The Symbiote of *Lakshadia communis.*

By

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(With 5 figures in the text and plate 5.)

This article is a continuation of series of communications ending with "the symbiotes of some important lac insects" [1]), so that further references to literature may here be avoided.

One of the most important factors considered in evaluating shellac is its colour. Many experts have believed the different localities which produce stick lac or the raw material offer one and the same biological entity. To M. HAUTEFEUILLE, the French lac-expert in Indo-China, goes the credit of being the first to clearly state, for example, that the difference in the colour of shellac as manufactured in Tonkin and in India is due entirely to two sorts of stick lac which themselves represent insects not biologically identical. However other factors have also been mentioned in producing variation in colour, such as the season of the year and the nature of the host plant.

One species most suited for such a study is *L. communis* which grows on a large number of trees. An encrustation on its most favourite host, *Ficus mysorensis*, has been previously illustrated [2]). An encrustation far more typical of this insect is shown here in Pl. 5 Fig. 1, as collected from *F. Benjamina*, var. *comosa*, Lalbag, Bangalore, 20 Oct. 1921. At the lower extremity of the encrustation is seen a large, somewhat deformed, crown shaped cell, the nature of which

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[1]) This Journal Vol. 73 p. 164.
[2]) This Journal Vol. 68 Pl. 17 Fig. 4.

has been explained previously. The encrustation is seen completely enveloping the twig which was growing more or less vertically. It has been previously explained that on such a twig from the very beginning the larvae settle all around it while on one which is horizontally growing the larval settlement takes place on the side facing the gravity so that the later development of the lac encrustation is very disproporionate. Pl. 5 Fig. 2 shows such a chunk of lac fresly collected from *Anona squamosa*, Lalbag, 1 Nov. 1921. Only the lower surface, the side not shown, contained the major portion of the encrustation. The upper side seen here however still shows the difference in colour when compared with Pl. 5 Fig. 1; encrustations on *A. squamosa* are always relatively yellower. It must be clearly mentioned the difference so pronounced in the fresh condition almost disappears on the encrustations drying, while alcoholic solutions offer no indication of any difference whatsoever. Why the fresh encrustation of *A. squamosa* alone should show a specific character is not yet clear to me. On this tree when the insects show scattered settlement the individual cells appear more circular, having a puffed out body shape, with a uniformly thin coat of lac resin enveloping the insect. The entire colony also shows the ultimate effect of individuals exhibiting such conditions of growth. Pl. 5 Fig. 2 shows on the upper left side and again in the middle a couple of cells which look very rounded and characteristic. This observation needs photographic evidence and would be explained at length in a future publication. In Pl. 5 Fig. 2 the lower extremity is seen having a large crown shaped cell wich represents a very natural appearance.

In order to show the influence of time two illustrations Pl. 5 Figs. 4 and 5 are offered from specimens collected in the Lalbag, Bangalore from *Caesalpinea sappan*, during 1887 and kept in the Economic Museum of the Mysore State until they were kindly presented to me by the courtesy of Mr. Grunada Acharia, the Superintendent in Charge. Both these specimens show damage by the caterpillars of the moth *Eublemma amabilis*, the large conspicuous holes in both the encrustations are indicative of this destruction. Through these apertures it is much easier for rain water to get into the encrustation and leach out some dye outside. However it seems to me encrustation, Pl 5 Fig. 4 was ceraitnly one year older than the one shown in Pl. 5 Fig. 5. On the lower right side of this figure a small crown shaped cell is seen lighter in colour and

raised above the surface of the encrustation adhering like a foreign body.

Pl. 5 Fig. 6 would offer an explanation as to Pl. 5 Fig. 4 appearing so dark. It was collected during July 1919 at Lingal, Mahboobnagar District, Hyderabad State from *Butea fronodosa* growing on the bund of an old Tank of Telugu origin, now in ruins for centuries, not far from an old Iron foundary. The encrustation was the only piece seen and a number of other twigs were found thickly covered with the dried cells of winged male larvae. The history constructed is as follows. A crown shaped cell, which meanwhile dropped off and was not collected, started life with Dec. 1917 and gave rise to a normal generation with preponderating females about June 1918. These insects formed the encrustation piece here shown in Pl. 5 Fig. 6. The generation issuing from this encrustation beginning about Dec. 1918 gave rise to all winged males which emerged and died as bachelors about Feb. 1919 so that only their dried larval cells could be collected about July 1919. The encrustation Pl. 5 Fig. 6 was thus already devoid of life by Dec. 1918 so that later showers of rain, specially during March 1919 and again during June, leached out the dye spreading it at the outer surface there undergoing darkening in colour. It was easy to soak a dry stick lac in water and ultimately allow it to grow dark under experimental conditions to imitate the probable process going on in nature. Thus the dark colour specimen representing growth during the monsoon season of 1886, Pl. 5 Fig. 4 was most probably just as dark when it was collected in 1887. At any rate it compares very well with respect to colour Pl. 5 Fig. 6, which was certainly not longer than 18 months from the time of larval settlement to the time when it was collected as a dry specimen.

