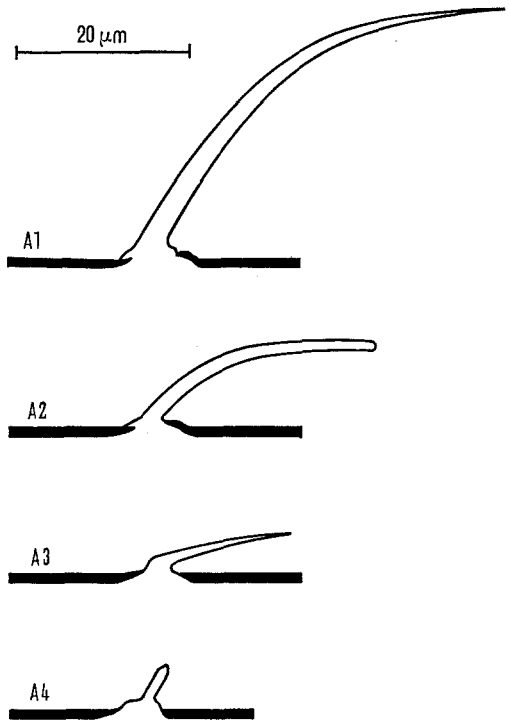

Haarförmige, dünnwandige Sensillentypen auf den Antennen weiblicher *Aedes aegypti*. Schematische Zeichnung nach auflichtmikroskopischen Beobachtungen der Antennen in vivo.

trischen Charakteristika bei elektrophysiologischen Ableitungen.

Bei Reizung von Sinneszellen des Typs A 3 mit dem konzentrierten Abschreckungsstoff DEET (N, N-Diethyl-m-toluamid) wurde eine sehr starke Impulsfrequenzsteigerung beobachtet. Dagegen zeigten Sinneszellen des Typs A 4 keine Beeinflussung ihrer Spontanaktivität bei Reizung mit DEET. Sie wurden aber von einer attraktiv wirkenden menschlichen Schweissfraktion[6] erregt. Diese Substanz war bei den Zellen des Typs A 3 wirkungslos[7,8].

Summary. A new type of thin-walled, hair-like setae was found on the antennae of the female mosquito. The length of the 3 different types, already described in the literature, is compared with the author's own measurements. This new type differs not only in length but in electrophysiological responses.

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Life Sciences Research, Stanford Research Institute, Menlo Park (California, USA), 30. Januar 1969.

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Susceptibility of Various Strains of Chicken to the Oncogenicity of *Yaba* Virus

In a previous paper[1] we reported that *Yaba* virus, an oncogenic pox virus of primates[2], produces tumors on the chorioallantoic membrane of 10- to 11-day-old embryonated hen eggs. Recently our virus laboratories have been moved to the Springville campus of the Institute. We were surprised to find that in the new location we are unable to repeat our previous experiments. Previously we have purchased embryonated eggs from the DeKalb 144 Leghorn strain of the Glor Co., Holland, New York (B) and more recently from the H and N (Nick) Leghorn strain of the Weidner Co., Eden, New York (S). Possible seasonal differences were also considered.

Methods and results. In order to study this problem, in 2 experiments, 10-day-old (B) and (S) eggs were inoculated simultaneously with *Yaba* virus and tumors harvested after 5 more days of incubation, as previously described[1]. The results (shown in Table I) revealed 90 to 95% takes in the (B) eggs and 6% takes in the (S) eggs, regardless of whether the experiment was performed in spring or fall. Figure 1 depicts typical tumors of the chorioallantoic membranes, they ranged in diameter from 0.5–1.0 cm.

In order to study the mechanism of susceptibility and resistance to *Yaba* virus of the 2 strains of chicken we have inoculated 4-day-old eggs into the allantoic cavity with 0.2–0.5 ml of allantoic fluid from the other strain. At the age of 10 days, the chorioallantoic membranes were inoculated with 0.3 ml of a 10% *Yaba* tumor homogenate obtained from infected stumptail monkeys[1,2]. On the sixth day after inoculation the chorioallantoic membranes of the viable eggs were harvested and the presence of macroscopic tumors determined. Control groups were inoculated with virus but not with allantoic fluid. Table II

Table I

Season	No. with tumors/No. inoculated (B) eggs	(S) eggs
Fall	46/51 (90%)	3/54 (6%)
Spring	38/40 (95%)	2/35 (6%)
Total	84/91 (92%)	5/89 (6%)

