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Research No.(V):

Toxoplasma gondii: Prolonged in-vitro maintenance of virulent tachyzoites in fluid media at low temperatures

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Background: Prolonged maintenance of infective *Toxoplasma gondii* tachyzoites (*T.g.*T.) is an important subject for research purposes. This study aimed to evaluate four fluid media for prolonged in vitro maintenance of *T.g.*T.

Methods: The four fluid media Phosphate buffered saline (PBS) pH 7.2 and Roswell Park Memorial Institute (RPMI-1640) with or without 3% fetal bovine serum (FBS) were evaluated for maintenance of virulent T.g.T. The four media were tested after incubation at three different temperature degrees in the darkness.

Results: Prolonged maintenance period for infective *T.g.*T. was recorded especially in the absence of FBS supplement. RPMI without FBS was able to maintain infective *T.g.*T. for 16 days post incubation (dpi) at refrigerator temperature. This period decreased to 10 dpi and 6 dpi after incubation in the same media at 18–22 °C and 37 °C, respectively. Cultivation of *T.g.*T. in RPMI supplemented with 3% FBS and in PBS proved to maintain infective *T.g.*T. for 14 dpi at refrigerator temperature, and for 9 and 5 dpi when the two media were incubated at 18–22 °C and 37 °C, respectively. Shorter periods for keeping the *T.g.*T. infectivity were recorded using PBS supplemented with 3% FBS under all tested temperature conditions.

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Conclusion: This method allows economic long-lasting maintenance of tachyzoites for 16th dpi in RBMI that can be reactivated by reinoculation in mice.