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Gastric Tumor Suppressor Genes Alterations Associated with cag A Positive H pylori among Patients with Gastric Cancer Systemic Review and Meta-Analysis

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Abstract

Introduction: *Gastric cancer is the fifth most frequent cancer worldwide After lung, breast, colorectal, and prostate cancers. Helicobacter pylori (H. pylori) is considered the most important causative agent of gastrointestinal diseases such as peptic ulcer, gastritis, gastric adenocarcinoma, and mucosa-associated lymphoid tissue (MALT) lymphoma.*

Objective: *to identify the tumor suppressor genes alterations associated with CagA in patients with gastric cancer.*

Methods: *All the available papers published before 2022 were collected by searching in PubMed and Scopus. The keywords included in the research were "H.pylori", "gastric cancer", "virulence factors", "tumor suppressor genes" " gene mutations" "cagA+" used by Boolean operators to obtain the articles with the keywords in their titles or abstracts.*

Result: *Initial searches yielded 111 articles, four articles were excluded as a duplication using the computer program Zotero (v5), then one hundred and seven articles were screened for the title and abstract evaluation using the Rayyan website, among them seventy-one articles were excluded. Thirty-six articles were scanned for full-text review and eligibility, furthermore, twenty-five articles were excluded because there were either Reviews and case reports, Not relevant studies, Insufficient data, and Unclear methods and results. Eleven articles were included for the literature review. In addition, the studies were in different regions of the world including Asia, Europe, North America, and Latin America. However, most of the studies were related to the USA.*

Conclusion: *Cag A can cause alterations on gastric tumor suppressor genes by either decreased expression by increasing the methylation, inducing point mutation as mentioned, inactivation by increasing the methylation levels, increasing the levels of degradation and methylation the promotor of the tumor suppressor gene as mentioned.*

Keywords: H.pylori, Gastric Cancer, Virulence Factors, Tumor Suppressor Genes, Gene Mutations, cagA+

Introduction

Gastric cancer is the fifth most common cancer in the world. Following cancers of the lung, breast, colorectal, and prostate. Almost two-thirds of stomach cancer cases are found in East Asia, Eastern Europe, and South and Central America. GC affects over 80% of adults in developing countries, while it affects 20% of people under thirty and 50% of the elderly in developed countries [1]. Lauren has distinguished between the intestinal type and the diffuse type of gastric cancer[2,3]. The intestinal type of gastric cancer is well-differentiated, progresses gradually, and forms glands. Atrophic gastritis is frequently the first pathological change that leads to intestinal metaplasia, dysplasia, and eventually malignancy in the development of this type of gastric cancer [4]. The diffuse type of gastric cancer spreads quickly to distant organs and tissues without developing glands and grows aggressively throughout the stomach. Additionally, it is made up of tumor cells that have not undergone proper differentiation and produces mucus. The stomach wall thickens, hardens, and rubberizes due to a morphological variation of diffuse-type cancer that invades the muscles of the stomach wall with significant fibrosis. A type of diffuse stomach cancer called scirrhous gastric cancer, also known as linitis plastic, exists [5].

Barry Marshall and Robin Warren made the initial discovery

of Helicobacter pylori (H. pylori) in Australia in 1982. It is spiral-shaped, has flagella, and is an extracellular, microaerophilic bacterium that lives in the human stomach's submucosa [6]. It may spread through fecal-oral, oral-oral, or gastro-oral means between individuals [7]. Additionally, it is regarded as the primary cause of gastrointestinal illnesses like gastritis, gastric adenocarcinoma, peptic ulcer, and mucosa-associated lymphoid tissue (MALT) lymphoma [8,9].

Though not everyone infected with H. pylori develops gastric cancer, it is thought to be one of the risk factors. The Cytotoxin Associated Gene A (cagA), a protein with more than 1,200 amino acids that is inserted into epithelial cells lining the stomach by T4SS, is thought to be the virulence factor of H. pylori that causes an increase in cell proliferation [10-13]. By interfering with more than 20 host proteins, the cage can alter a variety of host cellular processes, including cytoskeletal organization, cell-to-cell adhesion, and intracellular signal transduction [14]. Many of the prooncogenic properties of cagA are thought to be primarily attributed to its C-terminal domain. [14-17]. Oncogenesis is a multifactorial series of events, as is well known. Oncogene activation and tumor suppressor gene activation are typically the two main factors in the majority of known cancers. CagA can influence cellular tumor suppressor genes and activate cellular protooncogenes [18-20].

