Simultaneous genotyping of snails and infecting trematode parasites using high-throughput amplicon sequencing.

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Abstract

Several methodological issues currently hamper the study of entire trematode communities within populations of their intermediate snail hosts. Here we develop a new workflow using high-throughput amplicon sequencing (HTAS) to simultaneously genotype snail hosts and their infecting trematode parasites. We designed primers to amplify 4 snail and 5 trematode markers in a single multiplex PCR. While also applicable to other genera, we focused on medically and economically important snail genera within the Superorder Hygrophila and targeted a broad taxonomic range of parasites within the Class Trematoda. We tested the workflow using 417 *Biomphalaria glabrata* specimens experimentally infected with *Schistosoma rodhaini*, two strains of *Schistosoma mansoni*, and combinations thereof. We evaluated the reliability of infection diagnostics, the robustness of the workflow, its specificity related to host and parasite identification, and the sensitivity to detect co-infections, immature infections, and changes of parasite biomass during the infection process. Finally, we investigated the applicability of the workflow in wild-caught snails of other genera naturally infected with diverse trematode assemblages. After stringent quality control the workflow allows the identification of snails to species level, and of trematodes to taxonomic levels ranging from family to strain. Our HTAS workflow is sensitive to detect immature infections and changes in parasite biomass described in previous experimental studies. Co-infections were successfully identified, opening the possibility to examine parasite-parasite interactions such as interspecific competition. Altogether, these results demonstrate that our HTAS workflow provides a powerful tool to analyze the processes shaping trematode communities within natural snail populations.