

Measurement of ASCA, Lactoferrin and *Clostridium Difficile* in Feces of Patients with IBD

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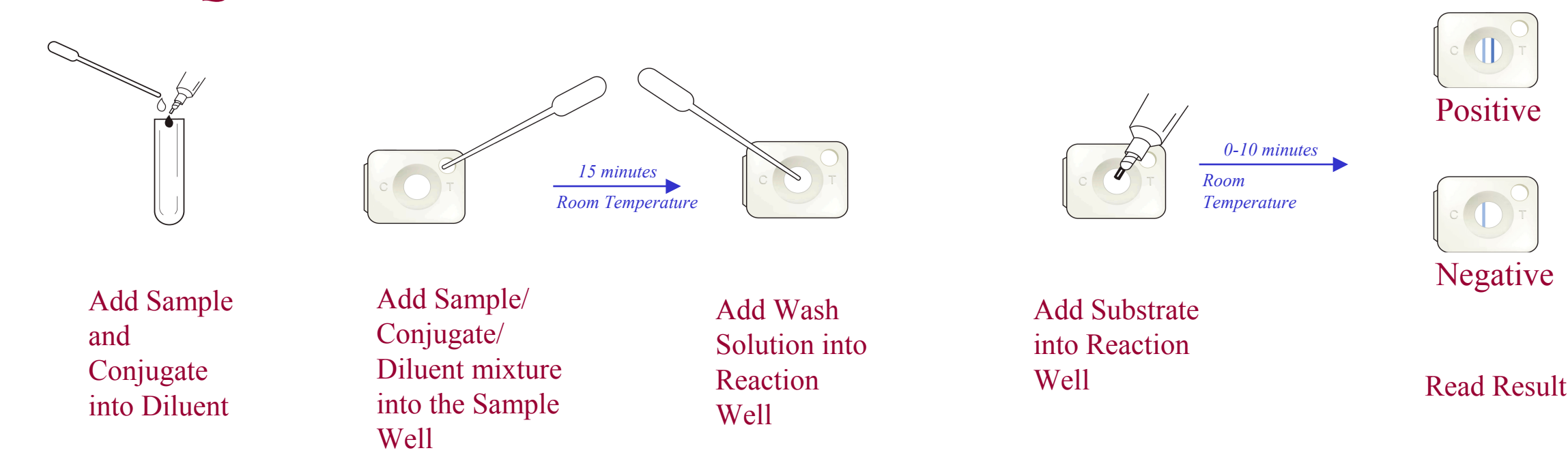
INTRODUCTION

Medical care for patients with inflammatory bowel disease (IBD) presents an ongoing challenge to gastroenterologists, posing a greater need for rapid diagnostics for the management of disease. The clinical assessment for confirmation of disease becomes even more difficult with the need to differentiate periods of disease activity from episodes of irritable bowel syndrome (IBS) and potential enteric infections that can mimic flares of IBD. Elevated fecal lactoferrin has been shown to be a sensitive and specific indicator of intestinal inflammation and the presence of fecal ASCA is correlated with Crohn's disease for both adult and pediatric IBD. Recent studies have shown *Clostridium difficile* (Cdiff), a nosocomial pathogen that causes antibiotic-associated diarrhea and pseudomembranous colitis, to be the most frequently identified pathogen in cases of relapsing IBD (10 to 20%). The aim of our study was to evaluate clinically characterized Crohn's disease (CD) subjects for the presence of fecal ASCA, measure lactoferrin (Lf) as an indicator of disease activity and to screen for the presence of Cdiff.

METHODS

- A single fecal specimen was collected from each of 23 consecutively recruited adult outpatients with clinically characterized Crohn's disease. The specimens were kept frozen until tested by a panel of rapid diagnostic tests for ASCA, elevated lactoferrin, Cdiff glutamate dehydrogenase (GDH; common antigen for Cdiff), and toxins A and B (toxigenic Cdiff). Test results were not linked to the diagnosis of Cdiff disease.
- IBD-SCAN** – Quantitative polyclonal-based ELISA for the measurement of fecal lactoferrin.
- ASCA-CHEK** – Qualitative ELISA for the detection of fecal ASCA.
- TOX A/B II** – Qualitative ELISA for Cdiff toxins A and B.
- C.DIFF CHEK** – Qualitative ELISA for GDH.
- TOX A/B QUIK CHEK™** - A new rapid membrane test for Cdiff toxins A and B.

