Role of Fecal Lactoferrin as a Non Invasive Biomarker in Diagnosis of Inflammatory Bowel Disease and Assessment of Disease Activity

Thesis

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بسم الله الرحمن الرحيم

قالُوا سُبْحَانكَ لاَ
عِلمَ لَنَا إِلاّ ما
عَلّمْتَنَا إِنَّكَ أَنتَ
الْعَلِيمُ الْحَكيم

صدق الله العظيم
سورة البقرة آية (٣٢)

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List of Abbreviations

Abb.	Full word
5-ASA	5-aminosalicylate
5-HT	Serotonin
6MP	6-mercaptopurine
AIDS	Adult immunodeficiency syndrome
AIEC	Adherent and invasive *E. coli*
ASCA	Anti-Saccharomyces cerevisiae antibodies
AZA	Azathioprine
CBC	Complete blood cell
CD	Crohn's disease
CDEIS	Crohn’s disease Endoscopic Index Of Severity
COPD	Chronic obstructive pulmonary disease
CRH	Corticotropin-releasing hormone
CRP	C-reactive protein
CT	Computed tomography
DBE	Double balloon enteroscopy
DCs	Dendritic cells
ELISA	Enzyme linked immunosorbent assay
ESR	Erythrocyte sedimentation rate
FBC	Full blood count
FC	Fecal calprotectin
FDA	Food and Drug Administration
FGIDs	Functional gastrointestinal disorders
FL	Fecal Lactoferrin
FLA	Fecal Lactoferrin assay

List of Abbreviations **(Cont...)**

Abb.	Full word
FODMAPs	Fermentable oligo-, di-, and monosaccharides and polyols
GI	Gastrointestinal
HADS	Hospital anxiety and depression scale
HSCT	Hematopoietic stem cell transplantation
IBD	Inflammatory bowel disease
IBS	Irritable Bowel Syndrome
IBS-C	Irritable bowel syndrome costipation subtype
IBS-D	Irritable bowel syndrome diarrhea subtype
IBS-M	Irritable bowel syndrome mixed subtype
IFN	Interferon
IL	Interleukin
IPAA	Ileal-pouch anal anastomosis
JAK	Janus kinase
M2PK	M2-pyruvate kinase
MCV	Mean corpuscular volume
MDP	Muramyl dipeptide
MMPs	Metalloproteinases
MPOs	Myeloperoxidases
MRI	Magnetic resonance imaging
MSC	Mesenchymal stromal cells
MTX	Methotrexate
NCCDS	Landmark National Cooperative Crohn's Disease Study
NF	Nuclear Factor
NK	Natural killer cells
NOD2	Nucleotide-binding oligomerization domain containing 2

List of Abbreviations **(Cont...)**

Abb.	Full word
NSAIDs	Nonsteroidal anti-inflammatory drugs
pANCA	Perinuclear antineutrophil cytoplasmic antibodies
PEG	Polyethylene glycol
PHQ	Patient health questionnaire
PI-IBS	Post infectious irritable bowel syndrome
PMN	Polymorphonuclear leucocytes
PMN-e	Polymorphonuclear neutrophil-elastase
PRRs	Pattern recognition receptors
RAGE	Receptor of advanced glycation end products
SBFT	Small bowel follow through
SeHCAT	Selenium homocholic acid taurine
SERT	Serotonin reuptake transporter
SES-CD	Simple Endoscopic Score For Crohn's Disease
SIBO	Small intestinal bacterial overgrowth
SSRIs	Selective serotonin reuptake inhibitors
TCAs	Tricyclic antidepressants
Th	T-helper cell
TNF	Tumor necrosis factor
UC	Ulcerative Colitis
UCCIS	Ulcerative Colitis Colonoscopic Index of Severity
UCDAI	Ulcerative colitis disease activity index
UCEIS	Ulcerative Colitis Endoscopic Index of Severity
US	Ultrasonography
WBC	white blood cell
WGO	World Gastroenterology Organization

