OBSERVATIONS ON THE MORPHOLOGY AND BIOLOGY OF THE SUBSPECIES OF *ANOPHELES PUNCTULATUS* DÖNITZ.

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(Three Text-figures and one Map.)

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Introduction.

The interpretation of the various forms occurring within the species *A. punctulatus* Dönitz has been, and still is, the subject of considerable discussion.

In a recent paper D. J. Lee and the author (Lee and Woodhill, 1944), on geographical and ecological evidence, substantiated two subspecies, *A. punctulatus punctulatus* Dön. and *A. punctulatus moluccensis* Swell, Swell de Graf., and put forward the theory that forms which were intermediate between these two were actually hybrids, which occurred where the subspecies existed together. At the same time it was pointed out that if the form with the dark proboscis, occurring in the New Hebrides, which was described by Laveran in 1902 as *A. farauti*, could not be differentiated from *A. punctulatus moluccensis* from Australia and New Guinea, then the former name would have priority and should be adopted. At the time of publication no detailed descriptions of the larvae of the New Hebridian and Solomon Islands forms were available, but recent work by Belkin, Knight and Rozeboom (1945) has provided the necessary information, since their description of *A. farauti* shows no significant difference from *A. punctulatus moluccensis*. However, the arguments previously used in relation to the subspecific status of this form still hold, and it is therefore proposed to adopt the name *A. punctulatus farauti* Laveran rather than the specific designation *A. farauti* Laveran as used by Belkin, Knight and Rozeboom (1945).

The following paper gives the results of some detailed observations at Salamaua during May-July, 1944, and also the results of re-examination of larval material from Australia and New Guinea.

Morphology of the Subspecies.

In the above-mentioned publication (Lee and Woodhill, 1944) the subspecies are differentiated as follows:

A. punctulatus punctulatus.

Adult.—The proboscis has the apical two-fifths pale scaled.

Larva.—The outer anterior clypeal hairs of the larva are simple; the posterior clypeals are simple and 0·2 to 0·5 as long as the inners; the antennae are frequently pale with dark tips. The inner and middle shoulder hairs are transparent with normally elongated shafts, and arise from small weakly pigmented tubercles which are not joined to form a single chitinous plate.

A. punctulatus farauti.

Adult.—The adult is not known to be separable from the subspecies *A. punctulatus punctulatus* except by the proboscis being entirely dark scaled.

Larva.—The outer anterior clypeals of the larva are simple or weakly branched; the posterior clypeals may be simple or have up to three or four branches; they vary from 0·1 to 0·5 the length of the inners; the antennae are usually concolorous. Characteris-

* The work embodied in this paper was carried out while the author was serving in the Australian Army Medical Corps.

tically the inner and centre shoulder hairs are heavily pigmented, the shaft of the inner hair is flattened, and the basal tubercles are heavily pigmented and either enlarged or joined to form a single chitinous plate. The appearance of the shoulder hairs, and particularly their basal tubercles, is the most reliable character for separating this subspecies from *A. punctulatus punctulatus*.

Since the above was published a further examination of larvae from Australia and various parts of New Guinea has shown that there are also differences of some value in the palmate hair of the first abdominal segment. These differences, which were originally noted by Lieutenant-Colonel W. V. King, are not constant for the whole range of the species, but hold in certain areas. In *A. punctulatus punctulatus* from Milne Bay, Koitaki, Lae, Salamaua and the Solomon Islands, the first abdominal palmate hair is weakly developed with from six to thirteen slender non-flattened tapering branches, i.e., not a typical palmate hair. In *A. punctulatus farauti* from Cairns, Merauke, Moresby, Milne Bay, Goodenough Island, Salamaua and the Solomon Islands, the first abdominal palmate hair is strongly developed with from twelve to sixteen flattened leaflets.

In the Northern Territory, however, *A. punctulatus farauti* has this hair weakly developed as in *A. punctulatus punctulatus* (see Fig. 3*e*).

With regard to the antennal colouration of the larvae, as described above, this is useful in differentiating the subspecies in certain localities, e.g., Milne Bay, but does not always hold.

At Salamaua one breeding place was found in which the larvae showed all variations in antennal colouration, but had typical *punctulatus* shoulder hairs; some 350 adults were bred out from this series and these were all *A. punctulatus punctulatus*. The breeding place was unusual for this subspecies, consisting of a weedy partly shaded grass-grown pool, with the larvae occurring under floating grass, and thus resembled the breeding places of *A. punctulatus farauti*. On the other hand, all the *A. punctulatus punctulatus* larvae found in sunlit muddy pools lacking all macroscopic vegetation, had pale antennae with dark tips. This would suggest that the antennal colouration is an environmental character, but definite experiments would be necessary to substantiate this.