When alcoholic solutions of such dark looking encrustations are made and compared with other normal looking encrustations they do not differ in intensity which is comparable with the observation that lac dye even in its unaltered form shows no solublity in this solvent. Hautefeuille mentions having come across dark coloured specimens of stick lac and hesitates offering one answer to such a question. It seems to me considering that the Tonkin lac is the next darkest variety of stick lac and that it is the richest sorurce of lac dye a similar process in nature is just as likely to be frequent.

Having experimentally established at least *A. squamosa* gives encrustations looking quite characteristic attempts were made to find if the symbiotic cells assumed any special form when the insects were growing on this host plant. Text-Fig. 1 shows representative

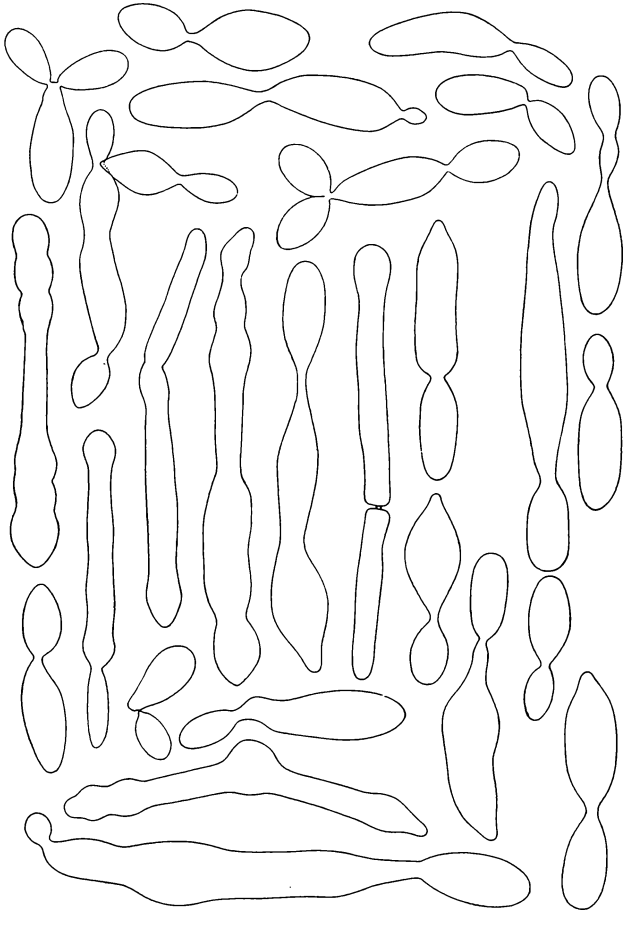

Fig. 1. The symbiote of *L. communis* on *A. squamoasa,* from a female larva before the third moult, collected 15 June 1925.

shapes of the symbiotic cells in the blood smears of *L. communis.* The specimen which supplied this picture was a female larva just before the third or last moult growing in Bangalore on 15 June 1925. A very thorough search showed that the host plants had no influence on the morphology of the symbiotic microorganism although specific differences between the insects could be thus established.

As a result of other observations and experiments to be reported separately it was found that the water supply to the host plant influences the sex determination of the next generation of this and another species. For example on two plots of *Acacia farnesiana* the species *Lakshadia sindica* was inoculated. One lot of plants were daily irrigated and the other left to nature. From the plants receiving water artificially the next generation consisted of only winged males. On plants which had not received water the mother insects were so predisposed as to give a preponderence of females in the next generation; but the deficiency of water so effected the mother insects that they died in great numbers and only a few survived by the time the next generation swarmed as larvae. For such experiments it became pertinent to know how much water could be given from time to time to prevent premature death due to drought and yet not liberal to predispose the females to give rise to a generation of males. It was of course very easy to produce large number of males which was brought about by daily and liberally watering the plants and thereby also reducing the death rate among the mother insects. After much labour and as much reflection it was found that the size of the rod shaped symbiotic cells was a good index. Text-Fig. 2 shows selected cells from a normal blood smear of *L. communis*, not fully grown, on *A. squamosa*, Bangalore, 20 Sep. 1924. The insects were well developed in size and the internal symbiotes were correspondingly long. A premature stage or late larval stage under similar favourable conditions of growth would give an identical picture. In other

