

# Detection of *Onchocerca volvulus* in Latin American black flies for pool screening PCR using high-throughput automated DNA isolation for transmission surveillance

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## Abstract

The posttreatment entomological surveillance (ES) of onchocerciasis in Latin America requires quite large numbers of flies to be examined for parasite infection to prove that the control strategies have worked and that the infection is on the path of elimination. Here, we report a high-throughput automated DNA isolation of *Onchocerca volvulus* for PCR using a major Latin American black fly vector of onchocerciasis. The sensitivity and relative effectiveness of silica-coated paramagnetic beads was evaluated in comparison with phenol chloroform (PC) method which is known as the gold standard of DNA extraction for ES in Latin America. The automated method was optimized in the laboratory and validated in the field to detect parasite DNA in *Simulium ochraceum sensu lato* flies in comparison with PC. The optimization of the automated method showed that it is sensitive to detect *O. volvulus* with a pool size of 100 flies as compared with PC which utilizes 50 flies pool size. The validation of the automated method in comparison with PC in an endemic community showed that 5/67 and 3/134 heads pools were positive for the two methods, respectively. There was no statistical variation ( $P < 0.05$ ) in the estimation of transmission indices generated by automated method when compared with PC method. The fact that the automated method is sensitive to pool size up to 100 confers advantage over PC method and can, therefore, be employed in large-scale ES of onchocerciasis transmission in endemic areas of Latin America.