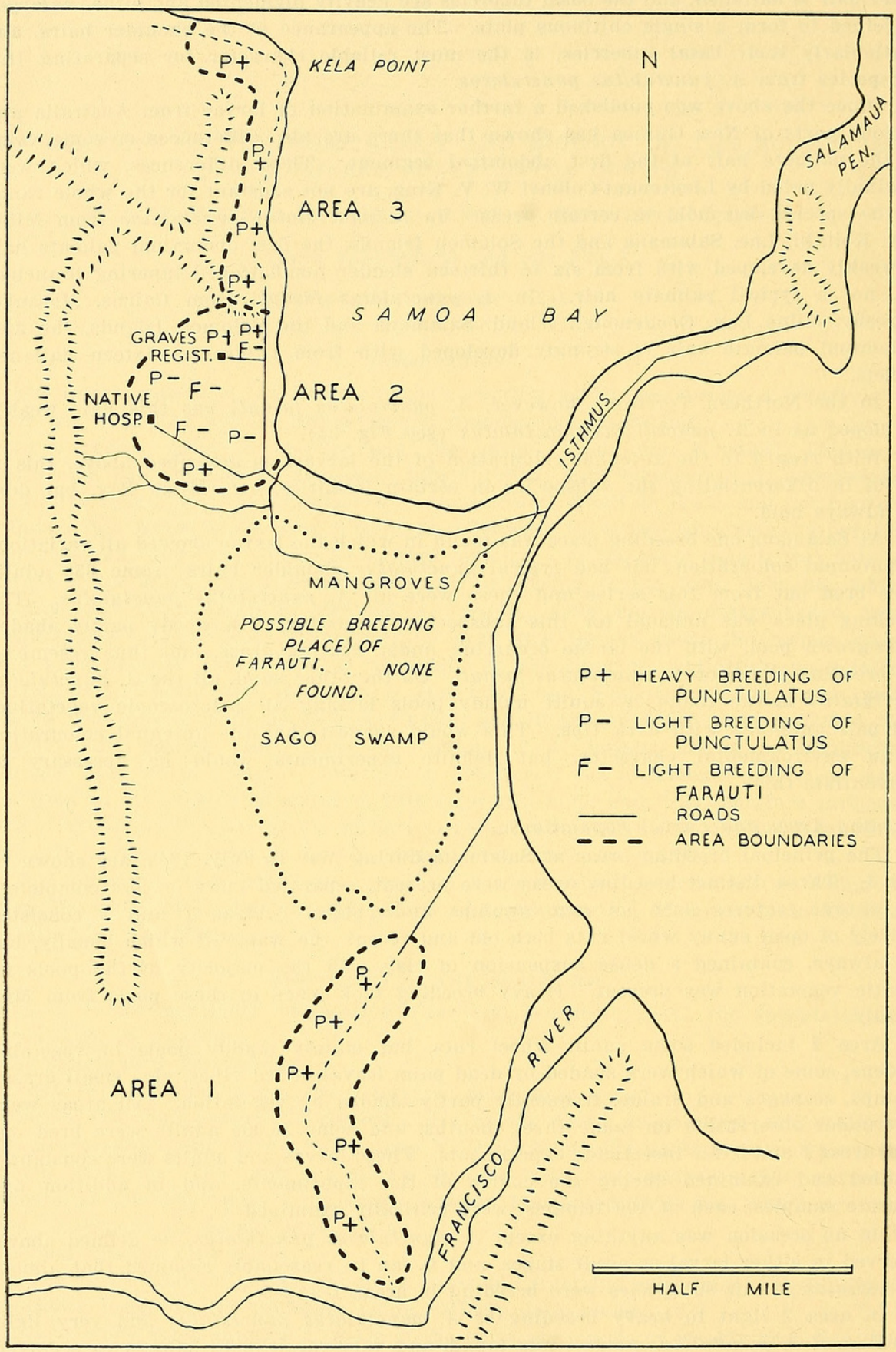
Breeding Areas and Larval Populations.

The principal breeding areas at Salamaua during May to July, 1944, are shown in Map 1. Three distinct breeding areas were present, separated more or less completely by natural features such as sago swamps and ridges. Areas 1 and 3 consisted entirely of open sunny wheel ruts both old and recent, the water of which usually, but not always, contained a dense suspension of clay. In the majority of the pools no aquatic vegetation was present. Heavy breeding took place in these pools from May to July.

Area 2 included some sunlit wheel ruts, but mainly muddy pools in vegetable gardens, some of which were shaded by dead palm leaves, dried vines, etc., small grassy swamps, seepages and drains, frequently partly shaded by vegetation. All areas were kept under observation for some three months, and some 15,000 adults were bred out from areas 1 and 3, for insecticide experiments. These larvae and adults were constantly handled and examined during the course of the experiments, and in addition five separate samples, each of 100 females, were critically examined.

On no occasion was anything except *A. punctulatus punctulatus*, as defined above, observed in either larval or adult stages, and it can be reasonably assumed that almost pure strains of this subspecies were breeding in areas 1 and 3.

In area 2 light to heavy breeding of *A. punctulatus punctulatus* and very light breeding of *A. punctulatus farauti* was found, and very rarely both subspecies occurred in the same pool. For reasons stated below, however, it appears obvious that fairly extensive scattered breeding of *A. punctulatus farauti* must have been taking place somewhere adjacent to this area, although only very small numbers of larvae could be located.



Map 1.—Breeding areas of *A. punctulatus punctulatus* and *A. punctulatus farauti* at Salamaua.

Estimation of Adult Population.

