

# The sequence of IL-1 $\beta$ gene in patients with *Helicobacter pylori* infections

Bushra Qasim Dhumad<sup>1\*</sup>, Wafaa Fadhil Hamad<sup>2</sup>

Department of Medical laboratories / College of health and medical techniques/ Baghdad<sup>1,2</sup>

Corresponding Author: 1\*



**ABSTRACT**— *Helicobacter pylori* is one of a risky bacteria that may lead to cancer. In this study, a total of 120 patients infected with *H. pylori* bacteria attended Baghdad Teaching Hospital during the period from 1<sup>st</sup> February to 15 September 2021. The Ages between (20-31) was more infected than other ages. The level of *H. pylori* among females 67 (55.3 %) was higher than males 53 (44.7%) with no significant difference ( $P>0.05$ ). The mean BMI  $\pm$  SD among the control group was ( $29.48 \pm 5.33$ ) and among patients was ( $27.99 \pm 4.55$ ), and the hypertension rate among patients was (23.3%) and among the controls was (16.4%). The prevalence rate of *Diabetes mellitus* among the patients was (13.7%), while it was (9.1%) among the controls, The thyroid dysfunction rate was (5.11%) among the patients, while it was (3.6%) among the controls. However, the renal/liver disease rate among the patients was (3.3%), but it was (9.1%). the association between SNPs of *IL1B* gene (T-31C) and infection with *H. pylori*. There was a significant association between *H. pylori* infection and T-31C ( $P<0.05$ ). There was a significantly higher frequency of TT genotype in the patient's group (50.4%) in comparison with the control group (38.88%), and there was an association between homozygotes TT state and the higher *H. pylori* infection risk in comparison with other genotypes. PCR was used for amplification of IL-1 $\beta$  gene, specific primers were used. The product was amplified with 240bp in region (TC; dbSNP: rs1143627).

**KEYWORDS:** Sequence, IL-1 $\beta$ , gene, *Helicobacter pylori*

## 1. INTRODUCTION

*Helicobacter pylori* is one of risky bacteria that may lead to stomach cancer [1], [2]. This bacterium may cause serious conditions to the intestinal lining, such as the stomach and intestines [3]. Diagnostic methods include cassette, and the sample is serum or stool after it is diluted and treated with special materials [4]. It is believed that *H. pylori* adhesion protein BabA2 plays an essential role in colonization of bacteria and in severe gastric inflammation induction, specially in combination with expressions of CagA & VacA [5]. There may be an association between *IL-1B* gene T-31C SNP and increased *H. pylori* infection risks among the Jordanian populations [6]. The virulent factors include cytotoxin-associated gene A (Cag-A) and vacuolating cytotoxin (Vac-A). Vac-A is a pore-forming toxin (PFT). Various other effects are used by Vac-A on target cells including apoptosis stimulation, mitochondrial function disruption and T-cell proliferation blockade so as to induce vacuolation [7]. Several studies demonstrated that IL-1B gene expression is usually affected by two allelic variants, IL-1B-31 & IL-1B-511, that are associated with the transcription of IL-1B [8]. A strong assembly was found between *H. pylori* and the host immune system during advance of gastrointestinal diseases, [9] where *H. pylori* stimulate the production of many proinflammatory cytokines, such as 1 beta (IL- 1 $\beta$ ), interleukin interleukin 10 (IL-10) and necrosis tumor factor-alpha [10]. The current study aimed to investigate whether the sequence of IL-1 $\beta$  gene in patients with *Helicobacter pylori* infections.

## 2. Materials and methods

*Helicobacter pylori* is one of risky bacteria that may lead to stomach cancer. A total of 120 patients infected with *H.pylori* attended Baghdad teaching Hospital during the period from 1<sup>st</sup> February to 15 September 2021. Stool samples were taken from the patients and each sample was kept in the cold box for detection of *H. pylori* bacteria by using serological tests. To amplify the *IL-1B* gene, for each primer pair, PCR reactions have been optimized with various annealing temperatures. The samples and standards were pipetted into the wells, and the present IL-1 $\beta$  were bound to the immobilized antibodies. Primers of *IL-1 $\beta$*  were used in Real-time to identify *Toxoplasma gondii* gene polymorphism, which was F: AGAAGCTTCCACCAATACTC & R: ACCACCTAGTTG TAA GGA with product 240bp.

