# THE STATUS OF ANOPHELISM AND MALARIA IN SARDINIA DURING 1951 AND 1952<sup>1</sup>

By

# THOMAS H. G. AITKEN, JOHN MAIER AND HAROLD TRAPIDO (Received for publication April 19, 1954)

From 1946 to 1950, Sardinia was the site of an island-wide experiment aimed at attempting to eradicate the indigenous malaria vector, Anopheles l. labranchiae Falleroni. It was a joint undertaking participated in by the Italian Government and The Rockefeller Foundation, and largely financed by UNRRA and ECA funds. Responsibility for carrying out the project was delegated to a para-governmental organization known as ERLAAS (Ente Regionale per la Lotta Anti-Anofelica in Sardegna). The ERLAAS project, conceived as a result of, and patterned after, the Anopheles gambiae eradication programs in Brazil and Egypt, was planned around the use of the then new chemical, DDT. the end of 1950, with eradication still not achieved (although seemingly tantalizingly close), the decision was made to cease activities and the project officially closed on July 15, 1951.

Anopheles labranchiae was not eradicated, but this formerly dominant anopheline has been reduced to a few small scattered populations. Few insects have been subjected to an onslaught as intensive and persisting as has A. labranchiae. Evidence obtained during the summer of 1952, which is discussed in a separate paper (1), suggests that the campaign may have left a residual popu-

lation of this mosquito which differs biologically from that of pre-ERLAAS days. The early phases of the project are described in a progress report by Logan (2) and the complete story of ERLAAS is the subject of a final report by Logan et al. (3). The present paper reviews the 1950 achievements and what has occurred during the first two post-ERLAAS years (1951–1952); presentation of the entomological data will precede discussion of the malaria statistics.

### A. Entomology

### The 1950 season

By October of 1950, when the final ERLAAS scouting operations terminated, Anopheles labranchiae had been found in 151 of the 5,229 administrative sectors into which the island had been divided. (A sector comprised the amount of land which one larvieider could cover in a week; the mean size was about 4.6 sq. km.). Ninety-five per cent of the collections were in areas which had been subject to eradication treatment the previous year. The principal period of scouting had covered the 4 months of June through September. During that time an average of 1,471 men per month (range 662 to 2,037 men) were looking for adults or larvae. Scouting results were as follows:

Larval inspections 2,2	14,490
Adult inspections 1	78,359
A labranchiae larval collections	420

A. labranchiae adult collections 11

<sup>&</sup>lt;sup>1</sup> The studies and observations on which this paper is based were conducted with the support and under the auspices of the Division of Medicine and Public Health of The Rockefeller Foundation with the cooperation of the Italian Government and the Government of the Autonomous Region of Sardinia.

A. labranchiae larval specimens 1,379
A. labranchiae adult specimens 28

On the average, it had taken approximately 400 man-days of effort to find an A. labranchiae breeding place and about 1,102 man-days to identify a positive sector. Eradication zones, totaling 125 and averaging 13 sectors per zone, had been established and subjected to insecticidal action for a mean period of 10 consecutive weeks (range 4 to 21 weeks).

### The 1951 season

Two programs operated in 1951. That of the national government was sponsored by the Alto Commissiariato all-'Igiene e Sanità Pubblica and directed by the 3 provincial health officers. It was directed solely against adult mosquitoes through DDT residual spraying measures, but because of an inadequate budget, fell far short of completion. The second program, created by the Sardinian regional government and under the supervision of the Assessore all'Igiene, Sanità e Pubblica Istruzione, was a continuation of an ERLAAS-like program; it consisted of scouting and the establishment of larvicide-treatment zones (using DDT and Paris green) whenever A. labranchiae was found.

Regional scouting activities encompassed the period from May to October and involved a maximum of 380 men. During this time A. labranchiae was found in 86 sectors, over 90 per cent of which had been treated the previous year. The bulk of the collections were made in the northeastern part of the island (figure 1). Seventeen adults (Anopheles maculipennis group) were collected (all from the Villarios area in the southwest) and the numbers of larvae remained small. In 309 collections there were 1,110 larvae represented, giving a mean of about 4 larvae per collection and about 13 larvae per positive sector.

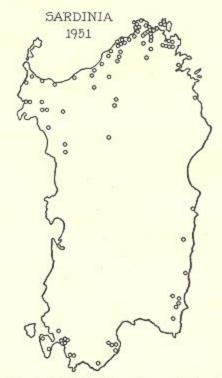


Figure 1. Location of sectors positive for Anopheles labranchiae in 1951.

Collections of single larval specimens were again encountered (there had been 22 in 1950), and 17 per cent of the positive sectors were thus represented. In 1951 it took approximately 139 mandays to locate an A. labranchiae breeding place and 498 to establish a positive sector. Thus it would appear that the species was easier to find than in the previous year. Nevertheless it was persisting in minimal numbers and in areas previously infested.

The numbers of immature stages and adults identified by species in 1951 are summarized in table 1.