A series of night catches was made in the three areas, but in areas 1 and 3 practically no adults were obtained, although the catches were made alongside heavy breeding places. In area 2, however, a number of very successful night catches were made, the numbers depending on the weather conditions, and possibly other undetermined factors. Although the temperature at Salamaua at 10 p.m. only varied between 76° and 82°F., with an average of 78°F., and the humidity between 89% and 96%, with an average of 92%, a slight drop in temperature, accompanied by a slight breeze, appeared to prevent any movement of adults and to reduce the catches considerably.

The inability to take adults at night in areas 1 and 3, as compared with area 2, may possibly be explained by the fact (*a*) that there were no human communities in areas 1 and 3 and the adults may have dispersed over a wide area on emergence, whereas in area 2 the catches were made in or adjacent to permanent living quarters of natives and army personnel, or (*b*) that catches in area 2 were made not only from the skin when feeding, but from resting surfaces such as walls of huts and tents which were illuminated by a dim light, whereas in areas 1 and 3 the catches were made in the open.

It is to be noted that approximately 50% of the females taken in area 2 were resting on walls, etc., and not biting.

As in many other localities, no adults could be found resting in tents or huts during the day time.*

The number of adult females taken in night catches in area 2 between 26th May, 1944, and 4th July, 1944, and determined as *A. p. punctulatus*, *A. p. farauti* and intermediates are shown in Table 1.

TABLE 1.

Date.	Number Catching and Time.	Total.	*A. p. far.*	*A. p. punct.*	Inter.
26. v.44 ..	1 native, 8 p.m. to 10 p.m.	39	29	5	5
31. v.44 ..	1 native, 8 p.m. to 10 p.m.	38	16	11	11
6. vi.44 ..	1 native, 8 p.m. to 10 p.m.	44	21	13	10
13. vi.44 ..	1 native, 8 p.m. to 10 p.m.	11	6	3	2
13. vi.44 ..	4 men, 8 p.m. to 6 a.m.	266	177	60	29
15. vi.44 ..	2 men, 10 p.m. to 2 a.m.	28	19	5	4
23. vi.44 to 27. vi.44 ..	4 men, 8 p.m. to 10 p.m.*	48	33	4	11
28. vi.44 ..	4 men, 8 p.m. to 6 a.m.	401	175	143	83
1.vii.44 ..	4 men, 10 p.m. to 12 midnight* ..	15	8	4	3
4.vii.44 ..	4 men, 11 p.m. to 2 a.m.	87	47	14	26
Total		977	531	262	184
Per cent.		100	54·3	26·8	18·9

* Bad weather.

It is significant that although light to heavy breeding of *A. p. punctulatus* was taking place in area 2, while only very light breeding of *A. p. farauti* could be recorded, nevertheless 54% of adults were *A. p. farauti* as compared with 26% *A. p. punctulatus* and 18% intermediates. Intermediates were of all types from those with only a small ventral pale patch on the proboscis to those with a considerable amount of pale scaling (see Figs. 1, 1*b*, 1*c* and 1*d*). The hourly figures from the two all-night catches are shown in Tables 2 and 3.

* It should be noted, however, that the resting habits of both subspecies vary considerably in different localities and circumstances. At Nadzab in July, 1944, large numbers of fed adults were found resting throughout the day on the walls of native huts. The huts were surrounded by jungle with some open patches of kunai grass, with a stream, including weedy backwaters, close by. Of 129 adults collected, 85% were *A. punctulatus farauti*, 3% *A. punctulatus punctulatus* and 12% intermediates.

Lieutenant Brannigan (unpublished data) has also reported large numbers of adults at Dumpu resting in tents and huts during the day.

TABLE 2.
All-Night Catches by Four Men, 13th June, 1944.

Time.	Total.	*A. p. farauti.*	*A. p. punctulatus.*	Intermediates.
8 p.m. to 9 p.m.	9	8	—	1
9 p.m. to 10 p.m.	10	8	1	1
10 p.m. to 11 p.m.	24	21	2	1
11 p.m. to 12 midnight ..	17	11	4	2
12 midnight to 1 a.m.	28	19	6	3
1 a.m. to 2 a.m.	62	39	15	8
2 a.m. to 3 a.m.	30	23	5	2
3 a.m. to 4 a.m.	38	23	10	5
4 a.m. to 5 a.m.	30	19	7	4
5 a.m. to 6 a.m.	18	6	10	2
Total	266	177	60	29

TABLE 3.
All-Night Catches by Four Men, 28th June, 1944.

Time.	Total.	*A. p. farauti.*	*A. p. punctulatus.*	Intermediates.
6.30 p.m. to 7 p.m.	6	4	2	—
7 p.m. to 8 p.m.	18	9	7	2
8 p.m. to 9 p.m.	19	9	8	2
9 p.m. to 10 p.m.	24	7	12	5
10 p.m. to 11 p.m.	25	11	8	6
11 p.m. to 12 midnight	35	12	16	7
12 midnight to 1 a.m. ..	50	13	25	12
1 a.m. to 2 a.m.	55	25	17	13
2 a.m. to 3 a.m.	55	30	17	8
3 a.m. to 4 a.m.	48	24	15	9
4 a.m. to 5 a.m.	42	19	12	11
5 a.m. to 6 a.m.	24	12	4	8
Total	401	175	143	83

It will be noted that there was a fairly steady rise during the night, with the peak period occurring between midnight and 5 a.m. without any significant difference between the two subspecies. On the other hand, Major Atherton (unpublished data), working at Lalapipi, found that the peak period of adult abundance of *A. p. farauti* was round about 9 p.m.

