Rapid Immunochromatographic Assay of IgM Antibodies to *Salmonella Typhi* in Diagnosis of Typhoid Fever in Adults

Thesis

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بسم الله الرحمن الرحيم

قالوا

سبحانك لا علم لنا
إلا ما علمتنا إنك أنت
العليم الحكيم

صدق الله العظيم

سورة البقرة الآية: ٣٢

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Contents

List of Abbreviations

DCA	Deoxycholate Citrate Agar.
DCS	Decreased Ciprofloxacin Susceptibility.
ELISA	Enzyme linked Immuno Sorbent assay.
Ig M	Immunoglobulin M.
Ig G	Immunoglobulin G.
IL 8	Interleukin 8
KD a	Kilodalton.
K/A	Alkaline/Acidic.
LPS	Lipopolysaccarides.
MDR	Multiple Drug Resistance.
MIO	Motility Indol Ornithine medium.
NPV	Negative predictive value
OMP Ag	Outer Membrane Protein Antigen.
PAMPs	Pathogen Associated Membrane Proteins.
PCR	Polymerase Chain Reaction.
PPV	Positive predictive value
Prt	Paratose synthetase gene.
S.typhi	*Salmonella enterica serotype typhi.*
S.S agar	Salmonella Shigella agar.
TLR	Toll Like Receptor.
TSI agar	Triple Sugar Iron agar.
Tyv gene	Tyvelose epimerize gene.
Via B gene	Virulence antigen B gene.

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I- INTRODUCTION

Typhoid fever is caused by Salmonella enteric serotype typhi (*S. typhi*) and remains a significant health problem in many developing countries. Estimates suggest an incidence rate of more than 21.5 million cases with 200,000 deaths worldwide annually. The estimated incidence of typhoid fever in Egypt ranges between 13 and 59 per 100,000 persons per year **(*Crump et al; 2004 and Srikantiah et al; 2006).***

Enteric fever is a potentially severe systemic febrile illness that is usually presented with non- specific symptoms and signs such as slow gradually progressive fever, nausea, vomiting, abdominal pain, malaise, headache, constipation then diarrhea and hepatospleenomegally. It can be complicated by serious complication such as intestinal hemorrhage, intestinal perforation in the distal ileum, septicemia, diffuse peritonitis, encephalitis and cholecystitis ***(Clark et al; 2010).***

As the clinical picture of typhoid fever usually is misdiagnosed, microbiologic culture of a blood sample is considered to be the gold test for the diagnosis of typhoid

fever even though it takes up to seven days for isolation of the causative organism ***(Parry et al; 2002)***.

Serological diagnosis using Widal test provides a cost and time efficient alternative for blood culture, however its performance remains unsatisfying with sensitivity of 77% among Egyptians using blood culture as a gold standard test and with the need for establishment of a local cut off titer prior to use which makes its interpretation more complicated ***(Wilke et al; 2002).*** Therefore a rapid accurate test with a performance comparable to that of blood culture would be desirable for both patient care and surveillance situations ***(Kawano et al; 2007).***

Assays that detect IgM antibodies to *S. Typhi* which develop early in acute typhoid fever suggesting current infection are more sensitive and specific than the Widal test, and can be performed more rapidly. In the absence of culture facilities, IgM antibody tests are more useful than Widal test in diagnosis of typhoid fever in endemic areas ***(Cheesbrough; 2006).***

The Enterocheck WB® test is a 15 minutes qualitative, sandwich immunoassay for the detection of IgM antibodies to lipopolysaccaride (LPS) specific to *S. typhi* in human serum/plasma or whole blood specimen.

Evaluation of this test as a diagnostic tool for typhoid fever in adults will provide a rapid detection method for diagnosis and interference with prevention of further disease complications ***(Anusha et al; 2011)***.

II- AIM OF THE STUDY

This study aims to evaluate the Enterocheck WB® test (immunochromatograghic assay for *S. typhi* IgM) in comparison to Widal test as a rapid diagnostic tool for typhoid fever in adults in terms of sensitivity ,specificity, positive predictive value (PPV) and negative predictive value (NPV) using the blood culture as the gold standard.

