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SHORT COMMUNICATION

Evaluation of a new *Strongyloides* ELISA IgG Test Kit

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SUMMARY

***Background: Strongyloides stercoralis* poses a significant clinical and public health challenge not only in endemic regions but also in non-endemic regions associated with travel. Ruling out this infection is essential in immunocompromised hosts given the high probability of reactivation and mortality.**
***Methods:* In this study we compared an ELISA based on two recombinant antigens in the United States with a highly sensitive and specific reference serological test.**
***Results & Conclusion:* There was 100% agreement between the two methods. ELISA assays based on *Strongyloides stercoralis* recombinant antigens has the potential to improve specificity. Further studies are warranted.**
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KEY WORDS

Strongyloides, NIE, SsIR, IgG

LIST OF ABBREVIATIONS

ELISA - Enzyme-linked immunosorbent assay
LIPS - Luciferase immunoprecipitation systems
HTLV - Human T cell Lymphotropic Virus
SsIR - *S. stercoralis* immunoreactive antigen
LPD - Laboratory of Parasitic Diseases
NIH - National Institutes of Health

INTRODUCTION

Strongyloides stercoralis is a parasitic nematode that is estimated to have infected between 30 and 100 million individuals worldwide [1]. This parasite has a potential to establish a long-term relationship with a given host but may also cause significant morbidity once reactivation and dissemination occurs in immunocompromised individuals. Climate change, faster and more convenient travel especially to endemic areas, and the expanding use of immunosuppressive and immunomodulatory therapies have all significantly paved the way for a large potential for reactivation and dissemination. To prevent this from occurring, serological screening is

performed in individuals currently receiving or being considered to receive immunosuppressants (especially corticosteroids), those infected with HTLV-1, and those who are candidates for transplantation, among others [2]. To this end, a highly sensitive and specific IgG test should be used to prevent both reactivation as well as unnecessary anti-parasitic therapy while withholding immunosuppressive/immunomodulatory treatment or transplantation. The currently-available *S. stercoralis* ELISAs in the United States are based on crude larval lysate of *S. ratti* and this has led to cross-reactivity with other nematodes such as filariae [3]. Two specific antigens of *S. stercoralis*, a 31-kDa recombinant antigen (termed NIE) and SsIR have been successfully incorporated in the LIPS assay for serodiagnosis of *Strongyloides* infection with sensitivity and specificity of 100% measured against the gold standard methods [3-5].

MATERIALS AND METHODS

In this study, we evaluated a new qualitative ELISA kit that has recently become available in the United States (*Strongyloides* ELISA IgG test kit; Gold Standard Diagnostics, Davis, CA, USA). A total of 44 de-identified sera (20 positive and 24 negative for *Strongyloides* IgG antibody) were kindly provided by LPD at the NIH. No further well-characterized samples were available for testing. Sera used for this study were previously collected from patients with multiple negative routine O&P and blood smear results and also tested negative by an established serology assay for filarie at LPD (data not shown). In brief, these sera were from patients with routine ova and parasite (O&P) positive results for *Strongyloides* and also tested positive using the aforementioned LPD's LIPS assay. All negative samples were from patients with previously negative O&P results and had previously been tested negative by the LIPS assay for both *Strongyloides* and Filariae. LIPS assay is routinely offered by LPD as a diagnostic test. The specimens were received by the Immunopathology Laboratory at the Cleveland Clinic and tested using the new ELISA kit. This test is an indirect ELISA. Per manufacturer's instructions, serum (1:100 dilution) was added to microtiter wells coated with recombinant NIE-SsIR fusion protein. After incubation and washing, adding protein A/G-conjugated horseradish peroxidase was followed by incubation, washing steps, substrate addition, and reading at 450 nm. Using kit calibrators, absorbance readings were converted to index values: < 9, 9 - 11, and > 11, as negative, equivocal, and positive, respectively.

RESULTS & CONCLUSIONS

All known positive and known negative sera from LPD tested positive and negative using the new kit, respectively, yielding a 100% concordance with the LPD method that had previously shown 100% sensitivity and specificity [4]. Given the fact that current *S. stercoralis* ELISA assays are based on the parasite's whole larval lysate and that many nematodes especially filariae share antigen with *S. stercoralis*, the possibility of cross-reactivity is high. This can be problematic in immunocompromised patients as it may change the course of management and cause undue frustration for patients and clinicians. Since this test is also done as a screen for previous *S. stercoralis* infection prior to institution of immunosuppressive or immunomodulatory therapies, any false positive result can unnecessarily withhold those therapies and may have adverse consequences for the patients. Given the 100% concordance with LPD results (as a surrogate gold standard), this test suggests a very high level of sensitivity and specificity. Further studies using larger panel of sera from patients directly tested by gold standard methods is warranted.

Ethics:
This study was deemed as exempt research by the Cleveland Clinic institutional review board (#19-1616).

Declaration of Interest:
None declared.

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No specific funding was available for this project. The kits were purchased as part of test validation and quality improvement process.

Contributions:
KK: design, analysis, writing; BGJ: testing, manuscript review.

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