Influence of Helicobacter Pylori Infection on Plasma Ghrelin level in Patients with Upper GIT Lesions

Thesis

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By

Haytham Abdel-Rahman El-Naggar

M.B., B.Ch. (Faculty of Medicine, Cairo University)

Supervised By

Dr. ***Mohamed Shehata Abdalla***

Assistant Professor of Clinical and Chemical Pathology

Faculty of Medicine, Cairo University

Dr. ***Dalia Ibrahim Ramadan***

Lecturer of Clinical and Chemical Pathology

Faculty of Medicine, Cairo University

Dr. ***Ahmed Amin Haekal***

Lecturer of Internal Medicine

Faculty of Medicine, Cairo University

FACULTY OF MEDICINE

CAIRO UNIVERSITY

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ABSTRACT

The stomach is the main source of circulating ghrelin. Gastric colonization by H. pylori showed to affect ghrelin dynamics. This fact is due to long-term persistent H. pylori infections leading to atrophic gastritis. Some authors reported that; the relation between H. pylori status and ghrelin level is uncertain.

So the aim of this study is to answer; whether H. pylori infection affects gastric ghrelin production and consequently alters plasma ghrelin concentration or not?

The research was performed between November 2008-August 2009. Patients were recruited from endoscopy unit of Kasr El-Aini Internal Medicine Hospital. All patients were subjected to history taking with exclusion criteria of; age <18 or >80 years, body mass index (BMI) >30 Kg/m^2, pregnancy, diabetes mellitus, cachectic state including cancer, systemic infection, thyroid and liver diseases, renal impairment and use of medications effective against H. pylori during the preceding three months, alcohol use, dug addiction and gastro-intestinal surgery.

Key Words:
History, Description, Genome structure, Cell structure and metabolism, Pathology

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LIST OF ABBREVIATIONS

ACTH	Adrenocorticotrophic hormone
AMPK	AMP-activated protein kinase
AN	Anorexia nervosa
BMI	Body mass index
CAMP	Cyclic adenosine monophosphate
CLO	Campylobacter-like organism
DMSO	Dimethyl sulphoxide
GABA	Gamma aminobutyric acid
GGDT	Ghrelin gene-derived transcript
GH	Growth hormone
GHRH	Growth hormone releasing hormone
GHS	Ghrelin receptor
GPCR	G-protein coupled receptors
H. pylori	Helicobacter pylori
HPLC	High performance liquid chromatography
HRPO	Horseradish peroxidase
IGF-I	Insulin like growth factor-I
IP3	Inositol triphosphate
L-NAME	L-nitro arginine methyl ester
MALT	Mucosa associated lymphoid tissue
MCFA	Medium chain fatty acids
MCT	Medium chain triglyceride
MTLRP	Motilin-related peptide
NPT	Near patient test
NPY	Neuropeptide Y
PAT	Phosphate acetyl transferase
PRL	Prolactin
P-VALUE	Probability
PWS	Proder-Willi syndrome
RIA	Radio-immunoassay
SD	Standard deviation
TNF	Tumor necrosis factor
UBT	Urea breath test

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Introduction and Aim of Work

INTRODUCTION AND AIM OF WORK

Helicobacter pylori is a gram negative bacteria known to be a major pathogenic factor in the development of gastritis, peptic ulcer disease and gastric malignancy. Attachment of helicobacter pylori to the gastric mucosa induces inflammation which is associated with the release of various cytokines including IL-1B. It has been reported that helicobacter pylori infection could modify the plasma and gastric ghrelin dynamics in Mongolian gerbils. In humans, however helicobacter pylori has been reported not to be associated with any changes in plasma ghrelin levels, although eradication of helicobacter pylori has been shown by some to be associated with increase of plasma ghrelin levels.

Ghrelin is a 28 amino acid peptide recently identified in the stomach as an endogenous ligand for growth hormone secretagouge receptor. It potentially stimulates growth hormone release but is also implicated in many other homeostatic mechanisms ghrelin influences appetite, energy balance, gastric motility and acid secretion. This hormone is produced by X/A like endocrine cells of the oxyntic glands as well as pituitary, hypothalmus, colon, kidney, placenta, ovary and testes. Ghrelin mesenger ribonucleic acid (mRNA) is mostly highly expressed in the stomach compared with other tissues. Plasma ghrelin level decreases by as much as 65% after gastrectomy. Collectively stomach is the main source of circulating ghrelin. Most cases of chronic gastritis are due to H. pylori infection. Long persistent gastric mucosal inflammation induced by this organism results in progressive atrophy with loss of pyloric and oxyntic glands. It is tempting to speculate that the inflammatory and atrophic events

associated with H. pylori infection negatively affect ghrelin production in the stomach and its release into the ciruclation. There are however contradictory reports in the human beings on the relationship between H. pylori and ghrelin. A Turkish study reported lack of effect of H. pylori on plasma ghrelin levels, whereas a British study demonstrated an increase of circulating ghrelin following cure of H. pylori.

This study was designed to further investigate this issue.