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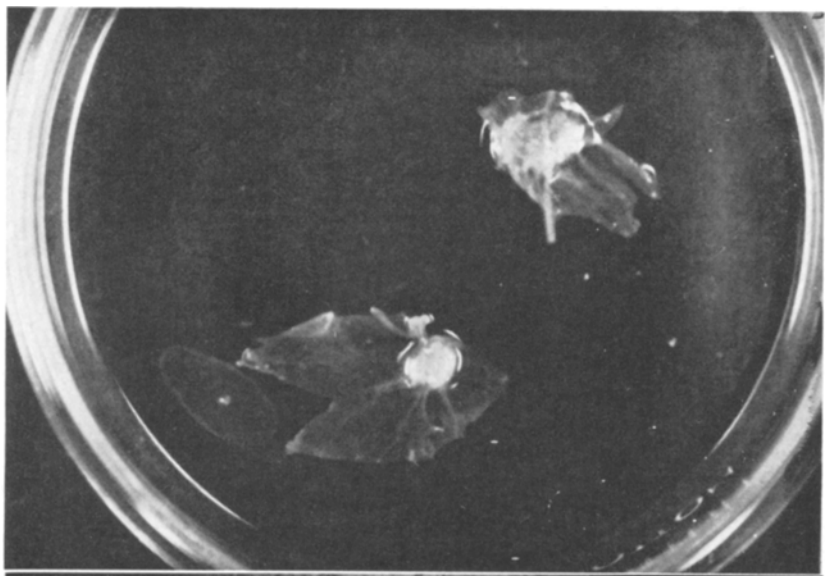

Fig. 1. *Yaba* virus induced tumors on the chorioallantoic membranes of embryonated hen eggs inoculated at 10 days of age and harvested after 5 days of additional incubation.

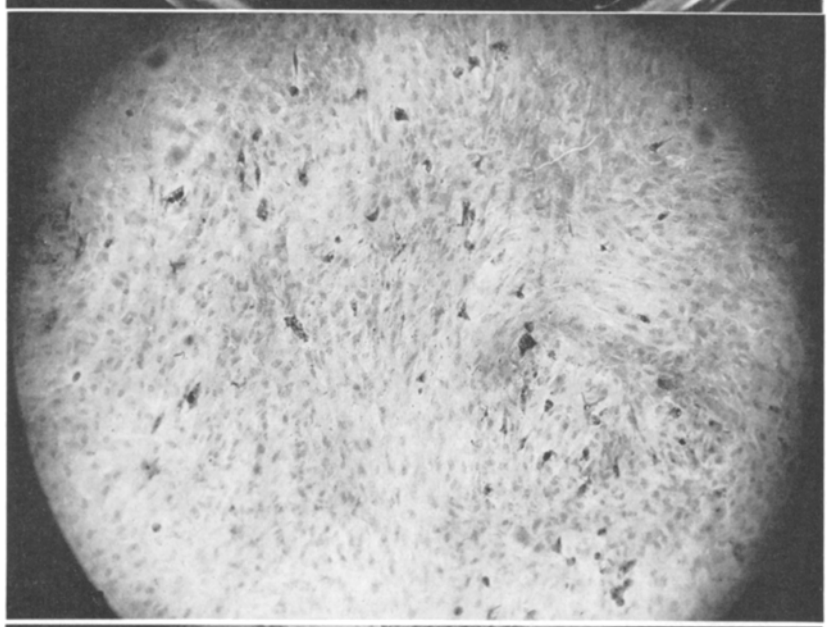

Fig. 2. Chicken fibroblast culture.

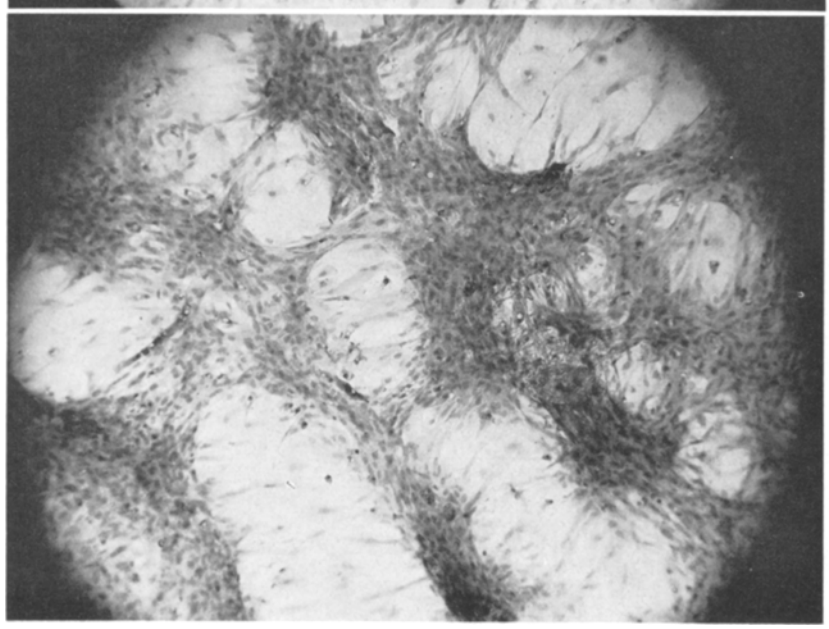

Fig. 3. Chicken fibroblast culture 7 days after inoculation with *Yaba* virus.

summarizes the results. In (S) eggs, in which no tumor takes were observed, inoculation of (B) allantoic fluid resulted in 30% takes. By contrast (B) eggs, in which tumor takes were 83%, inoculation of (S) allantoic fluid reduced the take to 58%. Mortality from allantoic fluid inoculation alone was 10% in the (B) into (S) series and 23% in the (S) into (B) series. It appears that allantoic fluid can transfer some susceptibility and also some resistance to *Yaba* virus. (S) allantoic fluid seems to be more toxic to (B) embryos than (B) allantoic fluid to (S) embryos.