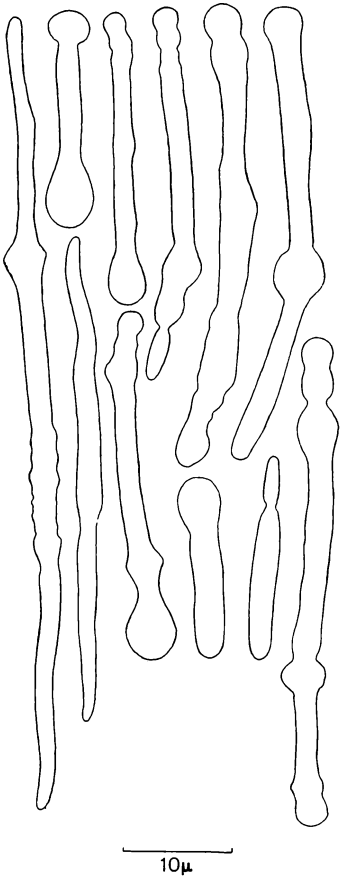

Fig. 2. Symbiotic cells selected from a blood smear preparation supplied by an individual not fully grown on *A. squamosa*, 20 Sept. 1924. The long rods inidicate good growth and a fair degree of water supply.

words the rate of growth can be roughly judged by the length of such rod shaped symbiotic cells from a mixed blood smear preparation.

Text-Fig. 3 is a selected picture from a blood smear of an immature female, shortly after fertilisation, growing on *A. squamosa*, Bangalore, 5 March 1923. The smaller size of the symbiotie cells indicate indirectly the relatively poorer rate of growth during the non-monsoon season, a fact which can be directly confirmed by comparing the size of the insects growing during the two seasons.

The rod shaped cells are by no means the preponderating ones but the more usual or yeast shaped forms often go together in chains of three or four cells and do not lend to such a convenient comparison.

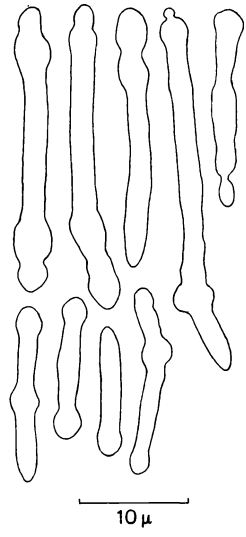

Fig. 3. Symbiotic cells selected from a blood smear of a female shortly after fertilisation on *A. squamosa*, 5 March 1923. The small size of the rods indicate drought conditions and a poorer rate of growth but a very favourable condition for the female sex predominance in the next generation.

As yet no illustration has appeared of a colony where all the individuals consist of winged males. Pl. 5 Fig. 3 supplies this deficiency. It shows part of a colony on *F. mysorensis*, Bangalore, 28 Jan. 1922. Some cells are in their second larval stage and show small white tufts of anal hairs; the majority of them would be in the third or prepupal larval stage. From one of the above cells in the second stage Text-Fig. 4 was drawn and shows nothing uncommon from the shape of the symbiotic cells seen in the female.

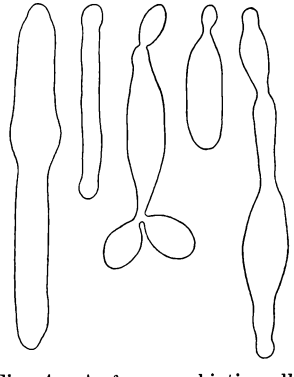

Fig. 4. A few symbiotic cells from a second stage larva of the winged males, 28 Jan. 1922.

In order to indicate the specific nature of the symbiotic organism of *L. communis* a carefully drawn picture of the symbiotic cellsof *L. mysorensis* as seen in a normal blood smear preparation is offered for comparison. Text-Fig. 5 was supplied by an individual with nearly fully developed eggs. The picture was made on 15 Dec.

1924, the larval swarming was observed on 29 Dec. The host plant was *Shorea talura*, growing at Dursanipalya, near Bagalore. Contrastig Text-Figs. 1 and 5 their specificity is at once evident and this fact is again emphacised in the obsence of all confirmations as to the specific difference between these two species by other workers on the subject.

