Research Note

Effectiveness of several locally available membranes used for artificial feeding of *Aedes albopictus* Skuse

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Abstract. Artificial feeding of mosquitoes with blood meal is an important technique in the studies of mosquito feeding. Owing to the difficulty in obtaining suitable artificial membranes for mosquito feeding from other sources, several easily obtainable membranes in Malaysia were tested for their suitability as a replacement. Skin of chicken, fish, and salted sausage were obtained and tested against cattle skin membrane as a control. The results showed that cattle skin is still the most favorable membrane to be used, with full engorgement rate of around 57% using fresh human blood. However, processed chicken skin was shown having potential for further testing since with feeding using human blood kept overnight at 4°C, an engorgement rate of 50% was obtained.

Artificial membrane feeding is an important method used in medical entomological research for virus susceptibility testing (Gubler et al., 1979), virus detection methods e.g. for immunofluorscence testing (Kuberski & Rosen, 1977), and others. Examples of artificial membranes cited in literature include collagen membranes and gauze sealed with wound dressing. However, using animal products that are easily obtainable from local markets present an attractive alternative to these methods and the conventional cattle membrane.

Laboratory-bred female *Aedes albopictus* mosquitoes with an age of not more than 5 days were obtained from the insectarium of the Medical Entomology Unit, Institute for Medical Research, Kuala Lumpur. Mosquitoes were placed into paper cups with nettings and starved overnight at room temperature.

The following membranes were tested: fish skin, chicken skin, thinned chicken skin and preserved sausage skin. For comparison, cattle membrane was used as the control. All membranes used, except the control, were processed by first soaking them in warm water at about 70 °C several times to remove the fat from their surfaces. In the case of the thin chicken skin, the endodermal layer was removed and only the epidermal layer used. For the preserved sausage, its contents were removed and the membrane cut open and treated with hot water to remove the fat.

The membranes were stretched across the glass feeding apparatus and the mosquitoes were exposed to it through the netting. Five ml of fresh human whole blood was used each time whenever possible. Feeding was allowed to continue for 40 minutes in each case at a mean temperature of 25 °C.

The degree of feeding was determined by observing the volume of blood taken up by each mosquito. This was done via magnified examination of individual mosquitoes. Engorged mosquitoes were termed fully fed while mosquitoes with

Membrane Type	Total No. Mosquitoes used	No. Fully fed mosquitoes	No. Partially fed mosquitoes	No. Unfed mosquitoes	Percentage fed	Percentage fully fed
Chicken skin	25	7	2	16	36.00	8.00
Thinned chicken skin	10	2	3	5	50.00	20.00
Preserved sausage membrane	6	0	1	5	16.67	0.00
Cattle membrane	14	8	0	6	57.14	57.14

 Table 1. Percentage of mosquitoes fed and fully fed with chicken skin, thinned chicken skin, preserved sausage membrane and cattle membrane

blood in them but not engorged were termed partially fed. Those without blood in them were termed unfed.

Fish skin was deemed unsuitable as a membrane because of the difficulty in applying the hot water treatment. The fish skin was easily cooked and unstretchable.

Owing to the availability of *Ae. albopictus* from the insectarium, not all feedings involved an equal number of individuals. Therefore the percentage that fed and the percentage that became engorged from feeding would be the indication on how much the *Ae. albopictus* mosquito's affinity for the feeding membrane. (Table 1)

Compared to the control i.e. cattle membrane, only the thinned chicken skin came close in terms of attracting Ae. *albopictus* to feed, reaching 50% in fed mosquitoes in which 20% of the total number became engorged. It is worthy to note that owing to constraints in the availability of fresh blood, the blood used for thinned chicken skin feeding was blood stored overnight at -4°C. This may have been a compounding factor in deterring the mosquitoes from feeding. Hence further testing with this membrane is warranted.

Compared to other reported feeding methods for *Aedes* feeding, all the feeding results were less than satisfying. Cosgrove & Wood (1995) reported a mean feeding number of 8.57 out of 10 *Aedes aegypti* from 7 replicate feedings. This difference in feeding preference could be because the mosquitoes used were not originated from a well-established lab colony.

Considering the above, future recommendations for membrane testing would be for the thinned chicken skin using fresh blood and well established *Aedes* mosquito colonies. Furthermore, nylon gauze sprayed with wound spray as described by Hagen & Grunewald (1990) can be tested as another alternative to a renewable membrane for feeding.

Acknowledgements. This experiment was conducted with assistance of the staff of the Unit of Medical Entomology, Institute for Medical Research. Their assistance is gratefully acknowledged. The author would also like to personally convey his gratitude to Dr. Lee Han Lim, Head of the Unit, for his suggestion on initiating the experiment and his support throughout its course.

REFERENCES

Cosgrove, J.B. & Wood, R.J. (1995). Probing and gorging responses of three mosquito species to a membrane feeding system at a range of temperatures. *Journal of the* American Mosquito Control Association **11(3)**: 339–342.

- Gubler, D.J., Nalim, S., Tan, R., Saipan, H. & Sulianti Saroso, J. (1979). Variation in susceptibility to oral infection with dengue viruses among geographic strains of Aedes aegypti. American Journal of Tropical Medicine and Hygiene 28(6): 1045–1052.
- Hagen, H.E. & Grunewald, J. (1990). Routine blood-feeding of Aedes aegypti via a new membrane. Journal of the American Mosquito Control Association 6(3): 535–537.
- Kuberski, T.T. & Rosen, L. (1977). A simple technique for the detection of dengue antigen in mosquitoes by immunofluorescence. *The American Journal* of *Tropical Medicine and Hygiene* 26(3): 533–537.