### The 1952 season

The two agencies mentioned in the previous section continued to operate in 1952. However, it was only the regional organization, now called the Centro Re-

Table 1

Numbers of specimens of anophelines by species

collected in 1951

	Larva	e	Adults	
Anopheles species	Number	Per	Number	Per
claviger	221,311	67	158	84
algeriensis	103,758	32	11	6
marteri	1,833	<1	1	<1
labranchiae	1,110	<1	17*	9
hispaniola	957	<1	0	0
Totals	328,969	99	187	99

<sup>\*</sup> Anopheles maculipennis group, probably A. labranchiae.

gionale Anti-Insetti, which functioned continuously. The residual spraying program sponsored by the central government through the agency of the provincial health officers, did not get under way until late in the season and was very restricted in scope through lack of funds.

The regional organization and program was as follows. The budget was roughly \$700,000, of which about 35 per cent was earmarked for scouting, 31 per cent for larviciding and residual spraying (personnel and insecticides), 11 per cent for administrative salaries, 10 per cent for transportation and supplies, 3 per cent for rent and building maintenance, and 10 per cent for contingencies.

The island was divided into 4 zones (each under a medical officer) and 15 groups (headed by former ERLAAS personnel) (Figure 2). The former ERLAAS units of District and Sector remained within the framework of the group. Each group had a varying number of scouting squads (depending upon the suspected A. labranchiae potential of the area), the average being 7 squads, each composed of about 6 experienced men. On the basis of presumed A. la-

branchiae incidence, the countryside was divided into A areas (suspected as being positive; roughly 20 per cent of the total sectors), B areas (doubtfully positive; roughly 25 per cent of the sectors) and C areas (presumably negative; roughly 55 per cent of the sectors). Scouts in A areas were assigned on the basis of 1 per 5 sectors (said to represent a 12day cycle for a theoretical complete coverage of all possible breeding places); in B areas, 1 scout per 7 sectors (3- to 5-week cycle); and in C areas, 1 per 30 sectors (2-month cycle in valleys and one inspection only in the higher mountains). There was employed a maximum of 606 scouts, plus 124 chief scouts, giving a total of 730 scouting elements (by comparison there were 380 scouts in 1951 and 2,000 to 3,000 scouts during the ERLAAS period).

The campaign started with the DDT



FIGURE 2. Group subdivisions and location of sectors positive for Anopheles labranchiae and A. sacharovi in 1952.

residual spraying of all man-made structures and grottoes in the A areas during the period February 18 to March 31. These areas were largely along the north coast, but there were a few on the south and east coasts as well. The insecticide, a 40 per cent emulsion concentrate of DDT, was diluted to 5 per cent and applied at an estimated 2 grams of DDT per square meter.

Scouting in A areas started during the second week of April and continued until October 31, in B areas from late April to October 15, and in C areas from June through September.

Larviciding in the eradication zones was done with fuel oil containing 1 per cent Triton X-100. No DDT was included. This was apparently an economy measure. Dosage was reported to be 20 to 25 gallons per acre. In certain instances Paris green was utilized where farmers objected to the use of excessive amounts of oil in animal water holes.

A general summary of the results of the 1952 program follows. Scouting operations covered the period from April

Table 2

Man-days of scouting by 2-week periods in 
Sardinia during 1952

Two-week period	Man-days	
April 6-19	3,976	
April 20-May 3	4,801	
May 4-17	5,911	
May 18-31	5,777	
June 1-14	6,330	
June 15-28	7,656	
June 29-July 12	7,549	
July 13-26	7,800	
July 27-August 9	7,819	
August 10-23	7,335	
August 24-Sept. 6	7,894	
Sept. 7-20	5,893	
Sept. 21-Oct. 4	4,100	
Oct. 5-18	3,534	
Oct. 19-Nov. 1	2,689	
Totals	89.064	

7 to the end of October. The first personnel reductions occurred during the last 10 days of September, at a time when the rains began. In the final 7 weeks, the mean weekly scouting force was only about 45 per cent of what it had been during the previous 7 weeks (1,736 man-days as compared with 3,873 man-days per week). Man-days of scouting by 2-week periods are summarized in table 2.

All sectors in the island were inspected at least once, and 1,790 sectors (34 per cent of the total) received only one inspection. The maximum number of sector inspections was 21 (one sector only). The mean number of sector inspections for the island as a whole was 4. For the A ("dangerous") areas it was 9, but should have been closer to 15 as originally planned (only 6 per cent were so inspected); for the B ("doubtful") areas it was 5 when it should have been between 5 and 8; and for the C ("presumed negative") areas the figure was about 2, as planned.

The first A. labranchiae collection (a single first instar larva) was made April 8 and when the last specimen was found (October 14), 161 collections representing 597 larvae and pupae had been made in 64 sectors (figure 2).

