* infections appeared normal.

Inoculation of patients with *P. malariae* following prior *P. falciparum* infection was done in only 2 cases, and little definitive information is available. However, in these 2 trials, there appeared to be little significant effect of the first infection on the course of the *P. malariae* infection.

P. ovale–P. vivax. Earlier experience with the Chesson strain of *P. vivax* appeared to exert only a slight effect on subsequent infections with Donaldson strain *P. ovale*. In 15 such subjects, 10 of whom had experienced more than one prior *P. vivax* infection, the average maximum parasite density was 7916 per mm³; non-immune *P. ovale* patients averaged 8314 per mm³. The average number of paroxysms in the multiply infected group was 5.8, while in the non-immune it was 8.1. It would appear that the symptomatic course of the infection may have been shortened somewhat by the prior *P. vivax* infection, but the difference did not seem great.

In 5 patients, *P. vivax* was induced subsequent to a prior *P. ovale* infection. In these cases there appeared to be an equally slight effect on the second infection. The maximum parasite average was 29 556 per mm³, and the number of paroxysms was 15, both of which are close to normal averages for this species. However, in none of these cases was early intervention with drug a necessity, and in all of them the clinical attack ceased and the elevated parasitaemias decreased spontaneously.

P. falciparum–P. ovale. In 11 patients, *P. falciparum* infection was followed by subsequent *P. ovale* inoculation (Donaldson and Liberian strains). In these cases there was an average maximum parasite density of 7073 per mm³, which is slightly lower than the average of about 9000 for this species. The average number of paroxysms in the group was 7.3, which is also somewhat less than the average of about 9 seen in non-immune subjects. There were no other apparent differences in the course of infection, and the degree of immunity conferred by the initial infection seemed minor.

Observations of 11 subjects initially infected with *P. ovale* and subsequently with *P. falciparum* pro-

duced some interesting findings. While in no case was the second infection suppressed by the earlier one, even in one instance where patent parasites of *P. ovale* were present at the time of the second infection, there was in every case an obvious modification of the second infection. The *P. falciparum* infections exhibited the following averages: maximum asexual parasite densities, 63 371 per mm^3; maximum gametocyte densities, 1408 per mm^3; number of paroxysms, 12.4. While these do not vary widely from the normal values shown in Table 3 (85 823 asexual parasites per mm^3, 1976 gametocytes per mm^3, and 11.8 paroxysms), there was a striking difference in the proportion of cases in which drug intervention was necessary and in the amount of drug needed for partial suppression of the parasitaemias. In 5 of the 11 cases, the symptomatic attack terminated spontaneously without resort to drugs after 5 to 15 paroxysms; in 4 of the 6 subjects receiving small drug dosages, the attack subsided either immediately or within a very few days. It should be noted that, in the large majority of first infections, with the strains of *P. falciparum* used, early intervention with suppressive or curative drugs was the rule, which effectively depressed the maximum parasite densities and the number of paroxysms but failed to terminate the attack. In this series of cases, all symptomatic attacks terminated spontaneously without the use of curative drug dosages although, in some patients, minor amounts of drug were used.

Other interspecific relationships

It was possible to observe a few cases in which *P. vivax* infection either preceded or followed one with *P. malariae*. In 2 patients where *P. malariae* was the second infection, there was no obvious immunity. Parasite densities proceeded to high levels and it was necessary to terminate both prior to the end of the clinical attack. In 2 of 3 cases in which *P. vivax* infection followed *P. malariae* infection, some modification seemed to occur. In both of these cases, the early attack was normal, but spontaneous reduction of parasitaemia and symptoms followed 8 to 10 paroxysms. In the third case, it was necessary to terminate the infection after 8 paroxysms because of an unusually high parasitaemia (126 000 per mm^3).

There was no opportunity to observe the possible interrelationships of *P. ovale* and *P. malariae* infections.

DISCUSSION

Perhaps the earliest report on the differing characteristics of first and subsequent infections was that of James & Shute (1926). In their classic report on the results of laboratory work on malaria in England, they noted the failure of a prior experience with *P. vivax* to prevent completely, in any instance, subsequent infection with heterologous or homologous strains, but they also found a significant alteration of the course of infection. Since that time there has been a considerable variability in the reports of such acquired immunity. Some authorities have reported a complete and solid immunity, persisting for long periods, to homologous and even heterologous strain reinoculation with *P. vivax*, while others have noted complete immunity in homologous- but not in heterologous-strain reinoculations. It is of interest that the majority of reports showing absence of total immunity did note varying degrees of modification of subsequent infections.

Extensive studies conducted over many years in Romania at Socola and Berceni have been reported by Ciuca (1955), summarizing the results of multiple inoculations of various strains and species of malaria in neurosyphilitics. Our smaller series of cases generally confirms the findings of Ciuca. With *P. vivax*, he noted reduced parasitaemias and clinical attacks in second infections, with very few showing a " true " immunity after reinoculation with either homologous or heterologous strains. Ciuca's experience with *P. falciparum* was more limited. In 6 such cases, one subject reinoculated after 5 months was completely resistant, while in the remaining 5, who were inoculated after about a year, there was a low degree of acquired resistance. With *P. ovale*, Ciuca found that the initial infection produced a partial resistance toward reinfection with the same strain that decreased if the interval between the 2 inoculations was greater than a year. In a large series of cases reinoculated with heterologous species, Ciuca found little or no cross-immunity.