INTRODUCTION

Inflammatory bowel diseases (IBD) including ulcerative colitis (UC) and Crohn's Disease (CD) are chronic inflammatory disorders of the gastrointestinal tract identified by episodes of relapse and remission **(Ponder *et al*., 2013).**

IBD had uncertain etiology; it imagined to be caused by interplaying among different environmental, genetic and immunologic factors **(Jussila *et al*., 2012).** Previous studies revealed that different factors such as infectious diseases and nutrition during infancy, tonsillectomy, appendectomy, lifestyle factors and diet, domestic hygiene, refrigeration of food, time scales of socioeconomic evolution, drugs (nonsteroidal anti-inflammatory drugs [NSAIDs] and oral contraceptive pills), smoking, intestinal pathogens, and measles vaccination play a role in IBD **(Vahedi *et al*., 2009).**

UC involves primarily the colon, and the symptoms include continuous or repeated blood in the stool and diarrhea. In contrast, CD may involve the entire gastrointestinal tract including the small intestine, colon, esophagus, and stomach. Perianal lesions are frequently observed in patients with CD. A discrepancy between clinical symptoms and endoscopic severity is observed in the clinical setting. Thus, endoscopic assessment is critical for the management of IBD. The assessment of the extent and severity of the disease is also important for decision making in the medical treatment of IBD **(Naganuma *et al*., 2015).**

In adult population, chronic diarrhea is a relatively common condition and often poses a diagnostic challenge despite its frequency **(Limburg *et al*., 2000).** In fact, diagnosis based on initial history and physical examination occurred only in about one-third of all patients with chronic diarrhea. One of the most frequent causes of chronic diarrhea in adults is probably irritable bowel syndrome (IBS) **(Delvaux *et al*., 2003).**

Patients with IBD and irritable bowel syndrome (IBS) share many clinical symptoms, including abdominal pain, diarrhoea and generalized malaise. A considerable proportion of patients, especially those presenting in primary care, will have functional disorders with no need for invasive examinations. In many cases, IBD and IBS cannot be separated from each other exclusively on the basis of clinical symptoms. Until recently, colonoscopy was required to rule out IBD. However, more than half of patients with symptoms suggesting IBD will have negative endoscopy and be diagnosed with IBS **(Van de Vijver *et al*., 2012).**

Fecal measurements of potential biomarkers are noninvasive diagnostic tests for intestinal mucosal inflammation and may correlate well with disease activity. Several neutrophil-granular proteins released by activated neutrophils may constitute fecal markers of intestinal inflammation, including lactoferrin (LF), calprotectin (Cal), polymorphonuclear neutrophil-elastase (PMN-e), and lysozyme (Lys), with calprotectin and lactoferrin appearing to be the most promising surrogate biomarkers (**Langhorst *et al*., 2005).**

Lactoferrin is an iron-binding protein; it covers most mucosal surface and interacts with exocrine organs or substances including parotid, tears, vaginal discharge, articular synovia, and latex (**Kane *et al*., 2003).** It is a component of neutrophil granulocytes and activated in acute inflammation. Fecal lactoferrin increases significantly as infiltration of neutrophils in intestinal tracts **(Desai *et al*., 2007).** It is stable for 5 days in feces whenever repeating freeze thawing ***(*Joishy *et al*., 2009).**

Studies investigating whether FL can be used as a noninvasive marker to distinguish IBD from non inflammatory conditions, especially IBS have yielded variable results **(Kopylov *et al*., 2014).**

During inflammation, lactoferrin is released by the injured tissue and has been found to modulate inflammation and act in the defense against infections as a part of the innate immune system **(Angriman *et al*., 2007).** It is resistant to degradation and proteolysis, and unaffected by freeze thaw cycles, making it a useful biomarker **(Mendoza *et al*., 2009).** As such, it is an ideal marker for intestinal inflammation. However, like calprotein it is unspecific for CD and UC, but can distinguish active IBD from inactive IBD and irritable bowel syndrome **(Schoepfer *et al*., 2008).**

AIM OF THE WORK

The aim of this study is to assess the role of fecal lactoferrin as a non invasive biomarker in diagnosis of inflammatory bowel disease and assessment of disease severity.