One all-night catch of *A. p. farauti* made at Cairns by Major Roberts (unpublished data) gave a fairly even abundance from 9 p.m. onwards with a slight peak round 1 a.m.

Oviposition from Laboratory Bred Females.

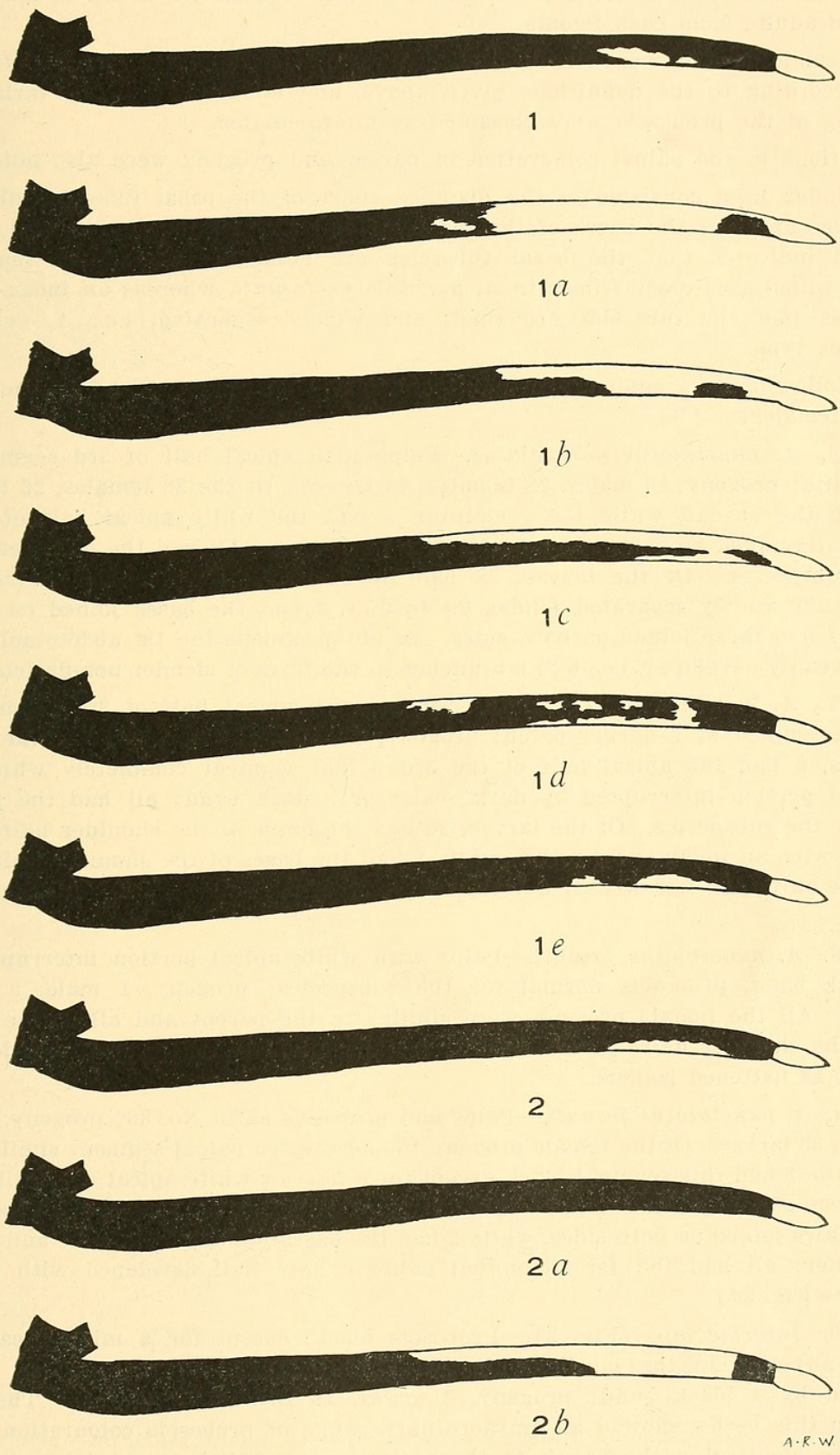
It had been noted previously (Lee and Woodhill, 1944) that at least one species of Australasian anopheline would deposit fertile eggs when bred in small laboratory cages, and an attempt was therefore made to establish a laboratory culture of *A. punctulatus punctulatus.**

Larvae were collected in the field and the resulting adults, approximately 200 males and 200 females, were allowed to emerge in a cage 8″ × 9″ × 15″, and were kept supplied with sweetened water and given a blood feed every alternate night. As the males died off approximately 150 additional laboratory bred males were added during a period of ten days. Under these conditions 170 eggs were deposited in four separate batches from the 7th to the 13th day after the females emerged. From these eggs about 50 adults were obtained, but these did not deposit eggs.