Similar detailed studies conducted at Horton Hospital, Epsom, England, have been briefly summarized by Covell & Nicol (1951). Their finding of a " remarkable solidarity of the tolerance acquired against the particular strain of malaria parasite used for the original infection " is somewhat in variance with our own. Covell & Nicol did note the presence of only partial protection when heterologous strains of *P. vivax* or *P. falciparum* were used and an absence of tolerance when heterologous species were involved.

Bray et al. (1962) in Liberia and Bruce-Chwatt (1963a, 1963b) in Nigeria have reported the results of inoculations of populations in endemic areas with

P. falciparum. The results of these two studies were quite similar and indicated that over one-half of the subjects inoculated developed patent parasitaemia that could be related to the inoculation. In a significant proportion of the recipients in the two studies (25% and 15%), these parasitaemias were accompanied by clinical symptoms. It seems likely that most of the inoculations reported actually represented superinfections, since the parasite rates in the groups under study were fairly high. In view of this probability, the imposition of a detectable secondary infection is of considerable interest.

Our studies have shown the complete absence of a " solid " immunity, even where homologous strains were concerned. While this observation appears to be incompatible with some earlier reports, it should be pointed out that in our cases the initial infection was invariably eliminated prior to challenge by a homologous strain. In reports of other investigators who found that the prior infection had prevented completely the re-establishment of the homologous strain, the presence of subpatent parasitaemias at the time of challenge was a possibility in many cases. Recent studies on the persistence of specific antibody (Collins, Jeffery & Skinner, 1964a, 1964b) have shown that high antibody levels persist during terminal periods of low parasitaemia and would undoubtedly be operative in the prevention of any increase in parasitaemia caused by homologous reinoculation. In most cases, this antibody level decreases sharply to a low but persistent level following the termination of the infection. This lower antibody level appears to be insufficient to prevent re-establishment of the infection, but its rapid stimulation to high levels by the reintroduction of the antigen serves to modify quickly the course of the second infection. This appears to be the case with both homologous and heterologous strains of the same species, and may operate to some extent in reinoculation with heterologous species.

Earlier reports have not credited an initial infection with much effect on later infection by heterologous species, and such subjects have usually been considered non-immune. Our studies appear to confirm this conclusion, but with some reservations that may be of importance. There appears to be no cross-protection between *P. falciparum* and *P. vivax* infections nor between those by *P. falciparum* and *P. malariae*. Where *P. vivax* and *P. ovale* infections were concerned, the effect of one on the other was slight, but perhaps significant, as is evident from a reduction in the maximum parasite densities and number of febrile attacks. Of particular interest was the absence of the necessity for partial suppression in *P. vivax* infection following earlier *P. ovale* infection, indicating that, in all cases observed, the *P. vivax* infection was self-limiting under these conditions.

Perhaps the most interesting observation concerned the *P. ovale–P. falciparum* relationship. *P. falciparum* infection exerted only a slight effect on later *P. ovale* infection, but when *P. ovale* infection preceded *P. falciparum* infection, there appeared to be a significant alteration in the *P. falciparum* infection. In view of the fact that little partial suppression was needed in the 11 cases studied, the moderate reduction in parasite densities probably has considerable significance. It is probable that at least 9 of the 11 cases would have terminated spontaneously, although small amounts of drugs were used in 4 of them, resulting in rapid and permanent abortion of the clinical attack. In 2 other cases the symptoms reappeared after partial suppression but terminated spontaneously after a few additional paroxysms.

Little of the information on strain and species immunity has been applied to the determination of its possible significance in the epidemiology or control of malaria, other than the general recognition of the role of acquired immunity in the prevalence and intensity of the infection in various age-groups. Macdonald's (1957) analysis of the effect of acquired immunity on the transmission of malaria under conditions of endemicity and control emphasizes the possible reduction of gametocytaemias in immunes as an overriding consideration, incriminating the non-immune portion of the population, i.e., infants and children, as an important factor in the continuation of transmission. It is of interest that Macdonald points out the extreme difficulty in the control of " stable malaria ", where a stable immunity exists in the greater proportion of the population, in contrast to the much greater ease of control of " unstable malaria ", where immunity would be quite variable. While this variation in the ease of control might not be a function of the immunity of the population, it appears obvious that the presence of the immune status does not contribute greatly to the ease of control.

The results of the present studies may be useful for the interpretation of the significance of partial immunity as it affects the transmission and perpetuation of malaria in endemic situations. However, it should be pointed out that immunity acquired after one, two or three infections, as in our studies, may not be comparable to that derived from superinfections continu-

ing year after year. Any conclusions as to the effects of partial immunity that may be based on our findings should be interpreted within the limits of the experiments that we have described.

