Detection of submicroscopic soil-transmitted helminth infections from fecal samples in Nangapanda, Ende, using real-time polymerase chain reaction

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Abstrak

Deteksi Infeksi Submikroskopis Necator americanus, Ancylostoma duodenale, dan Ascaris lumbricoides dari Sampel Feses di Nangapanda, Ende, Menggunakan Real-Time Polymerase Chain Reaction. Infeksi dari Soil-Transmitted Helminthes (STH) (N. americanus, A. duodenale (Hookworm), dan A. lumbricoides) dapat menyebabkan anemia, kekurangan zat besi, bahkan malnutrisi. Pemeriksaan infeksi STH dapat dilakukan menggunakan mikroskop, tetapi metode tersebut masih kurang sensitif. Penelitian bertujuan mendeteksi dan mengetahui persentase infeksi submikroskopis STH dari sampel feses anak (usia 5-18 tahun) di Nangapanda, Ende menggunakan metode real-time polymerase chain reaction (PCR). Sampel feses dikoleksi sebanyak dua kali, yaitu sebelum dan sesudah pemberian albendazole 400 mg. Total sampel yang diperoleh adalah 242 tetapi hanya 45 sampel yang negatif secara mikroskopis yang diuji dengan real-time PCR. DNA sampel diisolasi dan diamplifikasi menggunakan primer dari daerah internal transcribed spacer (ITS-1 dan ITS-2) rDNA. Deteksi dengan real-time PCR menghasilkan kurva amplifikasi pada fluorophore VIC, FAM, dan Texas Red. Sebanyak tiga sampel (6,7%) pada pre treatment termasuk low load of DNA (N. americanus and A. lumbricoides) (Ct > 35), empat sampel (9,1%) termasuk low load of DNA untuk N. americanus saja (Ct > 35), dan lima sampel (11,4%) termasuk moderate load of DNA untuk A. lumbricoides saja (30 < Ct < 35) pada post treatment. Hasil penelitian menunjukkan bahwa real-time PCR dapat mendeteksi infeksi submikroskopis dari Hookworm dan A. lumbricoides.
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Soil-transmitted helminth (STH) infections (Necator americanus (hookworm), Ancylostoma duodenale (hookworm), and Ascaris lumbricoides) can lead to anemia, malnutrition, and iron deficiency. Traditionally, STH infections have been diagnosed using microscopy to detect eggs in human fecal samples. However, there are several limitations of this method. The aim of this research was to detect the percentage of submicroscopic STH infections from human fecal samples (children, 5?18 years old) in Nangapanda, Ende, using the real-time polymerase chain reaction (PCR) method. The fecal samples were collected in two time periods, which were before and after treatment, using 400 mg of Albendazole. There were 242 samples in total, but only 45 negative samples from microscopic detection were tested with real-time PCR. The DNA samples were isolated and amplified wih primers of internal transcribed spacer (ITS-1 and ITS-2) region of rDNA. The detection of samples with real-time PCR generated an amplification curve in VIC, FAM, and Texas Red fluorophore. Three samples (6.7%) in pre-treatment were low load of DNA (N. americanus and A. lumbricoides) (Ct > 35). Four samples (9.1%) were low load of DNA (N. americanus) (Ct > 35) in post-treatment. Five samples (11.4%) were moderate load of DNA (A. lumbricoides) (30 < Ct < 35) in post-treatment. real-time PCR could detect submicroscopic infections from specific species of hookworm and A. lumbricoides.