It would appear that copulation under these conditions only takes place between a very small percentage of individuals and that laboratory cultures would be very difficult to maintain. It was felt that considerable information could be obtained if *A. punctulatus punctulatus* could be crossed with *A. punctulatus farauti* in this way.

* As far as is known to the author, Lieutenant F. B. Bang of the U.S. Army Medical Corps was the first to establish a laboratory culture of *A. punctulatus punctulatus.*

As it was not possible to obtain sufficient *farauti* larvae at Salamaua, the author suggested to Major I. M. Mackerras of the Medical Research Unit at Cairns that the attempt should be made there. Some progress has been made along these lines but no definite results are yet available.



Figs. 1-2*b*.—Proboscis colouration in intermediate females and their progeny. 1. Intermediate female No. 24. 1*a* to 1*e*. Progeny of No. 24. 2. Intermediate female No. 20. 2*a* and 2*b*. Progeny of No. 20. All × 54.

Morphology of Adults and Larvae from Area 2.

In this area, as described above, both *A. punctulatus punctulatus* and *A. punctulatus farauti* were breeding, and collections of adult mosquitoes gave 18% intermediate forms.

In studying the morphology the method used was to obtain eggs from single females caught in the field and to breed these through, thus obtaining a series of fourth stage larvae and adults from each female.

The female parents were classified as *A. punctulatus punctulatus* or *A. punctulatus farauti* according to the definitions given above, and all those showing variations in the scaling of the proboscis were classified as intermediates.

Variations in the palpal colouration of parent and progeny were also noted.

The index used consisted of the distance apart of the basal tubercles divided by the distance apart of the bases of the inner and middle shoulder hairs. An index of 0·1 to 0·3 indicates that the basal tubercles are enlarged and almost touching, a condition which occurs sometimes in *A. punctulatus farauti*, whereas an index of 0·4 to 0·7 denotes that the tubercles are small and widely separated, i.e., *A. punctulatus punctulatus* type.

The following is a summary of adult and larval characters found in the progeny of single females:

No. 34. *A. punctulatus punctulatus.*—Palps with apical half of 3rd segment completely white; progeny, 10 males, 28 females, 43 larvae. Of the 28 females, 22 had palps resembling the parent, while the remaining 6 had the white apical half of the 3rd segment interrupted by a few dark scales or a dark ring. All had the proboscis normal for the subspecies. Of the larvae, 38 had the bases of the shoulder hairs weakly developed and widely separated (index 0·4 to 0·7), 4 had the bases joined on one side only, and 1 had these joined on both sides. In all specimens the 1st abdominal palmate hair was weakly developed, i.e., 6 to 9 branches in the form of slender non-flattened hairs.

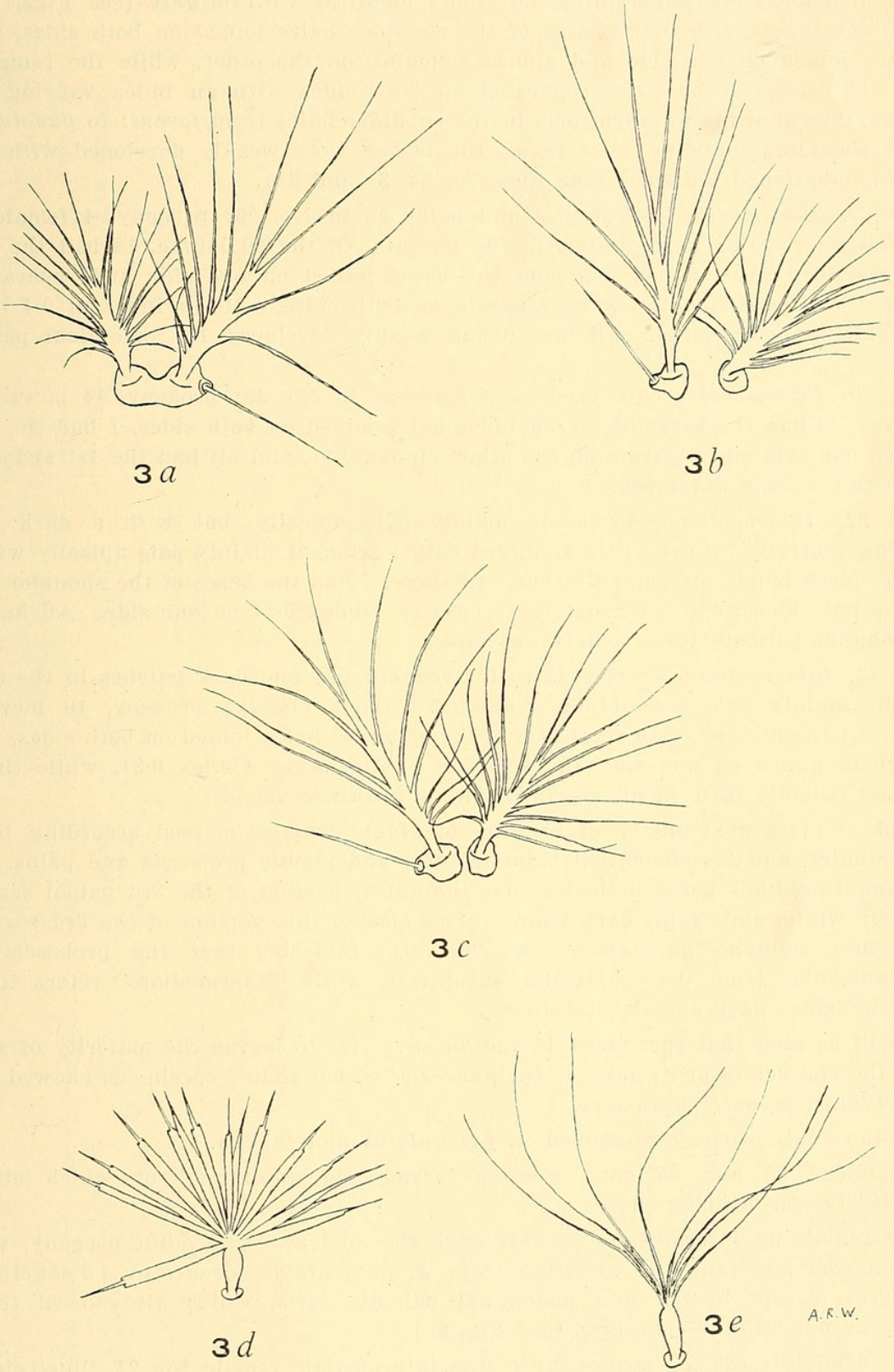
No. 45. *A. punctulatus punctulatus.*—Palps with apical half of 3rd segment only faintly interrupted by a darker patch; progeny, 6 males, 15 females, 22 larvae. Of the 15 females, 6 had the apical half of the 3rd palpal segment completely white, while 9 had this portion interrupted by dark scales or a dark band; all had the proboscis normal for the subspecies. Of the larvae, 20 had the bases of the shoulder hairs weakly developed, with an index of 0·4 to 0·7, while 2 had the bases of the shoulder hairs joined on one side only. All had weakly developed 1st abdominal palmate tufts as described for No. 34.