A new development appeared this year with the finding of Anopheles sacharovi, another malaria vector of the A. maculipennis complex, in 11 sectors (between May 24 and July 22) representing 6 distinct localities in the northern part of the island (figure 2). Identification was based on the number of antepalmate hair branches of the fourth and fifth abdominal segments of the larvae as well as eggs recovered from the natural breeding place. Thirty-nine collections, representing 160 larvae and pupae, were made. A. sacharovi had not been found during the entire period of ERLAAS

Table 3				
Anopheles labranchiae-positive sectors and collections by months from 1950 to 1952	9.			

Month 1950†	Number of sectors			Number of collections		
	19509	1951	1952	1950	1951	1952
April	0		7	0	_	9
May	6	0	5 (1)	17	0	8 (2)
June	56	0	18 (2)	116	0	60 (17)
July	56	22	29 (8)	200	115	102 (20)
August	26	26	1	77	78	1
September	6	28	6	9	93	7
October	1	10	9	1	23	13
Totals	151	86	75 (11)	420	309	200 (39

<sup>\*</sup> Including the number of A. sacharovi indicated in parentheses.

including the initial survey of 1946, and this in the face of the fact that over a million anopheline larvae had been examined during these 6 years and a constant lookout kept for its presence. The species had been reported from the island some years previously but was apparently present at that time in very small numbers, and was restricted in its distribution (3); it is known from Corsica where it is not uncommon.

Considering then A. labranchiae and A. sacharovi together, the total number of sectors positive for known effective vector species in 1952 was 75; of these, one of the A. labranchiae sectors is counted twice as it was again positive in October after 17 weeks. All told, there were 757 larvae in the 200 collections made. These figures represent a mean of about 4 larvae per collection and about 10 per positive sector, results essentially similar to those of 1951.

Scouting results by months for the 3year period 1950-1952 are summarized in table 3 (data for 1952 include A. sacharovi). The virtual absence of August collections in 1952 is noteworthy. There was a drop of 602 man-days during the week August 10 to 16, but other than this, scouting remained at a high level through the second week of September. In general, the mean number of man-days required to establish the presence of a vector species in a sector was about 1,188 (compare with this the figures of 1,102 and 498 for 1950 and 1951, respectively). Possibly the A. labranchiae population was in reality more or less at a standstill rather than increasing as suggested by the 1951 figures. However, the lack of sufficient information hardly warrants hazarding such an opinion.

Sectors positive by scouting area were as follows:

	Positiv	e sectors
Scouting area	(number)	(per cent)
A (dangerous)	48	64
B (doubtful)	22	29
C (presumed negative)	5	7
	-	- T
Totals	75	100

The distribution of positive sectors in relation to scouting areas is of interest in evaluating the incidence of A. labranchiae. The finding of the species in areas expected to be negative is sig-

<sup>†</sup> These figures differ from those reported by Logan et al. (3) as sectors repeatedly positive are excluded.

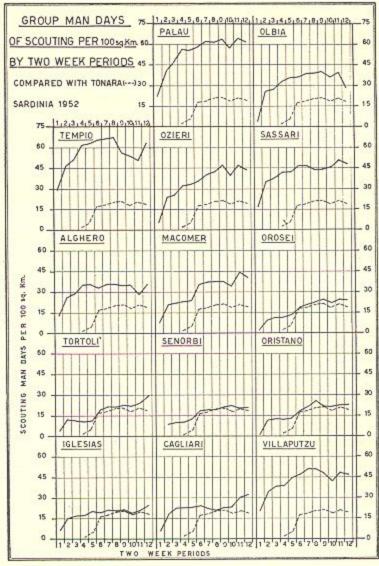


FIGURE 3. Relative biweekly scouting strengths of Sardinian groups in 1952.

nificant when one considers the small number of inspections made in these areas. We know from experience that only through repeated inspections may one hope to uncover the extent of A. labranchiae breeding when the species' numbers have been reduced to the present low level. For example, in 1950 about 50 per cent of the 151 A. labranchiae-positive sectors were found during the first two inspections and 78

per cent in 3 inspections; the remaining 22 per cent required from 4 to 9 visits before A. labranchiae was found (this with a scouting force of close to 2,000 men). The difficulties associated with this type of scouting are more fully discussed by Trapido (4).

The bias in the 1952 scouting effort, as indicated in the discussion regarding the assignment of scouts, takes on significance when scouting output is ana-

Table 4

Scouting intensity by group related to frequency of Anopheles labranchiae- and A. sacharovipositive sectors in 1952

Groups probably adequately scouted	Number of posi- tive sectors	Groups inade- quately scouted	Number of posi- tive sectors
1. Palau	8	9. Cagliari	12
2. Tempio	5	10. Iglesias	12
3. Villaputzu	11	11. Tortolì	1
4. Sassari	3	12. Orosei	5
5. Ozieri	0	13. Oristano	0
6. Macomer	6	14. Senorbì	0
7. Alghero	12	15. Tonara	0
8. Olbia	0		-

lyzed by regional group. The indices obtained relate area to man-days of scouting by 2-week periods. These are shown in figure 3, with Tonara, a mountain group, with the smallest scouting force, used as a baseline for comparison; the period covered is from April 7 to September 13. The groups are arranged in order from north to south. It will be noted that, with the exception of Villaputzu, all the southern groups had comparatively weak scouting forces.

