

Extraintestinal *Entamoeba moshkovskii* Infection, Eastern India

Appendix

Appendix Table. List of primers used in this study. Primers adopted from published sources were used with the reported annealing temperatures, and the original protocols were followed unless specified otherwise. The chitinase primer set was optimized in-house and confirmed to show no cross-amplification with closely related species, including *E. histolytica* and *E. dispar*. PCR amplification was performed using Takara Ex-Taq, following the manufacturer's recommended protocol.

Primer Name	Sequence (5'→3')	Target gene/region	Amplicon Size (bp)	Annealing Temp (°C)	Tm (°C)	Reference
EH1	GTACAAAATGGCCAATTCATTCAAT G	18S rRNA	135 bp	50 °C	54 °C	(1)
EH2	ACTACCAACTGATTGATAGATCAG	18S rRNA			52 °C	(1)
EM_779bpNF	AACTAACGAAGGAGATGAAGTGAG	18S rRNA	779 bp	52 °C	55 °C	(2)
EM_779bpNR	GCCAGAGACATCGATTTAAAATG	18S rRNA			52 °C	(2)
EmChitinase_F	TGTTGTTTGGAGAAATGAAAAGG	Chitinase gene upstream region	480 bp	50 °C	51 °C	This study
EmChitinase_R	TTGATGTTGCCCTCACTGAC	Chitinase gene downstream region			52 °C	This study

References

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2. Sardar SK, Ghosal A, Haldar T, Maruf M, Das K, Saito-Nakano Y, et al. Prevalence and molecular characterization of *Entamoeba moshkovskii* in diarrheal patients from Eastern India. PLoS Negl Trop Dis. 2023;17:e0011287. [PubMed <https://doi.org/10.1371/journal.pntd.0011287>](https://doi.org/10.1371/journal.pntd.0011287)