Materials and Method

Search Strategies

All the available papers published before 2022 were collected by searching in PubMed and Scopus. The keywords included in the research were “H.pylori”, “gastric cancer”, “virulence factors”, “tumor suppressor genes” “ gene mutations” “cagA+” used by Boolean operators to obtain the articles with the keywords in their titles or abstracts.

All records were entered into the computer program Zotero to remove the duplicated articles or merge them. After that, the Rayyan website was used for the title and abstract screening. The included articles were downloaded for full-text screening and eligibility. The Statistical Package for Social Sciences (SPSS v26) was used for analysis.

Inclusion and Exclusion Criteria

All the case-control, cross-sectional, cohort studies, and the letter to the editors regarding the H. pylori infection, gastric cancer, and tumor suppressor genes were considered eligible and included in the study.

The eligibility of studies was determined after reviewing and evaluating the titles, abstracts, and full text of the studies. The included studies needed to be focused on the main idea, using standard methods including culture, urea breath test, immunohistochemistry PCR, Sequencing, and PCR-RFLP. However, the studies published in a non-English language, review articles, case reports, studies based on nonclinical samples, laboratory animals, and other autoimmune diseases were excluded

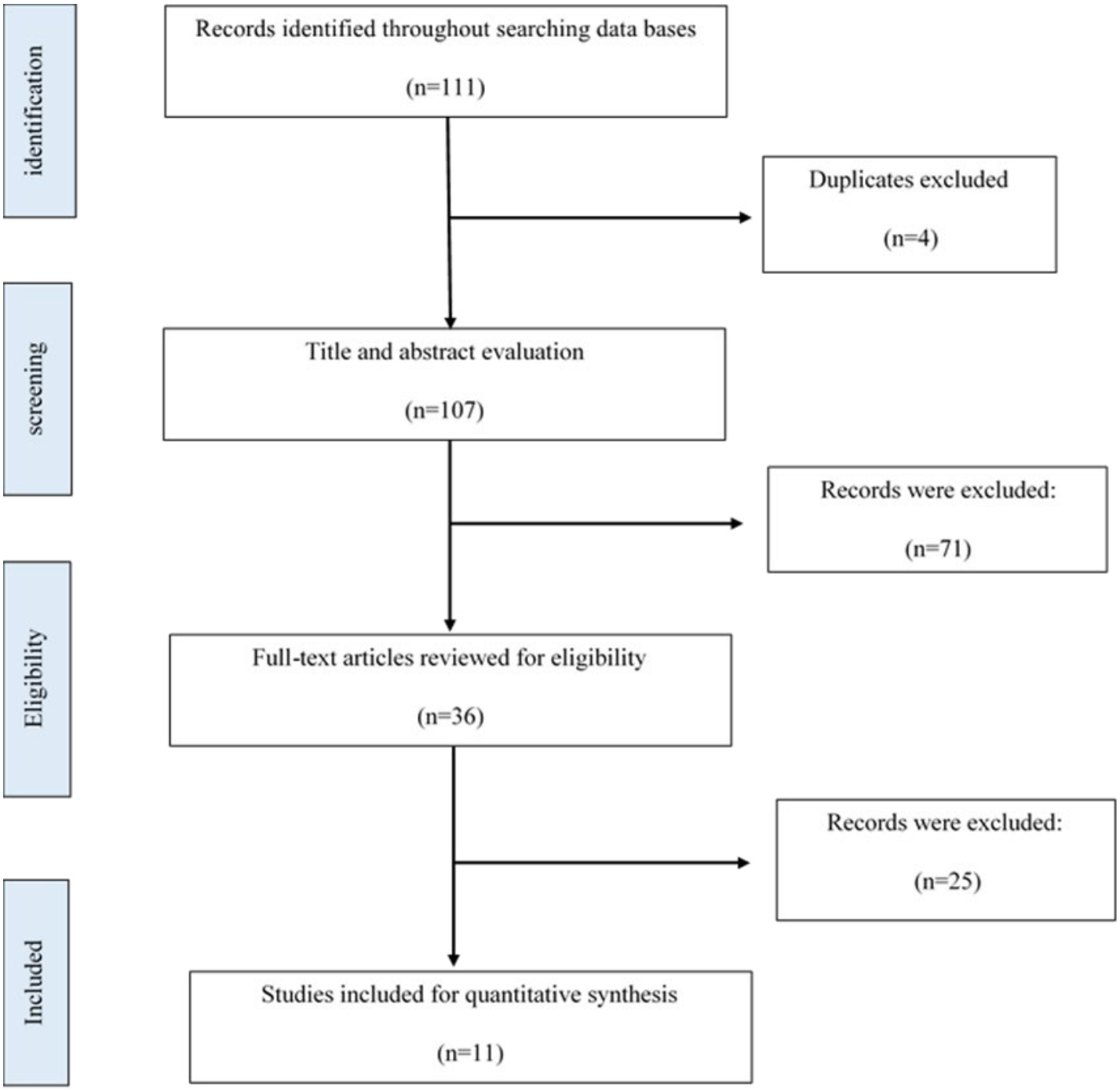

Figure 1: Flow Diagram Represents the Methodology of this Study and its Process

Background

H. pylori can trigger a strong immune response that results in inflammation of the gastric mucosa. Gastric cancer is largely caused by the failure to eradicate the organism from the stomach, where it causes a protracted infection [21]. However, it is a protracted process governed by the digestive environment, the host, and bacterial virulence factors, leading to gastric issues like peptic ulcer disease and gastric cancer [22]. H. pylori possesses numerous virulence factors, including Vacuolating cytotoxin A (VacA) the cytotoxin-associated gene pathogenicity island (cagPAI), an oncoprotein (i.e. cytotoxin-associated gene A (CagA)), and adhesion proteins, all of which are associated with the pathogenicity and development of gastric cancer [23,24]. The purpose of this review was to compile the most recent findings regarding changes in gastric tumor suppressor genes that are associated with CagA and contribute to the pathogenesis and development of gastric cancer.

Cytotoxin-Associated Gene Pathogenicity Island (cagPAI)