Considering the figure of 30 man-days of scouting per 100 square kilometers per 2 weeks as the very minimum force necessary to be reasonably effective (this means about one man per 3 square kilometers per day), and comparing scouting intensity with A. labranchiae positives in the various groups, one gets a clearer picture of the actual status of the species in a particular group. This has been done in table 4, where groups are listed in decreasing order of scouting intensity with the total number of A. labranchiae and A. sacharovi sectors in each. Groups below Olbia are considered to have had inadequate scouting (less than 30 man-days per 100 square kilometers per 2-week periods). table shows that A. labranchiae was easier to find in Cagliari and Iglesias than in the northern groups (possibly because of greater anti-anopheline activity in the north in 1951), and that the negative findings in Oristano, Senorbì, and Tonara are particularly open to question. It should be noted, however, that within these 3 groups the typical A. labranchiae habitats were subjected to intensive scouting.

The actual numbers of immature stages of A. labranchiae found during the 2 years are tabulated in table 5 (A. sacharovi were included in 1952). The high incidence of early instars in the collections is noteworthy.

The experience of collections represented by single immature specimens, which was one of the rather extraordinary aspects of the final stages of ERLAAS, continued to manifest itself in 1952. Of 75 sectors with either A. labranchiae or A. sacharovi present, no less than 22 (29 per cent of the total) were represented by collections of individual larvae. The frequency distribution of positive sectors by numbers of immature stages collected in 1951 and 1952 is summarized in table 6 (A. sacharovi included in 1952). The difficulties

Table 5

Numbers of larvae and pupae of Anopheles
labranchiae and percentage of total stages
in 1951 and 1952\*

Stage	Number		Per cent	
Stage	1951	1952	1951	1952
1st instar	281	94	25	13
2nd instar	213	184	19	25
3rd instar	277	239	25	30
4th instar	313	227	28	30
Pupa	26	13	2	2
Totals	1,110	757	99	100

<sup>\* 1952</sup> includes A. sacharovi.

inherent in larval scouting under present Sardinian conditions are demonstrated by the fact that 56 and 62 per cent of the positive sectors in 1951 and 1952, respectively, were based on collections of 5 larvae or less. The significance of these figures increases when one considers that, following laboratory identification, every A. labranchiae breeding place as well as all others in the area were immediately rechecked, prior to larviciding, to determine the extent of the infestation.

While the distribution of known A. labranchiae foci in 1952 followed the pattern of former years (i.e., reappearance in the same areas), there was a shift in the concentration of positive areas this year as demonstrated in table 7 (see also figures 1 and 2). While in 1950 and 1951 three-fourths or more of the foci were in the north (with the bulk of these in the northeast), they were about evenly distributed between the north and south in 1952.

With the exception of two sectors in

Table 6

Frequency distribution of sectors positive for Anopheles labranchiae by numbers of larvae and pupae collected in 1951 and 1952\*

Numbers of larvae and	Number o	of sectors	Per cent of total positive sectors		
pupae	19511	1952	1951	1952	
1	14	22	17	29	
2-5	33	25	39	33	
6-10	12	11	14	15	
11-20	12	9	14	12	
21-50	8	.6	10	8	
51-100	3	1	4	1	
101-150	2	1	2	1	
Totals	84	75	100	99	

<sup>\* 1952</sup> includes A. sacharovi.

Table 7

Distribution of A. labranchiae foci in the 3 years,
1950-1952

Region of island	Percentage of positive sectors			
region of island	1950	1951	1952	
Northeast	58	51	27	
Northwest	25	23	25	
Southeast	10	9	17	
Southwest	7	16	31	
Totals	100	99	100	

the upper Tirso valley of the north-central part of the island, the 1952 positives were again largely near the coast. The median distance of foci from the sea was 2.6 kilometers in 1951 (range 0.005 to 41.8 kilometers) and 2.7 kilometers in 1952 (range 0.002 to 48 kilometers). A. labranchiae generally was found at lower altitudes in 1952, the median elevation of foci being 19 meters (range 0.2 to 300 meters), as compared with 26 meters in 1951 (range 0.1 to 380 meters).

The spatial relationship of one A. labranchiae breeding place with another is of interest in showing the degree of infestation of a particular area. 1951, the median distance between breeding places was 2.2 kilometers (range 0.1 to 30.8 kilometers) and to the next nearest breeding place 5 kilometers (range 0.9 to 52 kilometers). In 1952 these figures for the first 63 positives were 4 (range 0.006 to 25 kilometers) and 8.7 kilometers (range 0.9 to 47.6 kilometers), respectively. The median distance of the 1952 foci (first 63 positives only) to those of the previous year was 5.6 kilometers (range 0.01 to 31 kilometers). These data are unavailable for the remaining 12 positives of 1952 and for 1951.

In pre-ERLAAS days, A. labranchiae was more or less uniformly distributed

<sup>†</sup> Two additional sectors positive for 3 adults only.