No. 38. *A. punctulatus farauti.*—Palps with white apical portion interrupted by a wide black band, proboscis normal for the subspecies; progeny, 1 male, 9 females, 11 larvae. All the female progeny were similar to the parent and all larvae had the bases of the shoulder hairs joined and the 1st abdominal palmate tufts well developed, with 16 to 18 flattened leaflets.

No. 35. *A. punctulatus farauti.*—Palps and proboscis as in No. 38; progeny, 5 males, 18 females, 33 larvae. Of the female progeny 16 had the 3rd palpal segment similar to the parent, while 2 had this segment black except for a narrow white apical ring; all had the proboscis completely black except for the labella. Of the larvae, 31 had the bases of the shoulder hairs joined on both sides, while 2 had the bases joined on one side and separate on the other; all had the 1st abdominal palmate hair well developed with flattened leaflets (see Fig. 3*d*).

No. 20. *Intermediate* (Fig. 2).—Proboscis black, except for a minute pale patch ventrally adjacent to the labella, palps with white apical portion of 3rd segment interrupted by a black band; progeny, 3 males, 19 females, 4 larvae. The female progeny in this series showed an extraordinary range of proboscis colouration varying from forms identical with *farauti* to those closely resembling *punctulatus* (see Figs. 2*a* and 2*b*).

The third palpal segment in 17 specimens had the white apical portion interrupted by a dark band, but in 2 specimens these segments were completely dark except for a narrow white apical ring. This is a character usually found more frequently in *farauti* than *punctulatus*, but nevertheless these 2 specimens show a considerable amount of pale scaling in the proboscis.



Figs. 3*a*-3*e*.—Structure of shoulder hairs and 1st abdominal palmate hairs in *A. punctulatus* larvae. *a*, *b*, and *c*. Variations in shoulder hairs of larval progeny of female No. 24 (see Fig. 1). *e*. First abdominal palmate hair of larval progeny of female No. 24. *d*. First abdominal palmate hair of larval progeny of female No. 35. (*A. punctulatus farauti* from Salamaua.) *a*, *b*, and *c* × 190, *d* × 416, *e* × 390.

The 4 larvae had the bases of the shoulder hairs joined, but the 1st abdominal palmate tufts were weakly developed with fine non-flattened branches, as in *punctulatus*.

No. 24. *Intermediate* (Fig. 1).—Proboscis and palps as in No. 20; progeny, 2 males, 8 females, 31 larvae. The females in this series showed a range of variation in proboscis colouration similar to the progeny of No. 20, with one specimen practically indistinguishable from *punctulatus*, but none identical with *farauti* (see Figs. 1*a* to 1*e*). Of the larvae, 5 had the bases of the shoulder hairs joined on both sides, 8 had the bases joined on one side and almost touching on the other, while the remaining 18 had the bases of the hairs separated on both sides with an index varying from 0·2 to 0·6, thus showing all variations in the shoulder hairs from *farauti* to *punctulatus*. The 1st abdominal palmate tufts in all the larvae were weakly developed with from 9 to 13 non-flattened branches (see Figs. 3*a*, 3*b*, 3*c* and 3*e*).