cagPAI contains about 40 kb of chromosomal DNA that contains about 32 open reading frames (ORFs), such as cag1 to cag26, cagA to cagZ [25]. cagPAI encodes the effector protein cagA and the syringe-like structure the bacterial type four (IV) secretion system (T4SS) which delivers the cagA into the epithelial cells of the stomach [26,27]. Although cagPAI integrity is required to encode intact T4SS for H. pylori contact with host cells, cagPAI is not present in all strains and is defective in some strains [28,29]. Infection with strains expressing the entire cagPAI has been linked to severe gastrointestinal problems such as chronic gastritis, peptic ulcer disease, and gastric cancer [30,31].

Cytotoxin-Associated Gene A (CagA)

H.pylori bacteria that can express this protein are thought to be very virulent, whereas strains that cannot express this protein are thought to be less virulent. This protein is approximately 125-145 kDa in length. T4SS assists in its translocation into gastric epithelial cells [29,32]. CagA is made up of a well-known five-amino-acid motif called EPIYA (glutamic acid-proline-isoleucine-tyrosine-alanine), which forms a sequence at the C-terminal region and, along with the nearby sequence, is critical for CagA's biological activity [24].

Depending on geographic variation, H. pylori strains can have four different EPIYA-sequences, namely EPIYA-A, -B, -C, and -D. CagA with the EPIYA-D sequence has a greater ability to de-regulate cellular activities than CagA with the EPIYA-C sequence. CagA containing the first two EPIYA-sequences (EPIYA-A and EPIYA-B) combined with the EPIYA-D sequence is thus considered more virulent than CagA containing the EPIYA-A, EPIYA-B, and EPIYA-C sequences [33].

CagA Translocation

CagA is translocated into gastric epithelial cells after H. pylori strains produce it, and at least 15 cagPAI-encoded proteins are involved in the formation of T4SS [34,35]. CagA is exposed on the bacterial surface via T4SS and interacts with PS patches on the plasma membrane of host cells that have been abnormally externalized due to H. pylori infection. CagA's N-terminal region interacts with the PS patches, causing the bound CagA to be flipped inside and internalized [36].

Tyrosine Phosphorylation

When CagA is delivered into gastric epithelial cells, its tyrosine (Y) residue in the EPIYA motifs is phosphorylated by cellular kinases such as Csk, Src family kinases (SFKs), and c-Abl [37-39]. CagA has the ability to bind promiscuously to the SH2 domain, which contains host proteins like the pro-oncogene Src homology 2 phosphatase (SHP2), PI3K, Crk, and the adaptor protein Grb2 [40-43].