Table 8

Distances of known Anopheles labranchiae breeding places from nearest population centers,
Sardinia (1949–1952)

Year	2	Mean distance (km)	Maximum distance (km)
1949		4	12
1950		4	12
1951		4	12
1952*		5	20

<sup>\* 1952</sup> includes A. sacharovi.

throughout the island, breeding places occurring both near and within population centers as well as long distances away from them. With the progress of the campaign and the passage of time, the species has virtually disappeared from the environs of villages and towns, suggesting that the residual population may be essentially nondomestic. Data for the past 4 years showing the distances of known breeding places from the nearest population center are summarized in table 8; those for 1952 refer to the first 63 positive sectors only and include A. sacharovi.

The numbers of anopheline larvae collected in 1952 are summarized according to species in table 9. A. claviger and A. algeriensis continued to dominate the

Table 9

Numbers and percentages of anopheline larvae by species collected in 1952 and compared with previous 2 years

Anopheles species	Number	Yearly percentage		
	of larvae in 1952	1950	1951	1952
claviger	442,039	93	67	57
algeriensis	311,738	5	32	40
marteri	13,158	1	<1	2
hispaniola	7,203	<1	<1	<1
labranchiae	597	<1	<1	<1
sacharovi	160	0	0	<1
plumbeus	29	<1	0	<1
Totals	774,924	99	99	99

collections as they have since 1949. The rapid increase of the latter species during the past 2 years suggests that it was invading habitats previously occupied by A. labranchiae but doing so at a slower rate than A. claviger. In fact, many rivers (such as the Tirso, Temo, Taloro, Araxisi, Flumini Mannu, etc.) which formerly were principally supporting populations of A. labranchiae are now producing large numbers of A. claviger and A. algeriensis. In the 4 groups of Cagliari, Iglesias, Alghero, and Sassari (and to a lesser extent Oristano). A. algeriensis in 1952 was practically always by far the most abundant anopheline collected (a complete reversal of the experience of former years). These are all low-lying coastal areas characterized by marshes and sluggish rivers.

With regard to adults, 94 A. claviger and 63 A. algeriensis were collected by regional scouts in 1952. They could find no A. labranchiae or A. sacharovi; however, one A. labranchiae adult was found by us during the course of a special investigation in the Geremeas valley (1). The absence of adult collections of these two vector species may reflect in part

Table 10

Numbers of larval anophelines (all species) collected in relation to man-days of scouting in Sardinia, 1946–1952

Year		Man-	Number of larvae				
	Period .	days of scouting	Collected	Per man- day			
1946	May 1-Aug. 31	231	20,730	89.74			
1947	May 1-Aug. 31	9,309	88,669	9.52			
1948	Jan. 1-Dec. 31	176,117	84,738	0.48			
1949	Jan. 1-Oct. 31	406,592	99,338	0.24			
1950	Mar. 1-Sept. 30	166,486	701,450	4.21			
1951	May 7-Oct. 20	42,866	328,969	7.67			
1952	Apr. 7-Oct. 31	89,064	774,924	8.70			

the lack of attention given to this phase of scouting. On the other hand, in the known positive area of Geremeas, where intensive searches for adults by various techniques (shelter inspections, nocturnal observations in pigsties and with Shannon trap, vegetation and rodent burrow smoking) were conducted by us on a routine daily basis, only one adult was recovered.

Further light is thrown on the effect of the ERLAAS project on the insular anopheline population by comparing the total numbers of larvae collected (all species) to the amount of work expended in the effort. Table 10 summarizes the data for the 7 years 1946 through 1952. During the initial survey of 1946, the capture rate was about 90 larvae per man-day of scouting. This figure rapidly diminished as eradication operations got under way. The year 1948 was the period of island-wide larviciding measures, but the full impact of this work was not apparent until 1949 when a figure of 0.24 larvae per man-day was obtained.

The following year, after the lifting of island-wide larviciding measures, there was a 20-fold increase in larval densities. Subsequently, this increase has continued but at a much lower rate, suggesting that larval populations of species such as A. claviger, in the absence of continued attrition, were reaching stabilization. The high larval densities in the early years refer mainly to A. labranchiae, and the fact that the figure in 1952 is only one-tenth that of 1946 seems to be reasonable evidence of the pre-eminent position of this species in the pre-ERLAAS days.

#### B. MALARIA

## The 1950 season

The status of malaria in Sardinia in 1950 has been described in the report of the ERLAAS campaign (3). All malaria indices continued the progressive decline which had been observed since control operations began. The spleen rate in 15,509 school children (aged 6 to 13 years) in 66 villages in all parts of the island, was measured in the period from November, 1949, to March, 1950, and was 13.2 per cent. The parasite rate in this group was 0.32 per cent. These were the lowest rates recorded in 3 annual surveys of these villages.

