

Invisible invaders: range expansion of feral *Neocaridina davidi* offers new opportunities for generalist intracellular parasites

Biological Invasions

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PCR settings for COI amplification with the LCO1490 and HCO2198 primers were as follows: initial denaturation at 94 °C for 60 s, five cycles of denaturation at 94 °C for 60 s, annealing at 45 °C for 90 s and extension at 68 °C for 90 s, followed by 40 cycles at 94 °C for 60 s, 50 °C for 90 s, 68 °C for 60 s, with a final extension at 72 °C for 5 min. Those used for the LCO1490-JJ and HCO2198-JJ primers were as follows: initial denaturation at 94 °C for 60 s; five cycles at 94 °C for 30 s, 45 °C for 90 s and 72 °C for 60 s, followed by 35 cycles with 94 °C for 30 s, 51 °C for 90 s and 72 °C for 60 s, with a final extension at 72 °C for 5 min. PCR settings for SSU rRNA amplification of microsporidians with the universal primers V1F and Micuni3R were as follows: initial denaturation at 94°C for 3 min, followed by 35 cycles at 94°C for 35 s and 68°C for 40 s, with a final extension at 68 °C for 5 min. Those for the EHP-specific primers ENF779 and ENR779 were as follows: initial denaturation at 94°C for 3 min, followed by 35 cycles at 94°C for 20 s, 58°C for 20 s, 72°C for 45 s, with a final extension at 72°C for 5 min. The PCR setting for the amplification of the spore wall protein of EHP (SWP1F - SWP1R and SWP2F - SWP2R primer pairs) were as follows: first step PCR with initial denaturation at 95°C for 5 min followed by 30 cycles at 95°C for 30 s, 58°C for 30 s, 68°C for 45 s, with a final extension at 68°C for 5 min; second step initial denaturation at 95°C for 5 min followed by 20 cycles at 95°C for 30 s, 64°C for 30 s, 68°C for 20 s, with a final extension 68°C 5 min.