## 3. Statistical analysis

The SPSS (16) program and Microsoft office excel were applied for statistical analysis. Numeric data were expressed as mean ( $\pm$ ) SEM (standard error of mean). The t-test, p-value <0.05 was considered as significant.

## 4. Results

Table (1) showed the prevalence of *H. pylori* with age groups. The highest rate of infection was among the age group (21-30) years (29.1%), but (31-40) years (24%), Sig. (P<0.05).

**Table (1):** Prevalence of *H. pylori* positive cases according to age groups

Age groups (Year)	<i>H. pylori</i>	%
7- 10	12	10
10 – 20	18	15
21 – 30	35	29.1
31 – 40	24	20
41 – 50	21	17.5
51-72	10	8.4
$\chi^2$ test (P-value)		P < 0.01 (HS)

The level of *H. pylori* among females 67 (55.3 %) was higher than males 53 (44.7%), with no significant difference (P>0.05) as shown in table (2).

**Table (2):** Prevalence of *H. pylori* positive patients according to gender

Gender	Patients	%
Male	67	55.3
Female	53	44.7

$\chi^2$  test (P-value) P> 0.05 (NS)

Table (3) showed that the mean BMI  $\pm$  SD among the control group was (29.48  $\pm$  5.33) and among patients was (27.99  $\pm$  4.55), and the hypertension rate among patients was (23.3%) and among the controls was

(16.4%). The prevalence rate of *Diabetes mellitus* among the patients was (13.7%), while it was (9.1%) among the controls, The thyroid dysfunction rate was (5.11%) among the patients, while it was (3.6%) among the controls. However, the renal/liver disease rate among the patients was (3.3%), but it was (9.1%) among the healthy persons ( $P < 0.01$ ).

**Table (3):** Demographic pictures of cases with *H. Pylori* infection

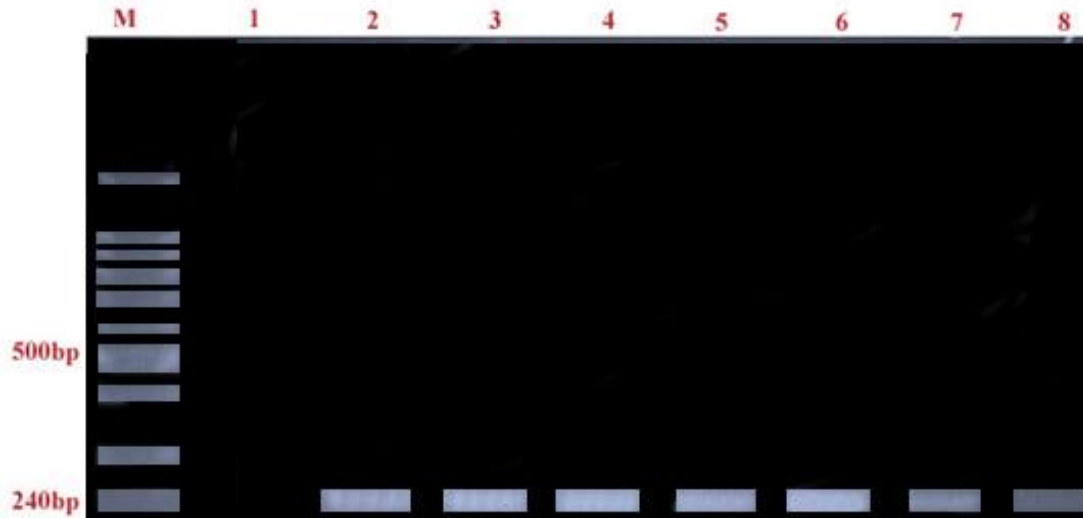
Parameters	Patients	Control
Mean BMI $\pm$ SD	29.48 $\pm$ 5.33	27.99 $\pm$ 4.55
Hypertension (%)	23.3%	16.4%
Diabetes mellitus	13.7%	9.1%
Thyroid dysfunction	5.11%	3.6%
Renal/liver diseases	3.3%	1.99%
$P < 0.01$ (HS)		$\chi^2$ test (P-value)