A smaller survey was carried out in 13 of these villages, in May, 1950, and again in September, involving 2,863 and 2,282 children, respectively. The spleen rates at these times were 17.8 per cent and 14.4 per cent; the parasite rates were 0.84 per cent and 0.66 per cent. Blood films were examined from 938 infants (aged 2 months to 2 years) in the 13 villages; all were negative for plasmodia.

Four cases of malaria, classified as primary on the basis of the history, were reported during 1950 in the official communicable disease notifications of the provincial health officers in Sardinia. These cases were investigated by the ERLAAS epidemiological service. Two were considered to be transfusion malaria, one was probably not malaria, and one was undoubted vivax malaria, perhaps primary. Forty further cases were reported from the island as relapses of previously acquired malaria.

#### The 1951 season

Malaria continued to regress in 1951.

The communicable disease reports of the provincial health officers for the calendar year showed a total of 6 cases of malaria classified as relapses, and 3 cases classified as primary.

The 3 primary cases were investigated, and the following information obtained: Case 1. Itireddu, Sassari province. A child of 7 years who developed quartan malaria (Plasmodium malariae) a few days after receiving two transfusions of paternal blood for the treatment of acute favism. The donor's blood was negative when examined by us about 1 month later. However, this case in all probability was transfusion malaria.

Case 2. Serrenti, Cagliari province. A child of 3 years with quartan malaria (P. malariae) who was said not to have had malaria previously. Conditions are unfavorable for A. labranchiae development and malaria transmission in this area, which is extremely dry during the summer months. The entire region was negative for A. labranchiae in 1951 and 1950; the nearest collection of larvae in 1949 was from a habitat 12 kilometers from the village where the child lived.

Case 3. Sa Castanza, Budduso commune, Sassari province. A child of 18 months alleged to have falciparum malaria. The child became ill in July, and after several days of fever was taken to a physician who diagnosed malaria, which was called primary in the absence of any previous history. A blood smear was taken before treatment, and was examined at the Olbia malaria dispensary, where "a few" P. falciparum were found. This film was not available for examination by us. A second smear made in September (after treatment) was examined by us; a single gametocyte of P. falciparum was found after prolonged search. The valley in which the hamlet is located was intensively scouted for A. labranchiae after this case was reported; neither larvae nor adults were found.

Spleen and parasite surveys were made, in August and September, on school children from 6 to 14 years of age. In addition, blood smears were made on infants under 24 months. Since it was impossible in the time available to examine all 66 of the villages in the ERLAAS surveys, the 1951 survey was limited to those which had a spleen rate of over 25 per cent in the first ERLAAS survey (winter of 1947-1948); there were 25 such villages. Two others were also included which had spleen rates of less than 25 per cent in the first ERLAAS survey, but where children positive for P. falciparum were found in 1950. These villages were Buggeru (spleen rate 12 per cent in the first survey) and Sant'Anna Arresi (spleen rate 23 per cent in the first survey). Other villages where P. falciparum-positive children were found in 1950, had spleen rates greater than 25 per cent in the first survey, and were thus included in the 1951 survey. This method of se-



FIGURE 4. The 66 Sardinian malaria survey villages. Only those underlined were visited in 1951 and 1952.

lection gave a fairly good geographical coverage of the island, as shown by figure 4.

The data obtained from the surveys are shown in table 11. The overall spleen rate was 13.79 per cent and the parasite rate 0.16 per cent, which compare with rates of 20.53 per cent and 0.70 per cent obtained in the same 27 villages during the preceding winter survev of 1949-1950. Seven plasmodiapositive blood smears were found; of these, 2 were P. vivax, 2 P. falciparum and 3 P. malariae. The two children with P. falciparum had not been examined in previous surveys; they were not ill when examined on this occasion, and had not been under treatment for malaria. Thus, the 1951 spleen rate was approximately one-third lower than the previous rate; the average spleen size also decreased, while the average enlarged spleen size increased slightly. The change in the parasite rate reflected a decrease in the number of positive slides from 36 to 7. Parasite carriers were found in 4 of the 27 villages in 1951, as compared to 16 "carrier" villages in the previous survey. Positives for P. falciparum decreased from 10 (including 4 mixed infections with P. vivax) to 2.

Blood examinations in 815 infants (under 2 years of age) in the 27 villages, were all negative for plasmodia.

In summary, the malaria experience of the island indicates that no significant transmission took place in 1951. The 18-month-old child in the hamlet of Sa Castanza must be regarded as having had a primary infection in either 1950 or 1951. It is difficult to explain the occurrence of an isolated primary case such as this, in view of all the other evidence of a virtual absence of malaria transmission, and in view of the apparent absence of A. labranchiae in this

area. An alternate explanation of this apparent transmission might be that A. claviger was in this case the vector. This species is a vector under certain local conditions in the Near East, but has not been considered as such in Sardinia where the adults are sylvatic in habit.

#### The 1952 season

No cases of malaria, either primary or relapse, were reported in the official communicable disease notifications during the year. While case reporting is subject to many inaccuracies, nevertheless the fear of the return of malaria in Sardinia has led to a degree of vigilance against malaria on the part of physicians and public health authorities which lends significance to this absence of cases.