Pylori CagA+ and Tumor Suppressor Genes

Apoptosis-Stimulating Protein of p53-2 (ASPP2) Tumor Suppressor

ASPP2 was initially discovered as a p53 binding protein, but it has now been shown to be a self-sufficient tumor suppressor that collaborates with p53 (and its family members p63 and p73) to inhibit tumor growth in vivo [44,45]. Furthermore, mounting evidence suggests that ASPP2's cellular roles include tight junction development and epithelial cell polarity maintenance. ASPP2 is downregulated in many aggressive tumors, including gastric cancers. Following Hp infection and CagA delivery, the level of ASPP2 increases. Furthermore, after Hp infection, CagA coimmunoprecipitates with ASPP2, altering its proapoptotic activity [46-54].

CagA injection enhances the interaction of p53 and ASPP2. Doxorubicin (Dox), a DNA-damaging chemical that activates p53, induced ASPP2 association with p53 as well as cell apoptosis. The link between CagA and ASPP2 was discovered 90 minutes after infection and grew stronger over time. In contrast, the relationship between ASPP2 and cytoplasmic p53 was discovered at 3 h and peaked at 7 h. This implies that when CagA interacts with ASPP2, the cytoplasmic pool of p53 is recruited.

P53 is a transcription factor that regulates gene expression. Although it is significantly stabilized following DNA damage or cellular stress, the proteasome degrades it quickly. Because ASPP2 binds cytoplasmic p53 during H. pylori infection, the transcriptional activity of p53 may be altered after CagA translocation. According to several lines of research, CagA-mediated suppression of p53 expression is ASPP2 dependent, whereas CagA-induced p53 degradation is mediated by the proteasome. Under normal conditions, the tumor suppressor activity of the ASPP2-p53 pathway is primarily mediated by stimulation of the apoptotic response. CagA, on the other hand, increases the association between p53 and ASPP2, resulting in increased p53 degradation and, as a result, transcriptional suppression in H. pylori-infected cells. CagA inhibits apoptosis by binding the tumor suppressor ASPP2, causing p53 to be damaged and its apoptotic activity to be suppressed [56].

Phosphatase and Tens in Homolog (PTEN)

PTEN (phosphatase and tensin homolog) is a tumor suppressor gene (810) found on chromosome 10q23. It has been discovered to regulate the protein kinase B (AKT) and mechanistic target of rapamycin signaling pathways, both of which are involved in apoptosis, cell cycle progression, and cell proliferation. PTEN deficiency has been linked to oncogenesis and somatic mutations in a number of cancers. Tet methylcytosine dioxygenase (Tet)1 has been found to interact with the p53enhancer of zeste 2 polycomb repressive complex 2 subunits (EZH2) signaling pathway to decrease tumors in gastric cancer. Tet1 inhibits cancer formation by activating p53 and inhibiting the carcinogenic protein EZH2, perhaps through DNA demethylation [57,58]. Zhang et al (59) recently reported that In human gastric cancer, the expression of PTEN was found to be dramatically reduced by CagA.

Cyclin-Dependent Kinase Inhibitor 2A-CDKN2A

E-cadherin and CDKN2A are two tumor suppressor genes whose promoter hypermethylation has been associated with H. pylori infection [59,60]. The tumor suppressor p16INK4A deletion has been implicated in the carcinogenic process in a number of malignancies [61–63]. It is common for p16INK4A to lose expression in GC, and hypermethylation of its promoter regions is thought to be the main factor in this gene's inactivation [64-66]. To the contrary, numerous earlier studies [67,68] claimed that promoter methylation and p16INK4A showed a strong correlation. The mechanism behind p16INK4A inactivation is unknown, despite the fact that it is acknowledged as a contributing factor to GC carcinogenesis. It is believed that H. pylori plays a significant role in this process [69, 70]. The H. pylori genotype has an impact on whether methylation or non-methylation mechanisms are used to inactivate p16INK4A, according to research by Zhang et al. [59]. Additionally, they noted that depending on where the tumor is located, different histological subtypes of GC exhibit different patterns of p16INK4A inactivation. Only in Nocardia tumors does methylation of the CDKN2A promoter render p16INK4A inactive in diffuse subtype cancers. In contrast, both cardia and non-cardia tumors exhibit promoter methylation, which is a crucial pathway for deactivating p16INK4A in intestinal cancers. Additionally, the methylation of the CDKN2A promoter is influenced by the H. pylori genotype.