Table (4) showed the shearing with SNPs of *IL1B* gene (T-31C) and the infection. There was a shearing between the infection and T-31C ( $P < 0.05$ ). There was a significantly higher frequency of TT genotype in the patient's group (50.4%) compared with the control group (38.88%), and there was an association between homozygotes TT state and the higher injuries when compared with other genotypes.

**Table (4):** The association of *IL1B* SNPs with *H. pylori* infection

IL-1 $\beta$ SNP	Cases	Controls	Odds-Ratio	P- Value
T31C	N (%)	N (%)		
CC	(17.92)	(19.2)	1.00	0.043
TC	(31.91)	(41.91)	1.70 (1.09–2.66)	
TT	(50.4)	(38.88)	1.40 (0.81–2.44)	

The findings in figure (1) showed that among 60 patients infected with *H.pylori* who were genotyped, there was an amplified product of 240bp in region (TC; dbSNP: rs1143627). The mutation occurrence in IL-1 $\beta$  gene in the sequence of region T31C showed an association with *H pylori* infection.



**Figure (1):** Amplified products of IL-1 $\beta$  gene in region (TC; dbSNP: rs1143627) after electrophoresis on 1% agarose gel, Lane 1 Negative and Lane (2-8) positive product

The mutation occurrence in IL-1 $\beta$  gene in the sequence of region T31C showed an association with *H. pylori* infection as shows in figure (2).

```

AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG 31C>T
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG
AGCCTCCTACTTCTGCTTTTGAAAGCCATAAAAAACAG

```

**Figure (2):** The mutation occurrence in IL-1 $\beta$  gene in the sequence of region T31C showed an association with *H. pylori* infection

## 5. Discussion

*Helicobacter pylori* is one of a risky bacteria that may lead to stomach cancer. The prevalence rate of *H. pylori* with age groups showed that the highest rate was among The ages twenty to Thirty one was more infected than other ages. The results agreed with (SAID, M. Kh. 2019), [9], who reported that the participants within the age group Thirty one to fourty years represented the highest rate 57 (32.4%) of positive *H. pylori* infections ( $P = 0.031$ ). The level of *H. pylori* among females 67 (55.3 %) was higher than males 53 (44.7%), and these results were in disagreement with [10], who revealed a mean age of  $(44.46 \pm 13.45)$  years, with a rate of 49.9% ( $n = 2392$ ) to be males and a male-to-female ratio to be 1:1.01). This may be due to the geographical conditions inside Iraq, or the reason may be that Iraqi women are more susceptible to infection than men in the world, or there may be a demographic situation. The association of *H. pylori* with the seriousness of this bacteria with the disease states as well as with the weight were positive results when compared to the control group. [5] indicated that there is an increase in the levels of weight mass, as well as high pressure, and people with diabetes, kidney and liver diseases are directly proportional to infection with the injures. Also was shearing with SNPs of the *IL1B* gene (T-31C) with the injures [5]. This increases the virulence factors of bacteria and is more dangerous for infected patients [6].

The mutation occurrence in IL-1 $\beta$  gene in the sequence of region T31C showed an association with injuries. These findings were matched with [11] who showed that there are mutations in interleukin-1 beta in the C31T region, and this confirms the susceptibility to infection with pylori and the increase in the risk to the human body [12].

## 6. Conclusion

The sequence of IL-1 $\beta$  gene in patients with *Helicobacter pylori* might be associated with an enhanced danger of *H. pylori* infection among the Iraqi population.