Malaria surveys were repeated in the same 27 villages used in 1951; spleen and blood examinations were carried out on 4,369 school children. The spleen rate showed an increase to 18.43 per cent, but the parasite rate declined further to 0.09 per cent, only 4 carriers having been found. In view of the absence of reported malaria and of the fall in the parasite rate, it seems improbable that the increased spleen rate is attributable to malaria. Unfortunately, the trained malariologist who had conducted all the previous spleen surveys, including those made under ERLAAS, was not available in 1952. He was replaced by an untrained physician, without experience in this kind of work, who was not supervised sufficiently. It is therefore considered that the increase in the spleen rate is in all probability an artifact. Evidence for this interpretation exists in the fact that a large part of the increase was accounted for by spleens of size 1 in the Hackett classification (palpable only on deep inspiration), which

are the most difficult to palpate correctly. The average enlarged spleen size actually decreased. On the other hand, the thick smear blood examinations can be considered to be reliable, since they were made by the same competent and conscientious technician who performed all the previous examinations beginning with the ERLAAS winter survey of 1948-1949.

Of interest is the finding of one smear positive for P. falciparum. This was from a child of 12 years in the village of Lotzorai, who had been examined in 7 of the ERLAAS surveys. shown an enlarged spleen on each previous examination (size 2 on 3 occasions and size 3 on 4). Parasites had not been found until the two most recent ERLAAS surveys, in May and September, 1950, when P. falciparum trophozoites were present. He had not been examined in 1951. At the time of the 1952 survey, he had not been ill and had not received treatment for malaria. It is therefore considered probable that this represents a parasite relapse of a previously acquired infection. The evidence indicating that in Sardinia P. falciparum may, in the absence of treatment, persist in the host for longer periods than generally supposed, has been reviewed in the ERLAAS report (3).

Blood examinations on 1,001 infants in the 27 villages were again all negative.

It is of interest that the northeast region of the island, which included 51 per cent of the A. labranchiae-positive sectors in 1951, 27 per cent of those positive in 1952, and which also accounted for much of the infested area in 1949-1950, nevertheless has not been one of the highly malarious regions of the island, either historically or in the ERLAAS surveys. Six villages in this general area were routinely surveyed by ERLAAS; only one of these, Loiri, had a high spleen rate in the first ERLAAS survey in 1948. (Loiri is at the southeastern limit of the area, near the Baronia region, a coastal region which was highly malarious.) Table 12 shows the complete survey records of these villages, and indicates the low initial spleen rates in all except Loiri, the decline in the spleen rate in Loiri, the disappearance of Plasmodium falciparum from all villages, and the decline in the parasite rate to zero in 5 of the 6. This may be further evidence for the belief held by Trapido and Aitken (1) that A. labranchiae in Sardinia possessed the full genetic complement of the species, and included nondomestic as well as domestic strains.

Table 11

Results of malaria surveys on school children in 27 Sardinian villages,
in 1949–1950, 1951 and 1952

Date of survey	Num- ber ex- amined	Spleen size (Hackett)					)	Num- ber en- larged	Spleen rate (per cent)	Average spleen size	Average enlarged spleen	Number of positive smears by species*			5	Num- ber positive smears	Para site rate (per
		0	1	2	3	4	5	spleens	centi	0		v	F	М	VF		cent)
Dec. 1949-	5,140	4,085	839	181	32	2	1	1,055	20.53	0.25	1.24	19	6	7	4	36	0.70
Feb. 1950 Aug. 1951-	4,408	3,800	472	105	28	2	1	608	13.79	0.18	1.28	2	2	3	0	7	0.16
Sept. 1951 lug. 1952- Sept. 1952	4,369	3,564	634	151	17	1	2	805	18.43	0.23	1.24	1	1	2	0	4	0.09

<sup>\*</sup> V = P. vivax; F = P. falciparum; M = P. malariae; VF = P. vivax and P. falciparum.

Table 12

Malaria survey records of 6 villages of northeastern Sardinia, an area where Anopheles labranchiae has persisted

Village	Date of survey	Number chil- dren examined	Spleen rate (per cent)	Parasite rate (per cent)	Number of positive amears by species*
Arzachena	Nov. 1947	182	13.8	4.4	6V, 2F
	Nov. 1948	165	15:2	0.6	1V
	Dec. 1949	188	16.6	0.5	1V
Luogosanto	Nov. 1947	137	13.2	0.0	1 8
	Nov. 1948	151	8.8	0.0	
	Dec. 1949	129	17.5	0.0	
Luras	Nov. 1947	298	6.7	1.7	2V, 3F
	Nov. 1948	282	3.9	0.0	200000000000000000000000000000000000000
	Dec. 1949	222	9.5	0.0	
Berchidda	Nov. 1947	321	12.7	1.3	4V
	Dec. 1948	325	12.8	0.3	1F
	Dec. 1949	280	11.5	0.0	
Laerru	Dec. 1947	202	7.5	6.5	11V, 2F
	Nov. 1948	217	15.5	0.9	1V
	Feb. 1950	162	14.2	0.0	
Loiri	Feb. 1948	39	35.8	7.7	3V
	May 1948	31	51.6	0.0	The second
	Sept. 1948	57	59.7	3.5	1V, 1F
	Dec. 1948	57	40.4	0.0	
	May 1949	41	58.5	0.0	12224
	Nov. 1949	53	37.7	1.9	1V
	Feb. 1950	59	33.9	1.7	1M
	May 1950	69	31.9	1.5	1F
	Sept. 1950	38	31.6	2.6	1F
	Aug. 1951	42	22.9	0.0	
	Sept. 1952	49	12.2	0.0	

\* V = P. vivax; F = P. falciparum; M = P. malariae.