III- REVIEW OF LITERATURE

Salmonella is a gram-negative, non- endospore forming food-borne pathogen of the general group called enteric bacteria. Salmonella species cause typhoid fever and illnesses of varying severity, collectively called salmonellosis or Salmonella enterocolitis. The straight rod-shaped Salmonella cells measure 0.5–1.0 micrometer (μm) wide and 1–5 μm long. Most species are motile, using a single polar (at one end of the rod) flagellum. Salmonella grows on a variety of nutrients as a facultative anaerobe. The genus belongs to family Enterobacteriaceae, order Enterobacteriales, and class Gammaproteobacteria of the phylum Proteobacteria ***(Guibourdenche et al; 2010)***.

Typhoid Fever

Enteric fever is a potentially severe systemic febrile illness caused mainly by *Salmonella enterica* serotype Typhi (*S. typhi)* causing typhoid fever and *Salmonella enterica* serotype Paratyphi A, B, and sometimes C (*S.* Paratyphi A, B and C) causing paratyphoid fever which is similar but less severe disease compared with *S. typhi*. The estimate for global incidence of typhoid fever is more than 27 million cases worldwide each year resulting in an

estimated 217,000 deaths annually ***(Crump and Mintz; 2010).***

While in most developed countries the rates of typhoid fever throughout the first half of the 20th century has declined due to vaccinations and advances in public sanitation and hygiene, infection remains endemic in many developing countries and is usually associated with poor sanitation, reduced access to treated drinking water and poor food hygiene ***(Crump and Mintz 2010 and Nuhu et al; 2010).***

A- Epidemiology:

In contrast to the rich countries, typhoid fever remains an important cause of illness in the developing world. According to the last surveillance supported by the World Health Organization (WHO) *S. typhi* ranked as the third (8%) most prominent *Salmonella* spp. isolated in Africa after *S.* typhimurium (26%), and *S.* enteritidis (25%) ***(Galanis et al; 2006).***

Typhoid fever is considered an endemic disease in Mediterranean North African countries with estimated medium incidence of 10 to 100 cases per 100,000 persons. Egypt remains a country with intermediate incidence of

one to 100 per 100,000 cases of enteric fever/year ***(Afifi et al; 2005 and Connor and Schwartz 2005)*** **figure (1).**

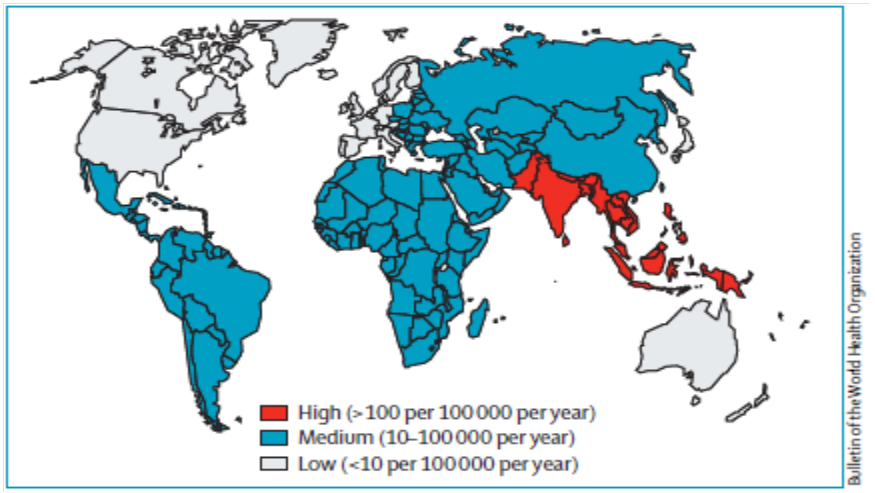

Figure (1): Geographical distribution of typhoid fever ***(Crump et al; 2004).***

Without effective treatment, typhoid fever is a life-threatening illness of several weeks duration with long-term morbidity often involving the central nervous system and has a case-fatality rate of 10-30%, with appropriate therapy typhoid fever is typically a short-term febrile illness requiring a median of 6 days of hospitalization and the mortality rate number is reduced to 1-4% ***(Crump et al; 2004).***