## Acknowledgement

We would like to thank the administration of Baghdad Medical City/Educational Laboratories for their assistance in completing this work, as well as all of the volunteer participants who kindly provided the samples.

## 7. References

- [1] Bernard, M. and Josenhans, C. (Pathogenesis of *Helicobacter pylori* infection. *Helicobacter*, 19(Suppl.1): 2014): 11-18.
- [2] Stubljär, D. Jeverica, S. and Jukic, T. et al, The influence of cytokine gene polymorphisms on the risk of developing gastric cancer in patients with *Helicobacter pylori* infection, *Radiol Oncol*. 2015 Sep; 49(3): 256–264.
- [3] Vakil , N. *Helicobacter pylori* Infection, *HEALTH TOPICS & CHAPTERS*,2021: 6: 2021.
- [4] Kasirga, E. The importance of stool test in diagnosis and follow-up of gastrointestinal disorders in children, *Turk Pediatri Ars*. 2019; 54(3): 141–148.
- [5] Wen, S. Dominique Velin, D. and Felley, Ch. P. et al, Expression of *Helicobacter pylori* Virulence Factors and Associated Expression Profiles of Inflammatory Genes in the Human Gastric Mucosa, *Journal List Infect Immun* v.75(11); 2007 Nov, PMC2168299.
- [6] Shakhatreh, M. A. K. Khabour, O. F. and Alzoubi, K. H. et al, The Influence of IL-1B Gene Polymorphisms on *H. pylori* Infection and Triple Treatment Response Among Jordanian Population, *Appl Clin Genet*. 2020; 13: 139–145.
- [7] Saeidi, E. Sheikhaahrokh, A. Doosti, A. and Ranjbar, R., *VacA* Genotype in *Helicobacter Pylori*, *Home Books Bacteriology*, DOI: 10.5772/intechopen.81203.
- [8] Li, J. Xiaoxiao Sun, X. and Luo, Sh. et al, The Positivity Rate of IA-2A and ZnT8A in the Chinese Han Population With Type 1 Diabetes Mellitus: Association With rs1143627 and rs1143643 Polymorphisms in the IL1B Gene, *Front. Pharmacol.*, 11 2021 doi.org/10.3389/fphar.2021.729890.
- [9] 3. Kumar S, Dhiman M. Inflammasome activation and regulation during *Helicobacter pylori* pathogenesis. *Microb Pathog*. 2018;125:468–474. doi:10.1016/j.micpath.2018.10.012
- [10] 4. de Brito BB, da Silva FA, de Melo FF. Role of polymorphisms in genes that encode cytokines and *Helicobacter pylori* virulence factors in gastric carcinogenesis. *World J Clin Oncol*. 2018;9(5):83–89.

[11] SAID, M. Kh. PREVALENCE OF HELICOBACTER PYLORI INFECTION AMONG PATIENTS WITH PEPTIC ULCERS AND THE ASSOCIATED RISK FACTORS IN MBAGATHI LEVEL V HOSPITAL, NAIROBI COUNTY, KENYA, P150/CE/23142/2019.

[12] Wang, W. Jiang, W. and Zhu, Sh. et al, Assessment of prevalence and risk factors of helicobacter pylori infection in an oilfield Community in Hebei, China, Wang et al. BMC Gastroenterology (2019) 19:186.

[13] Alexander, S. M. Retnakumar, R. J. and Chouhan, D. et al, Helicobacter pylori in Human Stomach: The Inconsistencies in Clinical Outcomes and the Probable Causes, Front. Microbiol., 17: 2021 : doi.org/10.3389/fmicb.2021.713955.

[14] Ranjbar R. and Chehelgerdi M. Genotyping and antibiotic resistance properties of Helicobacter pylori strains isolated from human and animal gastric biopsies, Infection and Drug Resistance 2018:11 2545–2554.



This work is licensed under a Creative Commons Attribution Non-Commercial 4.0 International License.