P53 and p27

The p53 gene, which is located on the short arm of chromosome 17 (17p13.1), produces a protein that acts as a transcription factor and controls a number of physiological processes, including cell division, DNA damage response, apoptosis, and angiogenesis. The main transcriptional target of p53 is WAF1 (also known as CIP1, SDI1, mda-6, or CDKN1A). A phosphorylated 21-kDa protein with tumor-suppressing properties is encoded by the p2WAF1/CIP1 gene. P53 mutations are found in between 38% and 71% of gastric cancer tumors, making them relatively common [70,71].

Another CIP/KIP tumor suppressor protein is encoded by the p27KIP1 gene, which is located on chromosome 12p13. Because p27Kip1 Protein (p27) and p21 share a 42 percent structural similarity, this explains how their ability to inhibit the cyclin D/CDK4, cyclin E/CDK2, and cyclin A/CDK2 complexes to stop the progression of the cell cycle is similar [72,73]. Reduced or absent p27 protein expression is associated with more aggressive characteristics and tumor growth in people with gastric carcinomas [74-76]. Lower expression of p27 has been reported to be a predictor of aggressive behavior and a poor prognosis in a variety of malignant tumors, including breast, colon, liver, stomach, lung, brain, prostate, and malignant melanoma [77-80].

According to reports, H. pylori can result in a mutation in the p53 tumor suppressor gene, which in turn can lead to stomach cancer [81]. Although p53 alterations have been examined in several studies of gastric cancer associated with H. pylori infection [81–85], there is still some controversy. Furthermore, H. pylori in gastric cancer has been linked to decreased p27 expression [86,87]. However, there is no scientific agreement among studies, and there aren't any studies linking these two suppressor genes to H. pylori [89].

Fragile Histidine Triad (FHIT)

Is a tumor suppressor gene that can be found on chromosome 3p14.2. Early studies on a large number of gastric cancer samples discovered that the tumor suppressor protein fragile histidine triad (FHIT) was lost in the majority of cases (more than 70%), and that it was more common in GC with the diffuse and mixed histotypes than the intestinal histotype. Changes in FHIT gene expression have been found in primary tumors and cancer cells from the lung, breast, head and neck, esophagus, colon and rectum, pancreas, kidney, cervix, and hepatocellular carcinoma. The FHIT protein is involved in a variety of biological functions, including cell cycle regulation, DNA damage sensitivity, and pro-apoptotic signaling [90-95].

CDH1

CDH1 is a tumor suppressor gene that produces the E-cadherin protein, which is required for cell-cell interactions. Inactivating this gene increases the likelihood of metastasis. The methylation of the CDH1 promoter during the early stages of gastric carcinogenesis remains a mystery. It is found in epithelial cells and participates in cellular processes like adhesion, morphology, migration, and development. It plays a role in cellular processes such as adhesion, morphology, migration, and development, as well as in cell architecture and tissue integrity [96,97].

It has been found in a variety of cancers, including gastric cancer, and its inactivation has been linked to tumor growth via invasion and metastasis. CDH1 mutations were found in approximately 50% of diffuse histological type gastric carcinomas, and CDH1 hypermethylation was discovered to be the second source of gene expression inactivation in two families with familial stomach cancer and CDH1 mutations [98].

Result

Initial searches yielded 111 articles, four articles were excluded as a duplication using the computer program Zotero (v5), then one hundred and seven articles were screened for the title and abstract evaluation using the Rayyan website, among them seventy-one articles were excluded. Thirty-six articles were scanned for full-text review and eligibility, furthermore, twenty-five articles were excluded because there were either Reviews and case reports, Not relevant studies, Insufficient data, and Unclear methods and results. Eleven articles were included in the literature review. In addition, the studies were in different regions of the world including Asia, Europe, North America, and Latin America. However, most of the studies were related to the USA.

Among eligible studies, the data of 298 (59.6 ± 28.7) patients with gastric cancer were studied. Regarding the histological classification of gastric cancer, intestinal-type was in 92 of the studied patients while the diffuse type was seen in 44 of the studied pa-

tients. on the other hand, the cagA+ gene was in 151 of the studied patients among them cagA was in 55 patients with intestinal-type while it was in 35 of patients with the diffuse type (Table 1).