#### SUMMARY

- Field activities of the Anopheles labranchiae eradication project in Sardinia were terminated at the end of 1950. While close, the goal of eradication was not achieved.
- During 1951 and 1952 a similar but more modest program with the same objective was continued by the autonomous regional government of Sardinia. At the same time the central government carried on some DDT residual spraying activities.
- 3. There has been no rapid build-up in the residual population of A. labran-chiae remaining at the end of 1950. The species continues to exist in a few scattered areas and the population appears to be more or less at a standstill, although scouting bias may not warrant these assumptions.
- 4. With a maximum scouting force of 380 men in 1951, A. labranchiae was found in 86 of the 5,229 administrative sectors into which the island is divided. Localities were largely coastal and

mainly in the northeast. Approximately 498 man-days were required to establish a positive A. labranchiae sector. A total of 1,110 A. labranchiae larvae and 17 adults (Anopheles maculipennis group) were collected between May 7 and October 20.

- 5. An analysis of the intensity of scouting (i.e., man-days related to area) in 1952 indicates that it was inadequate in 7 of the 15 administrative groups into which the island was divided.
- 6. In 1952, a maximum scouting force of 730 men found A. labranchiae in 64 sectors. Anopheles sacharovi was encountered (for the first time since 1940) in 11 additional sectors. Localities were still coastal, but were more evenly distributed between the north and south. The tendency for A. labranchiae to be found most frequently at some distance from towns and villages, a characteristic since 1949, has persisted. A. sacharovi was restricted to the north. Approximately 1,188 man-days of scouting effort were required to discover a sector positive for these species. A total of 597 larvae and one adult of A. labranchiae and 160 larvae of A. sacharovi were collected between April 7 and October 31.
- 7. Other species present and their relative proportions in 1952 include Anopheles claviger (57 per cent), A. algeriensis (40 per cent), A. marteri (< 1 per cent), A. hispaniola (< 1 per cent) and A. plumbeus (< 1 per cent). The first two species, particularly, were very abundant and all (except A. plumbeus) have invaded habitats formerly occupied by A. labranchiae.
- In 1951, DDT larvicides were employed; in 1952 fuel-oil larvicides containing spreading agent only were used, but at much higher dosages.
- Nine cases of malaria were officially reported by the government in 1951; 6 were classified as relapses and

3 as primary cases. Of the latter, one was transfusion malaria (*Plasmodium malariae*), the second may have been an old infection (*P. malariae*), and the third case (in an 18-month-old child) probably represents a primary infection due to *P. falciparum*.

- 10. Fall spleen and parasite surveys in 4,408 children (6 to 14 years of age) in 27 villages yielded rates of 13.79 per cent and 0.16 per cent, respectively. The average spleen size (classification of Hackett) was 0.18 and the average enlarged spleen was 1.28. Seven positive smears were found: 2 P. vivax, 2 P. falciparum and 3 P. malariae. Blood examinations of 815 infants under 2 years of age were negative.
- No cases of malaria were officially reported in 1952.

12. The 27-village survey of 1952 in 4,369 school children disclosed a spleen rate of 18.43 per cent and a parasite rate of 0.09 per cent. The average spleen size was 0.23 and the average enlarged spleen was 1.24. Four positive smears represented one vivax, one falciparum and two malariae infections. Blood examinations of 1,001 infants were negative

#### REFERENCES

- Trapido, H., and Aitken, T. H. G. Study of a residual population of Anopheles l. labranchiae Falleroni in the Geremeas Valley, Sardinia. Amer. Jour. Trop. Med. and Hyg., 1953, 2: 658-676.
- Logan, J. A. Anopheles labranchiae eradication in Sardinia: An interim report. Amer. Jour. Trop. Med., 1950, 30: 313– 323.
- Logan, J. A. with the collaboration of Aitken, T. H. G., Casini, G. U., Knipe, F. W., Maier, J., and Patterson, A. J. The Sardinian Project: An Experiment in the Eradication of an Indigenous Malarious Vector. Amer. Jour. Hyg. Monograph Series, No. 20. The Johns Hopkins Press, Baltimore, Md., 1953.
- Trapido, H. Factors influencing the search for anopheline larvae in Sardinia. Jour. Nat. Malaria Soc., 1951, 10: 318-326.