No. 37. *Intermediate*.—Proboscis and palps as in No. 20; progeny, 4 females, 20 larvae. All 4 females were similar to the parent. Of the 20 larvae, 18 had the bases of the shoulder hairs joined, one had the bases joined on one side and separate on the other, while one had the bases separate on both sides with an index of 0·4 to 0·5, i.e., similar to *punctulatus*. All larvae had weakly developed 1st abdominal palmate tufts.

No. 50. *Intermediate*.—Proboscis and palps as in No. 20; progeny, 14 larvae. Of the larvae, 13 had the bases of the shoulder hairs joined on both sides, 1 had the bases joined on one side and separate on the other (index 0·4), and all had the 1st abdominal palmate tuft weakly developed.

No. 22. *Intermediate*.—Proboscis mainly pale apically, but with a dark patch projecting ventrally into the pale area, 3rd palpal segment mainly pale apically without a distinct black band; progeny, 8 larvae. Of these, 7 had the bases of the shoulder hairs joined on both sides, while 1 had these separated (index 0·3) on one side. All had the 1st abdominal palmate tufts weakly developed.

No. 43. *Intermediate*.—Proboscis with irregular pale and dark patches in the apical area, but mainly pale resembling *punctulatus* fairly closely; progeny, 10 larvae, 4 females. Of the larvae, 8 had the bases of the shoulder hairs joined on both sides, while 2 had them joined on one side and separate on the other (index 0·2), while the 1st abdominal palmate tufts in all specimens were weakly developed.

Table 4 gives a summary of progeny of single females classed according to the shoulder hairs, and 1st abdominal palmate tufts and female proboscis and palps. The term "*annulipes*-like" palps indicates that the apical portion of the 3rd palpal segment is entirely white, and "faint dark band" refers also to this portion of the 3rd segment. In the first column "*punctulatus*" or "*farauti*" indicates that the proboscis was indistinguishable from that particular subspecies, while "intermediate" refers to any form of irregular dark and light scaling.

It will be seen that specimens 34 and 45 gave rise to larvae the majority of which had all the characters of *A. punctulatus punctulatus*, but that 7 specimens showed some *A. punctulatus farauti* characters.

All the adult progeny resembled *A. punctulatus punctulatus*.

Specimens 38 and 35 gave rise to larvae and adults all of which showed *A. punctulatus farauti* characters.

The remaining intermediate parents gave rise to larval and adult progeny, which showed a complete range of variation from *A. punctulatus farauti* to *A. punctulatus punctulatus*, except in the first abdominal palmate tufts, which all showed the *A. punctulatus punctulatus* character (see Fig. 3*e*).

Of special interest is the progeny of the intermediate female No. 24, illustrated in Figs. 1*a* to 1*e* and 3*a* to 3*c*. It will be noted that although the female parent had only a very small ventral pale patch on the proboscis, some of the adult progeny were indistinguishable from *A. punctulatus punctulatus*, while the shoulder hairs of the larvae showed a complete range of variation from the *A. punctulatus punctulatus* to the

TABLE 4.

Number and Description of Female Parent Proboscis and Palps.	LARVAE.							ADULT FEMALES.					
	Bases of Shoulder Hairs.					First Abdominal Palmate.		Proboscis.				Palpi.	
	Number of Larvae.	Number Joined on Both Sides.	Number Joined on One Side Only.	Separate on Both Sides		Number with Slender Branches.	Number with Flattened Leaflets.	Number of Females.	*punctulatus.*	*farauti.*	Intermediate.	Normal Palpi.	Annulipes-like Palpi.
				Index 0·1 to 0·3.	Index 0·4 to 0·7.								
34. *Punctulatus*, *annulipes*-like palps	43	1	4	—	38	43	—	28	28	—	—	8	22
45. *Punctulatus*, faint dark band	22	—	2	—	20	22	—	15	15	—	—	9	6
38. *Farauti* palps normal	11	11	—	—	—	—	11	9	—	9	—	9	—
35. *Farauti* palps normal	33	31	2	—	—	—	33	18	—	18	—	18	—
20. Intermediate palps normal	4	4	—	—	—	4	—	19	—	2	17	19	—
24. Intermediate palps normal	31	5	8	12	6	31	—	8	1	—	7	8	—
37. Intermediate palps normal	20	18	1	—	1	20	—	4	—	—	4	4	—
50. Intermediate palps normal	14	13	1	—	—	14	—	—	—	—	—	—	—
22. Intermediate, *annulipes*-like palps ..	8	7	1	—	—	8	—	—	—	—	—	—	—
43. Intermediate palps normal	10	8	2	—	—	10	—	4	—	1	3	4	—

A. punctulatus farauti type. Again, intermediate female No. 20 gave rise to some adult progeny (Fig. 2*a*) with a completely black proboscis, i.e., the *A. punctulatus farauti* type.

Discussion.