In order to further study this phenomenon we investigated the effect of *Yaba* virus on primary fibroblast cultures, from 10-day-old chicken embryos of both strains. To establish the fibroblast cultures modified Parker 199 medium was used[3] with 20% tryptose phosphate broth and 20% calf serum. After 2 or 3 days growth *Yaba* virus was added, the medium changed to 50% Eagle's basal medium[4], 50% bovine amniotic fluid and 0.2 ml cell-free filtrates from 10% suspensions of chorioallantoic membrane grown tumors or uninoculated chorioallantoic membranes of (B) eggs. Within one week after virus inoculation the uniform layer of fibroblasts (Figure 2) changed into a number of foci with increased cell density and areas lacking cells in between. The foci were interconnected with multiple 'pseudopodia' between which round empty spaces appeared (Figure 3). Careful examination failed to reveal signs of cytopathogenicity. The cells appeared to have concentrated into the foci rather than died off in between. A nonspecific contraction of cell sheets may occur in congested cultures. This was, however, not the case in these experiments. The phenomenon was not observed in uninoculated cultures. As summarized in Table III, foci occurred in all virus inoculated cultures regardless of the origin of the fibroblasts. No foci were seen in the control cultures.

In a second series of experiments 0.2 ml supernatant fluid from a (B) fibroblast culture (second transfer generation) was used to inoculate primary fibroblasts of (B) and (S) origin. Only in the (B) cultures did foci develop. It appears that susceptibility to *Yaba* virus varies with the strain of the embryo from which cell cultures originate. However, filtrates of (B) grown tumors can transmit susceptibility to (S) cultures similar to the transmission by allantoic fluid. In the second transfer generation the susceptibility-inducing agent may be diluted out (or failed to multiply, or did not multiply at a sufficient rate).

When superantant fluids from (B) cultures with foci were further subcultured, foci continued to appear probably indicating the continued presence of the focus-inducing agent without resulting in significant cytopathogenicity. When supernatant fluids from (S) cultures were subcultured, no foci appeared. Incubation of *Yaba* virus suspensions with anti-*Yaba* immuneserum from monkeys[1] prevented both focus formation in (B) fibroblast cultures and tumor formation in (B) eggs.

Discussion. Resistance to *Yaba* virus in certain strains of chicken embryos may be based on the presence of (a) viruses which induce the production of interferons or interferon-like substances and (b) viruses which directly interfere with *Yaba* virus. For example, in chicken cells the growth of *Rous sarcoma* virus may be inhibited by infection with viruses of the avian leukosis complex without production of interferon. In this instance interference seems to be based on production of viral coat protein which prevents infection by viruses with similar coats probably by interfering with penetration[5]. Resistance inducing viruses may be responsible for the high mortality of susceptible embryos when allantoic fluid from resistant eggs is inoculated.

Susceptibility to *Yaba* virus, on the other hand, may require the presence of helper viruses similar to *Rous sarcoma* virus[6]. A complex interrelationship between these factors may explain why allantoic fluid may transmit both susceptibility and resistance from and into different strains of embryonated eggs. This may be the reason why filtrates from egg grown tumors can transmit not only the virus but also susceptibility to it. These experiments emphasize the complex relationship between oncogenic viruses, host factors and oncogenicity[7,8].

Table II

Origin of eggs	Origin of allantoic fluid inoculated into eggs	Mortality of embryos w/o virus inoculation	Tumor incidence w. virus
(S)	–	0/6	0/6 (0%)
(S)	(B)	6/62 (10%)	17/56 (30%)
(B)	–	0/6	5/6 (83%)
(B)	(S)	14/59 (23%)	26/45 (58%)

Table III

Source of eggs	(B)		(S)	
Passage of virus	1st	2nd	1st	2nd
Occurrence of foci	16/16	6/6	8/8	0/4
Occurrence of foci of uninoculated cultures	0/7	0/2	0/4	0/2

Résumé. Le virus *Yaba*, pox virus oncogène pour les primates, a produit des microtumeurs sur la membrane chorioallantoïque des embryons de poulet de quelques variétés et de différents âges. La sensibilité a pu être transférée d'une variété à l'autre par l'inoculation de liquide allantoïque.

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[7] This study was supported by the American Cancer Society No. E-351A, In-54-F-6, the United Health Foundation of Western New York No. G-65-RP-25, and by a Fellowship from the National Institutes of Health, USPHS to Dr. STRANDSTRÖM No. FO-5889.
[8] Acknowledgment. We wish to acknowledge the devoted technical assistance of Miss SIRKKA KERANEN.