In Deguch et al, they aimed to assess the possible association between CagA+ Helicobacter pylori infection and gastric carcinogenesis in gastric cancer patients [100]. they studied about 64 patients with gastric cancer, and their specimens were gastric biopsy. They use more than one procedure, they determine H pylori infection using culturing, a molecular technique such as flaA-PCR, and an immunological technique like serum antibody against CagA was used.

For detecting mutations in P53 and other genes they used PCR-SSCP and direct sequencing. They found that, out of 64 patients, 85.9% (55/64) patients were infected with H pylori, in addition, the intestinal type of gastric cancer was found in 45 patients while the diffuse type was found in 19 patients. regarding the CagA antibody, it was positive in 78.9% (15/19) of patients with the diffuse type while it was in 48.9% (22/45) of patients with the intestinal type (p=0.003). regarding p 53 mutation, it was found in 11(29.7%) out of 37 patients with cagA+ while it was in 2(11.1%) out of cagA- patients.

Furthermore, they reported that the alterations in p53 were found more frequently in patients with cagA+ h pylori (29.7%) in contrast to cagA- H pylori patients (7.4%) with a significant p-value (p=0.033).

Another study by André et al aimed to verify the relationship among H [101]. pylori infection, p53 mutations, and p27Kip1 Protein (p27) expression in gastric adenocarcinomas (GA). they included about seventy-four tumor samples classified as gastric adenocarcinomas, in addition, they used PCR to amplify the urea gene by using specific primers to detect the H pylori infection and the presence of CagA too. Moreover, to assess the alterations in the p53 gene they used the Single-strand conformational polymorphism (SSCP).

Regarding the type of gastric cancer, they found the intestinal type was more frequent 59% than the diffuse type, in addition, they found the H pylori infection in 95% (70/74) of GA patients. moreover, they detect the cagA gene in 63% (44/70) of their studied cases. On the other hand, they noticed that in fifty-three cases 72% showed band mobility shifts in exons 5–8 of the p53 gene.

Regarding the association between p53 mutations and the presence of cagA, they noticed that the majority of cases with p53 mutation (67.9%) were cagA positive, the p-value of the difference between the cases with p53 mutation and presence of cagA was statistically significant (p=0.034).

On the other hand, regarding p 27, they found a reduction in the expression of p27, they noticed no statistical significance between the p27 expression and the presence of cagA although the cases of cagA+ (47.1%) were p27 negative (p=1.000).

Zhang et al conducted a study in 2018, that aimed to investigate the functions of cagA in human gastric cancer and to assess the association between cagA and Phosphatase and tensin homolog (PTEN) ad other genes in gastric cancer [102]. They included 12 patients with gastric cancer, they confirmed gastric cancer pathologically. They used Quantitative polymerase chain reaction (qPCR) to screen gene expression in HGC-27 human gastric cancer cells overexpressing CagA. In addition, they used western blotting for protein expression, and for methylation status, they used methylation-specific PCR. They found that the expression levels of PTEN and other genes were decreased in the patients' group with cagA+ compared to the control group (p value<0.05). In addition, they found that the decreased expression of PTEN was associated with increased methylation levels in the cells.

Another study by Alves et al, wanted to assess the association between p16INK4A inactivation and H. pylori genotype (vacA, cagA, cagE, virB11, and flaA) according to the location and histological subtype of the tumors [103]. They included 77 patients with adenocarcinoma who undergo gastrectomy. They extract the DNA by using the cetyltrimethylammonium bromide (CTAB) technique, in addition, they determine the amount of DNA by using a NanoDropTM 3300 fluorospectrometer (Wilmington, DE, USA). Moreover, they determined the methylation status of the CDKN2A gene by using Sodium bisulfite treatment and methylation-specific PCR (MS-PCR). For the detection of H.pylori and the presence of vacA, cagA, cagE, virB11, and flaA genes they used ureC gene primers for PCR and primer sets respectively. Moreover, they performed Immunohistochemical staining using CINtec p16INK4A Cytology Ki. They found that p16INK4A expression and CDKN2A promoter methylation were found in 77 gastric adenocarcinoma samples by immunohistochemistry and methylation-specific PCR. Furthermore, they found a strong negative correlation between immunostaining and CDKN2A promoter region methylation. Moreover, they reported that the process of methylation of the CDKN2A promoter seems to depend on the H. pylori genotype.