TOX A/B QUIK CHEK™

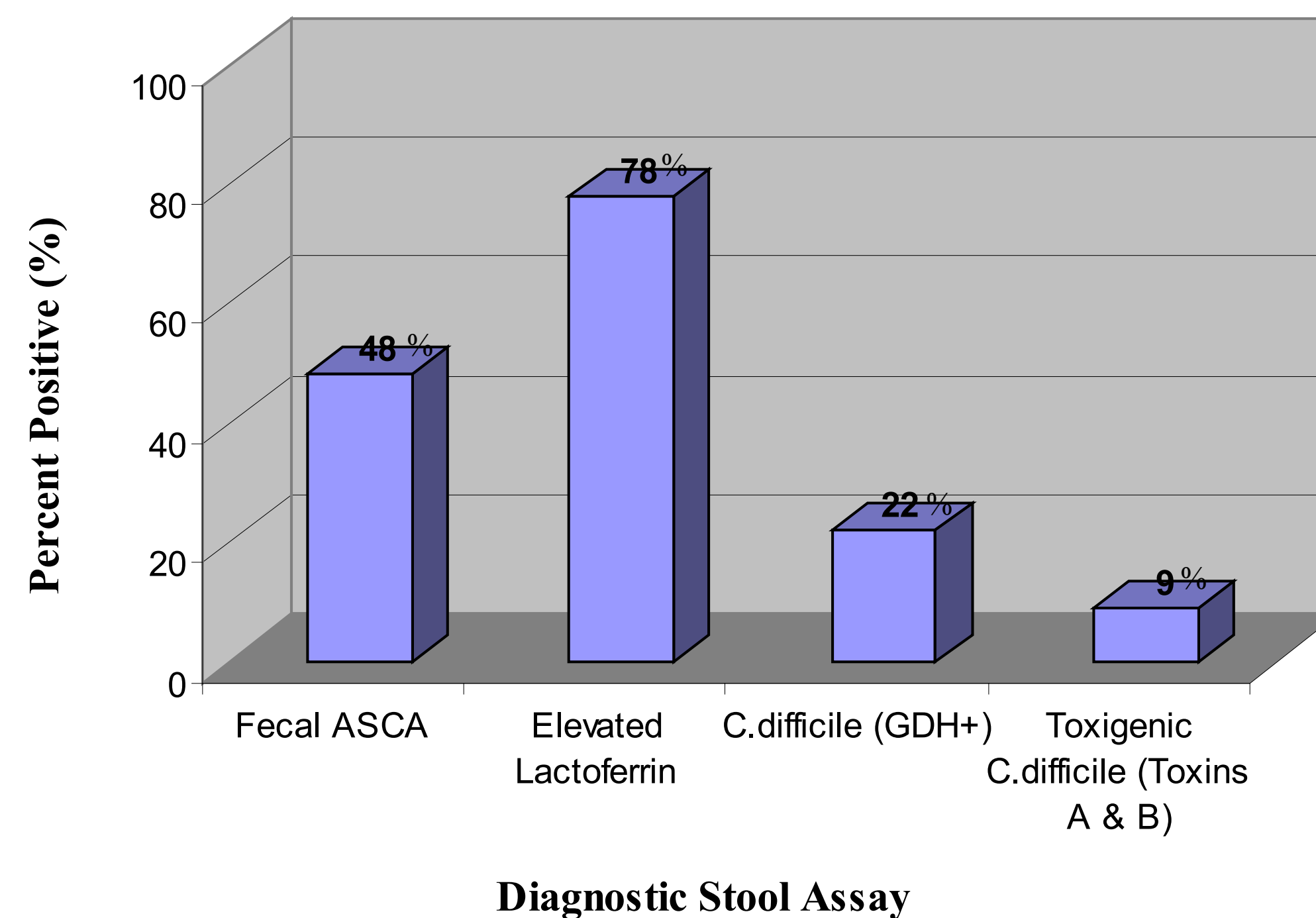


- TOX B Test** - Tissue culture assay using human foreskin cell monolayers and toxin B neutralizing sera.
- PCR analysis** – Direct PCR on fecal DNA for GDH, toxin A and B genes (GDH, *TcdA* and *TcdB*)

RESULTS I

Figure 1. Percent of Positive Diagnostic Test Results for Crohn's Disease Patients

Positive Test Results for Crohn's Disease Patients



RESULTS II

Table 2. Comparison of Diagnostic Results for patients with Cdiff-positive fecal specimens

Subject #	ASCA ELISA	Lactoferrin ELISA	Cdiff (GDH) ELISA	Cdiff (toxins) ELISA&Tissue Culture	Fecal DNA PCR
S1	Positive	168 µg/mL	Positive	Negative	Negative
S2	Positive	58 µg/mL	Positive	Negative	GDH+
S3	Negative	2 µg/mL	Positive	Negative	GDH+
S4	Negative	35 µg/mL	Positive	Positive	GDH+/Toxin+
S5	Negative	69 µg/mL	Positive	Positive	GDH+/Toxin+

Table 3. Clinical Data for patients with Cdiff-positive fecal specimens

Subject #	Disease Location	Disease Phenotype	History of Antibiotics	Immune Modulators	Clinical Signs Cdiff
S1	Ileocolonic	Inflammatory	No	Azathioprine	No
S2	Ileocolonic	Inflammatory	Bactrim	No	No
S3	Small bowel	Fibrosenosing	No	No	No
S4	Colon	Perforating	Flagyl / Augmentin	Azathioprine	No
S5	Ileocolonic	Inflammatory	No	Azathioprine	No

CONCLUSIONS

- Elevated lactoferrin can be detected in Crohn's disease patients with either small bowel and colonic disease.
- Our results support an incidence rate of 10% for toxigenic Cdiff in an IBD patient population.
- Toxigenic Cdiff should be considered during the assessment of patients with Crohn's disease.

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Table 1. Summary of Lactoferrin Levels for Different Disease Locations

Disease location	No.	Mean ± SE lactoferrin level (µg/mL)	Range
Ileocolonic	8	148 ± 53 µg/mL	4 – 376 µg/mL
Small bowel	8	205 ± 72 µg/mL	1 – 1405 µg/mL
Small bowel & Upper GI	2	554 ± 393 µg/mL	56 – 554 µg/mL
Colonic	5	112 ± 50 µg/mL	3 – 492 µg/mL