The theory that the intermediate forms are hybrids between *A. punctulatus farauti* and *A. punctulatus punctulatus* appears to be supported by the field observations detailed above, since as far as could be ascertained only *A. punctulatus punctulatus* was breeding in areas 1 and 3 (Map 1), whereas both subspecies were breeding together in area 2 where the intermediate forms were obtained. It is significant also that in areas where only *A. punctulatus farauti* occurs, e.g., the mainland of Australia and at Merauke, no intermediates have been recorded from very extensive collections. Intermediates have been taken from most localities in New Guinea where *A. punctulatus punctulatus* and *A. punctulatus farauti* occur together, the percentage of these varying from 80% at Salamaua (Gilmour, unpublished data) to only a few recorded specimens at Milne Bay (Allman, unpublished data). The fact that no intermediates occurred in many thousands of adults bred from open sunny pools in areas 1 and 3 indicates that their breeding habits probably resemble those of *A. punctulatus farauti.*

The difference in breeding habits of *A. punctulatus farauti* and *A. punctulatus punctulatus* is emphasized by the fact that in area 2 only light breeding of *A. punctulatus farauti* could be located, but adult counts showed over 50% of this subspecies; as previously recorded by many observers in coastal New Guinea, *A. punctulatus punctulatus* larvae are found in all the obvious open sunny breeding places, while *A. punctulatus farauti* larvae are usually hidden and far more difficult to locate.

The breeding experiments from individual females also indicate that the intermediate forms are hybrids between the two subspecies since both larval and adult progeny of intermediate females show all variations from the *A. punctulatus punctulatus* to the *A. punctulatus farauti* type. Since it has been found possible to maintain continuous laboratory cultures of *A. punctulatus punctulatus* (Lemerle, unpublished data), it is to be hoped that workers in this field will attempt to cross *A. punctulatus punctulatus* with *A. punctulatus farauti* on a large scale and thus finally demonstrate whether inter-fertility occurs between the two subspecies.

In view of the recent work by Owen (1945) and by Belkin, Knight and Rozeboom (1945) in the Solomon Islands, it must be pointed out that the findings of these authors in relation to the "*punctulatus* complex" in that area do not apply in certain areas in New Guinea and Australia. Reference to Figs. 1–3 will show that female No. 24 has proboscis colouration identical with that described for *A. koliensis* Owen, while other individuals which are the progeny of this same female parent have the proboscis either identical with *A. punctulatus punctulatus* or intermediate between that subspecies and *A. koliensis.* Again a single female (Fig. 2) with a proboscis of the type described for *A. koliensis* gave rise to progeny, some of which (Fig. 2*a*) had a proboscis identical with *A. punctulatus farauti*, i.e., completely dark.

Owen also states that *A. koliensis* can usually be separated from *A. punctulatus* by the absence of a small dark spot on the costal margin of the wing between the basal and median dark spots; in the progeny of two individual females from Salamaua (Nos. 34 and 45) with probosces of the *A. punctulatus punctulatus* type, 7 out of 38 and 7 out of 21 respectively, had this black spot absent, and in many hundreds of specimens of both *A. punctulatus punctulatus* and *A. punctulatus farauti* examined from New Guinea and Australia this character was found to be extremely variable. Again, Belkin, Knight and Rozeboom separate *A. farauti* larvae from those of *A. koliensis* and *A. punctulatus* by the presence of a true palmate tuft on abdominal segment I and the fusion of the tubercles of prothoracic hairs 1 and 2. Reference to Figs. 3*a*, 3*b* and 3*c* shows that the larval progeny of female No. 24 (with *koliensis* type proboscis) had shoulder hairs showing all stages between widely separated and completely fused tubercles. Also the larvae of *A. punctulatus farauti* from the Northern Territory of

Australia (where there are no forms with any pale scaling on the proboscis) all have hair 1 of abdominal segment I with narrow hair-like branches. The branching of the outer occipitals (used as a distinguishing character for *A. koliensis*) is also found to be extremely variable for *A. punctulatus punctulatus* and *A. punctulatus farauti* in New Guinea and Australia, varying from 1 to 5 branches in *punctulatus* and from 2 to 6 in *farauti*.

During the course of examining some thousands of specimens of the subgenus *Myzomyia* from various widely separated parts of the Australasian Region from the Solomon Islands to Morotai, and including the whole of Australia, the outstanding fact which has been brought out is the extreme variability of such characters as the relative length of pale and dark bands on the palps, the number of black patches on the costal margin of the wing, the amount of scaling on the abdomen, the number of branches in the posterior clypeal and sutural hairs and the form of the shoulder hairs and first abdominal palmate hairs. Very frequently these characters are found to be quite constant in a restricted area, but when specimens from widely separated areas within the Australasian Region are examined, variations in the above characters almost invariably occur.

It would seem therefore that *A. koliensis* Owen exhibits constant characters in the Solomon Islands, but that specimens from New Guinea which are morphologically identical with *A. koliensis*, cannot be considered as such since they give rise to *A. punctulatus punctulatus* and *A. punctulatus farauti* in their progeny.

In view of the above it would surely be more logical to regard *A. koliensis* as a subspecies of *A. punctulatus* rather than as a distinct species.

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