

# This Week's Citation Classic™

**Singh K R P.** Cell cultures derived from larvae of *Aedes albopictus* (Skuse) and *Aedes aegypti* (L.). *Curr. Sci. India* 36:506-8, 1967. [Virus Research Centre, Poona, India]

This paper describes the establishment of cell lines from the newly hatched minced larvae of *Aedes albopictus* and *Aedes aegypti* mosquitoes. Methods employed for subculturing up to the fifteenth passage level, as well as preliminary characterization of the cell lines, are given. [The SCI® indicates that this paper has been cited in over 230 publications since 1967, making it the most-cited paper ever published in this journal.]

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"It is unfortunate that my predecessor K.R.P. Singh, the author of the cited paper, died within a decade of its publication. At the request of the director of the National Institute of Virology, Khorshed Pavri, I am writing this commentary.

"In the early 1960s, soon after joining this institute, Singh began studies on the vector virus relationship of some of the arboviruses which were important to public health. He realized the importance of arthropod tissue culture in studies on arboviruses and received full encouragement from T. Ramachandra Rao, then director of the institute, and Charles R. Anderson, the chief scientific representative of the Rockefeller Foundation. Singh was awarded a Rockefeller Foun-

dation fellowship to visit the leading cell culture laboratories all over the world to gain experience in this discipline. On his return, he devoted himself wholeheartedly to the problem and succeeded quickly in establishing cell lines from *Aedes albopictus* and *Aedes aegypti*. The paper was sent to *Nature* for publication, but was returned. It was then sent to *Current Science* where it was promptly accepted.

"Singh's success was mainly due to the choice of his material, which consisted of *Aedes* eggs capable of withstanding dry conditions and the rigors of surface sterilization. They can be accumulated in large quantities to provide a sufficient amount of tissue. Above all, Singh seemed to have possessed a 'green thumb.'

"Singh's cell lines were the first continuous mosquito cell lines developed anywhere in the world. These were flown to Yale Arbovirus Research Unit in New Haven, Connecticut, where Sonja Buckley maintained a subline. Their distribution to scientists all over the world was undertaken from this institute and from Yale.

"Singh's *Aedes albopictus* cell line (ATC 15) found immense favor among arbovirologists because it was susceptible to several mosquito-borne arboviruses. Distinctive cytopathic effect with some flaviviruses facilitated their easy detection.<sup>12</sup> It is a rapidly growing cell line, requiring simple media and easy maintenance. Thus, Singh provided a tool which arbovirologists and cell biologists had been trying to obtain for a long time.

"Successful cloning of ATC 15 cells by Igarashi<sup>3</sup> gave a further boost to Singh's work. This clone is highly susceptible to both dengue and chikungunya viruses and is now popular all over the world.

"For his work, in 1968 Singh was awarded the Shakuntala Amir Chand Prize of the Indian Council of Medical Research."

1. **Singh K R P.** Growth of arboviruses in *Aedes albopictus* and *A. aegypti* cell lines. *Curr. Topics Microbiol. Immunol.* 55:127-33, 1971.
2. **Buckley S M, Hayes C G, Maloney J M, Lipman M, Aitken T H G & Casals J.** Arbovirus studies in invertebrate cell lines. (Kustak E & Maramorosch K, eds.) *Invertebrate tissue culture.* New York: Academic Press, 1976. p. 3-19.
3. **Igarashi A.** Isolation of a Singh's *Aedes albopictus* cell clone sensitive to dengue and chikungunya viruses. *J. Gen. Virol.* 40:531-44, 1978.