Aim of the Work:

The **aim** of this work is to study the influence of H. pylori infection on serum ghrelin level in patients with upper GIT lesions. This was achieved through recruitment of a suitable number of patients complaining of abdominal pain and dyspepsia. They were subjected to the falling:

- History taking to exclude the following:
  Age <18 and >80, pregnancy, DM, systemic infection, thyroid and liver disease, renal impairment, use of medications effective against H. pylori during the last 3 months, alcohol use, drug addiction, NSAID use, gastric surgery, cachectic state.
- Blood samples were withdrawn from patients after about 12 hrs fasting for detection of ghrelin, H. pylori IgG antibody, liver functions (AST, ALT, albumin), kidney function (creatinine), thyroid function (TSH), lipid profile (T. cholesterol, triglyceride) and a fasting blood glucose.
- Endoscopic biopsies were taken from antrum and corpus of the stomach and were subjected to histopathological examination by Hx & eosin also giemsa staining for detection of helicobacter pylori bacteria.

Review of Literature

HELICOBACTER PYLORI

HISTORY:

Timeline of peptic ulcer disease and Helicobacter pylori:

In 1875, German scientists found helical shaped bacteria in the lining of the human stomach. The bacteria could not be grown in culture and the results were eventually forgotten. In 1893, the Italian researcher Giulio Bizzozero described helical shaped bacteria living in the acidic environment of the stomach of dogs **(Bizzozero and Giulio, 1893)**.

Professor Walery Jaworski of the Jagiellonian University in Kraków investigated sediments of gastric washings obtained from humans in 1899. Among some rod-like bacteria, he also found bacteria with a characteristic helical shape, which he called Vibrio rugula. He was the first to suggest a possible role of this organism in the pathogeny of gastric diseases **(Jaworski, 1899)**.

The bacterium was rediscovered in 1979 by Australian pathologist Robin Warren, who did further research on it with Barry Marshall beginning in 1981; they isolated the organisms from mucosal specimens from human stomachs and were the first to successfully culture them. In their original paper Warren and Marshall contended that most stomach ulcers and gastritis were caused by infection by this bacterium and not by stress or spicy food as had been assumed before **(Marshall and Warren, 1984)**.

The medical community was slow to recognize the role of this bacterium in stomach ulcers and gastritis, believing that no microorganism could survive for long in acidic environment of the stomach. The

community began to come around after further studies were done, including one in which Marshall drank a Petri dish of H. pylori, developed gastritis, and the bacteria were recovered from his stomach lining after a second endoscopy ten days after inoculation revealed signs of gastritis and the presence of "H. pylori". Marshall was then able to treat himself using a fourteen day dual therapy with bismuth salts and metronidazole. Marshall and Warren went on to show that antibiotics are effective in the treatment of many cases of gastritis.

In 1994, the National Institutes of Health (USA) published an opinion stating that most recurrent gastric ulcers were caused by H. pylori, and recommended that antibiotics be included in the treatment regimen.

Evidence has been accumulating to suggest that duodenal ulcers are also associated with H. pylori infection **(Pietroiusti et al., 2005).** In 2005, Warren and Marshall were awarded the Nobel Prize in physiology and medicine for their work on H. pylori.

Before the appreciation of the bacterium's role, stomach ulcers were typically treated with medicines that neutralize gastric acid or decrease its production. While this worked well, the ulcers very often reappeared. A very often used medication against gastritis and peptic ulcers was bismuth subsalicylate. It was often effective, but fell out of use, since its mechanism of action was a mystery. Nowadays it is quite clear that it is due to the bismuth salt acting as an antibiotic. Today, many stomach ulcers are treated with antibiotics effective against H. pylori.

The bacterium was initially named Campylobacter pyloridis, then C. pylori (after a correction to the Latin grammar) and in 1989, after DNA sequencing and other data showed that the bacterium did not belong in the

Campylobacter genus, it was placed in its own genus, Helicobacter. The name pylōri means "of the pylorus" or pyloric valve (the circular opening leading from the stomach into the duodenum), from the Greek word πυλωρός, which means gatekeeper.

While H. pylori remains the most medically important bacterial inhabitant of the human stomach, other species of the Helicobacter genus have been identified in other mammals and some birds, and some of these can infect humans **(Mobley et al., 2001)**. Helicobacter species have also been found to infect the livers of certain mammals and to cause liver disease **(Starzyñska and Malfertheiner, 2006)**.

DESCRIPTION:

Helicobacter pylori (H. pylori) is a Gram-negative organism that has a helical or spiral shape and has 6-8 flagella at one end. The size of the organism measures about 2-4 μm x 0.5-1.0 μm. H. pylori are found in a very acidic environments, at a pH of 2.0 or less. The bacterium has been cultured in microaerobic (low oxygen conditions) but it adapts to high oxygen at high culture densities. It is commonly found inside the lining of the stomach and the duodenum. H. pylori are slow growing organisms that can cause peptic ulcers and gastritis that can lead to gastric cancer and gastric MALT (mucosa-associated lymphoid tissue) lymphoma **(Blaser Martin, 1997)**.

H. pylori has a unique way of adapting in the harsh environment of the stomach. The inside of the stomach is bathed in about half a gallon of gastric juice every day. Gastric juice is composed of digestive enzymes and concentrated hydrochloric acid, which can readily tear apart the toughest food or microorganism. Bacteria, viruses, and yesterdays steak dinner are