Stec-Michalska et al conducted a study in Poland, they aimed at determining whether FHIT expression is affected by Helicobacter pylori infection, strain virulence (vacA and cagA genes), and histopathological changes in the gastric mucosa of patients with functional dyspepsia having first-degree relatives with gastric cancer [104] .They included about eighty-eight patients with gastric cancer. To identify the H pylori infection they used bacterial DNA amplification, for the level of FHIT gene expression they used qRT-PCR, and they found that in patients having first-degree relatives with gastric cancer FHIT expression was lower (mRNA by ca. 40–45% and protein by 30%) compared with the control patients ($p < .05$). in addition, H. pylori infection decreased the FHIT mRNA level by 10–35% and the protein level by 10–20%.

Bacterial strain vacA(+) cagA(+) lowered FHIT mRNA by ca.

30–35% in the antrum samples of both groups and corpus samples of patients with first-degree relatives with gastric cancer ($p < .05$). they conclude that The decreased FHIT gene expression associated with hereditary factors and with H. pylori infection, especially with vacA(+) cagA(+)-positive strains, may be related to gastric carcinoma development. Kague et al [105] conducted a study to investigate the methylation status of CDH1 in chronic gastritis samples and correlated it with the presence of H. pylori. They used about Sixty gastric mucosal biopsies; they used PCR for the detection of H pylori used PCR for the urease C gene and the presence of page. In addition, CDH1 was analyzed using methylation-specific polymerase chain reaction and direct sequencing of the PCR. They found that H pylori were in 90% of chronic gastritis samples; among these 33% were cagA positive. Regarding the methylation of CDH1, they found it in 63.3% of chronic gastritis samples and 95% of them were also H. pylori-positive. They conclude that CDH1 gene methylation and H. pylori infection are frequent events in samples from Brazilian patients with chronic gastritis and reinforce the correlation between H. pylori infection and CDH1 inactivation in the early steps of gastric tumorigenesis.

Table 1: Shows the total number of patients in studied articles and the type of gastric cancer

Total	number of patients	type of gastric cancer		cagA+ patients	cagA in diffuse-type	cagA in intestinal-type
		diffuse	intestinal			
	298	44	92	151	35	55
mean	59.6	22	46	37.75	17.5	27.5
SD	28.71	4.24	1.41	19.25	3.53	7.77

Table 2: Tumor suppressor genes and alterations associated to CagA

Tumor suppressor genes	Alteration(s)	Method (s)	Authors
phosphatase and tensin homolog (PTEN)	Decreased expression (increased methylation)	Quantitative polymerase chain reaction (qPCR), western blotting	Zhang et al (102)
fragile histidine triad (FHIT)	Decreased expression (increased methylation)	Multiplex PCR, real-time quantitative	Stec-Michalska et al (104)
		RT-PCR, Western Blotting.	
p53	point mutation (alterations in exons 5, 6, 7 and 8, respectively)	Insta Gene Matrix	Deguch et al (100)
		(Bio-Rad, Richmond, CA), PCR-SSCP	
p53	mutations within exons 5– 8 of the p53 gene	Single-strand conformational polymorphism (SSCP), PCR	André et al (101)
	inactivation (increased methylation)	Sodium bisulfite treatment and methylation-specific	Alves et al (103)
CDKN2A		PCR (MS-PCR), PCR	
p27	Decreased expression by increasing	Single-strand conformational polymorphism (SSCP), PCR	André et al(101)
	its degradation		
Cadherin 1 (CDH1)	inactivation (Promoter methylation).	PCR, MSP (methylation-specific PCR)	Kague et al (105)

Conclusion

Cag A can cause alterations on; P53, phosphatase and tensin homolog (PTEN), fragile histidine triad (FHIT), CDKN2A, p27, and Cadherin 1 (CDH1) tumor suppressor genes by either decreased expression by increasing the methylation, inducing point mutation as mentioned, inactivation by increasing the methylation levels, increasing the levels of degradation and methylation the promotor of the tumor suppressor gene as mentioned.

Declarations

Ethical Approval

Not applicable

Competing Interests

The authors declare that they have no competing interests.

Authors' Contributions

AW carried out the systematic analysis of the databases, extracted the data from the articles, did the data analysis, and presented the findings.
ME organized the script and edited the grammar and paraphrase.
NA was the supervisor.

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Availability of Data and Materials

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