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SUKUNTHA SINGTHONG : CRYOPRESERVATION OF THIRD- STAGE LARVAE OF *ANGIOSTRONGYLUS CANTONENSIS* (THAILAND ISOLATE).
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The research objective was focused on finding the effective cryopreservation methods that yielded higher survival percentages of cryopreserved third-stage *Angiostrongylus cantonensis*. The experiment was performed by comparative use of 3 kinds of cryoprotectants: DMSO (dimethylsulfoxide), glycerol, combined DMSO and glycerol at concentrations of 5%, 10%, 15%, and 20% supplemented with 0%, 10%, 20%, and 40% fetal calf serum in the RPMI-1640 medium. The time for third-stage larvae (L_3) cryopreservation in liquid nitrogen at –196°C was 2, 4, 12, and 24 weeks. Four replications of the experiment were done in 4 Wistar rats. The L_3 used in the study were collected from laboratory infected snails, *Biomphalaria glabrata*.

The survival rate and infectivity of after-thawed cryopreserved L_3 were detected. Fifty surviving after-thawed cryopreserved L_3 were fed to a laboratory Wistar rat which was scarified after 50-55 days of post-infection, and the adult worms collected from the Wistar rat's lungs and heart were counted. The control Wistar rats group was fed with uncryopreserved L_3 that were collected from laboratory infected snails, with first-stage larvae (L_1) of *A. cantonensis* collected from laboratory infected Wistar rat feces.

The results showed that the optimum of cryopreservation method that yielded the highest percentage of survived cryopreserved L_3 was composed of RPMI-1640 medium mixed with 5% DMSO and 20% fetal calf serum per 100 L_3. The optimum time for cryopreservation in liquid nitrogen at –196°C was 2 weeks.

The infectivity of surviving after-thawed cryopreserved L_3 showed that there were 15, 9, and 11 adult worms collected from the lung and heart of infected Wistar rats that were fed with surviving after-thawed cryopreserved L_3 in 5% DMSO, 5% glycerol and 5% combined DMSO and glycerol cryoprotectants, and freezing in the liquid nitrogen at –196°C, for 2 weeks. There were no definitely surviving after-thawed cryopreserved L_3 that were frozen for 4, 12, and 24 weeks. The numbers of adult worms collected from the infected group and the control group were significantly different at ratios of 56.5%:7.5%; 64%:4.5%; 63%: 5.5% ($p < 0.0001$) using cryopreserved L_3 in 5% DMSO; 5% glycerol; and 5% combined DMSO and glycerol; respectively.