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The aims of this study were to assess how the development of Ostertagia ostertagi eggs into first-stage larva (L1) affects the copy numbers of the Internal Transcribed Spacer region 2 (ITS2) of the ribosomal DNA; and based on these results, to suggest optimal storage conditions for faecal samples prior to detection and quantification by real-time quantitative polymerase chain reaction (qPCR) . Fresh O. ostertagi eggs were isolated from cattle faeces and stored at 4°C or 25°C under aerobic or anaerobic conditions. Embryonation was monitored by microscopy and the ITS2 copies were determined by qPCR at predetermined intervals for up to 336 h. Under aerobic conditions, L1 was observed after 24 h at 25°C, while development to L1 took 336 h at 4°C. A corresponding significant increase of the ITS2 copies was also observed ($p < 0.0001$). However, anaerobic conditions inhibited embryonation at both temperatures and no significant effect of storage on ITS2 copies was noticed ($p = 0.8984$). ITS2 copies were significantly higher in L1 compared with copies in unembryonated eggs ($p < 0.0001$) and with lower coefficients of variation for L1 (33%) compared with unembryonated eggs (266%). In conclusion, storage conditions affect the outcome of qPCR analysis for the quantitative determination of O. ostertagi eggs in cattle faeces. Cold storage at 4°C for up to 3 days or anaerobic vacuum packing at 25°C for up to 336 h will entail no undesirable effects on ITS2 copies.

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