The effect of this partial immunity on the epidemiology and control of malaria may, in some ways, be disadvantageous to human populations. While the individual most certainly benefits from such immunity by reason of the early remission of the milder clinical attack, the early establishment of this host-parasite equilibrium also serves to enhance the success of the parasite in maintaining itself in the population. The more severe and long-lasting clinical attack of malaria in a non-immune person would, with considerable frequency, result in either early termination by radical treatment or death, while the brief symptomatic attack that characterizes reinfection with the same species, and even on occasion with other species, is less likely to be subjected to curative drug therapy.

Although the potential for transmission in these secondary infections may be somewhat lower than in non-immune cases, it cannot be overlooked as a source of mosquito infection. Our studies indicate an early and persistent infectivity to mosquitos, often of a higher quality than that produced by comparable gametocytaemias in primary cases. In the case of *P. falciparum*, low but infective gametocytaemias persisted as long as in those infections induced in non-immune subjects; further, the non-immune cases usually required intervention with a non-curative drug to avoid the necessity of curative intervention as an alternative to severe consequences. With *P. vivax* and *P. ovale* infections, there was also a frequent extension of low, asymptomatic parasitaemias in association with the secondary infections. These parasitaemias were capable of infecting mosquitos to a limited extent and must also be considered an important reservoir for transmission.

Numerous investigators have noted the comparative ease of treatment of immune patients in endemic areas as compared with treatment of non-immune patients, even to the point of describing a " synergism " of drug and immunity. Our observations offer little additional evidence in this regard, but in general would confirm the enhanced activity of various dosages of drug on infections in patients with prior malarial experience. The use of the minor dosages of drug and the ensuing symptomatic control of the infection in semi-immune patients may parallel similar practices in the field, where inexpert medication or self-medication with non-curative drugs and dosages may eliminate the clinical attack in an individual but not necessarily his potential as a source of infection in the community. This unhappy situation may become more common as efforts toward treatment and control of malaria are expanded. Curative drugs and facilities for their use in clinical malaria are becoming more easily accessible in many areas, making possible the radical cure which, in many cases, will leave the individual susceptible to new infections that may not require treatment but that will allow him to be a source for perpetuation of the disease. This would suggest that the treatment of clinical cases only would be quite inadequate as a measure for malaria control in an area where transmission potential is continual. If chemotherapeutic measures are to be relied upon to any great extent, their use must be extended to a much larger segment of the infected population through the use of such measures as dietary drugs, repository drugs that can be universally administered, or thorough and repeated case-finding and treatment. Whether or not any of these methods is practicable remains to be seen.

RÉSUMÉ

De nombreuses années d'expérience du traitement de la neurosyphilis par la malariathérapie ont permis d'observer l'évolution des infections paludéennes chez des malades dont l'infection antérieure par des *Plasmodium* de même souche ou de souches ou d'espèces différentes était connue. L'auteur a étudié l'effet d'une atteinte initiale sur la réponse immunitaire de l'hôte, les symptômes cliniques, la parasitémie, la transmissibilité et les mesures chimiothérapeutiques, au cours d'une réinoculation après un intervalle de plusieurs semaines à plusieurs années.

L'expérimentation a montré l'absence d'une immunité solide, même en cas de réinoculation de souches homologues. Ces résultats ne semblent pas confirmer ceux d'observations antérieures, mais l'auteur insiste sur le fait que chez ses patients l'infection initiale avait été complètement éliminée avant la réinoculation d'une souche homologue. Dans tous les cas, on a pu provoquer une infection supplémentaire, que la première infection ait été due à une souche ou espèce homologue ou hétérologue. La réinoculation d'une espèce homologue a entraîné une infection dont l'allure était sensiblement modifiée par rapport à l'atteinte initiale. Ce phénomène était plus accentué en cas de réinfection par la même souche, la régression spontanée des symptômes et la

chute de la parasitémie survenant plus précocement. L'inoculation d'une espèce hétérologue de *Plasmodium* a donné des résultats variables. On a noté un faible degré d'immunité croisée entre *P. falciparum* et *P. vivax* ainsi qu'entre *P. falciparum* et *P. malariae*. Une infection antérieure à *P. vivax* ou *P. falciparum* n'a pas modifié nettement l'évolution en cas de réinfection par *P. ovale*; en revanche, après une première infection à *P. ovale*, la gravité des réinfections à *P. vivax* et *P. falciparum* a été sensiblement atténuée, indice de l'existence d'une certaine immunité. Le recours aux méthodes d'immuno-fluorescence pourrait faciliter la compréhension du mécanisme de l'immunité partielle dont l'influence sur la chimiothérapie et la lutte antipaludique dans les régions d'endémie est manifeste. Si cette protection est avantageuse pour l'individu, elle implique souvent un traitement moins actif et les infections de sujets semi-immuns sont une source permanente et inapparente de réinfection de la collectivité. La chimiothérapie antipaludique ne sera réellement efficace que si l'on adopte des méthodes beaucoup plus énergiques que le traitement purement symptomatique à faibles doses.

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