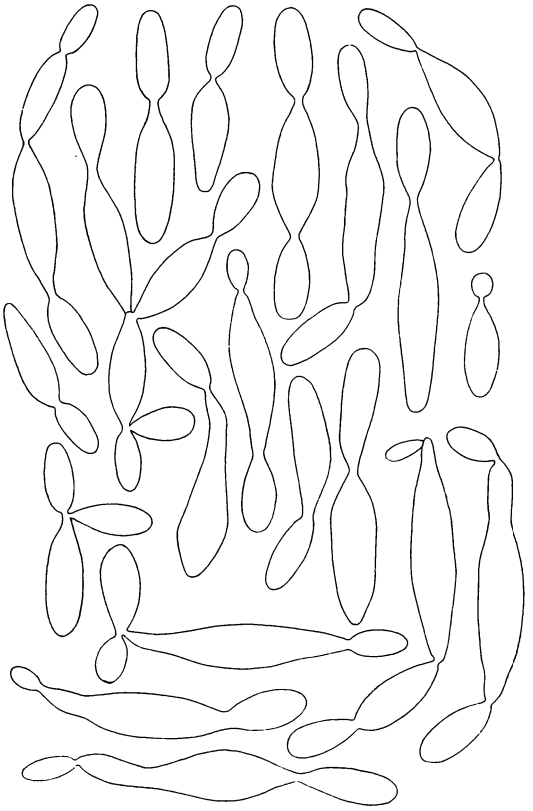

Fig. 5. The symbiote of *L. mysorensis* on *Shorea talura*, 15 Dec. 1924. The cells indicate a specific difference from those of *L. communis*, Fig. 1.

It may be added that the rod shaped forms of symbiotes have never been observed free from other forms in artificial cultures although so met with sometimes in insect body. Extracts of lac insects with different range of acidity and other alterations never offered a clue to the forms observed in blood smears. In this connection I have had the opportunity of consulting Prof. Lindner of Berlin and Prof. Šatava of Prague and I beg to thank these authorities again for their kindness and interest.

Summary.

The species *L. communis* produces the darkest coloured sort of stick lac. Under circumstances the dye may be leached out and spread outside the surface of the encrustation where it may darken and give the encrustation an almost black appearance. This has not been observed with other sorts. However the alcoholic solution does not dissolve the brown-black colour just as the lac

dye itself is not sululble in this solvent. On *A. squamosa*, *L. communis* builds charateristic encrustations, among others possessing a relatively yellow colour in the fresh condition. The symbitotic microorganisms do not morphologically differ when the species grows on different host plants.

In blood smear preparations the symbiotic cells show a great degree of polymorphism, among others rod shaped forms. These offer as test object for the condition of water supply to the insect and the corresponding rate of growth. It was found that liberal water supply before fertilisation predisposed the females to produce all males in the next generation; scarcity of water supply on the contrary induced the production of preponderating females but the mother insects died from drought. It was necessary to known how to controle water supply reducing death rate yet favouring the preponderance of females in the next generation. The greater length of the rod shaped symbiotes shows excess of water and speaks for the mother insects being predisposed to produce males while the shorter length of the rods indicates limited water supply ultimately conducive to the production of the female sex in the next generation.

The cost of illustrations was generously met by a grant sanctioned by the Council of the Indian Institute of Science and in this connection it is a pleasure to thank again Dr. M. O. Forster F.R.S., the Director and Prof's. Fowler and Norris of the same Institute.

But for the kindness of Mr. Turab Ali, then Superintendet of the Criminal Settlement at Lingal, I should not have been able to make some of the important observations recorded in this paper and in this connection I have also to thank Mr. Wakefield C.I.E., now Foreign Secretary at Kashmir for kindly sanctioning my visit to Lingal.

Explanation of Plate.

Plate 5.

Fig. 1. Encrustation of *L. communis*, freshly collected on *Ficus Benjamina* var. *comosa* 20 Oct. 1921.

Fig. 2. A fressly collected encrustation on *A. squamosa*, 1 Nov. 1921. The encrustation is seen from above; the twig was horizontally growing.

Fig. 3. A colony of only winged males, in the second and third larval stages, on *F. mysorensis*, 28 Jan. 1922.

Fig. 4. Encrustation from *Caesalpinea sappan* grown during the monsoon season of 1886, collected and dated Bangalore 1887.

Fig. 5. Encrustation collected as indicated in Fig. 4 but representing growth during the monsoon season of 1887.

Fig. 6. Encrustation from *Butea frondosa* growing during the monsoon season of 1918, collected at Lingal, Hyderabad State, July 1919.


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