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# *INTERLEUKIN 6 POLYMORPHISM AND GASTRIC CARCINOMA*

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# *INTERLEUKIN 6 POLYMORPHISM AND GASTRIC CARCINOMA*

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## Abstract

Background and aims: Genetic susceptibility has been proposed as an important determinant in gastric carcinogenesis. Several human polymorphisms have shown an increased risk for gastric carcinoma. Our aim was to correlate human IL-6 polymorphisms with the histopathologic features of gastritis and the risk of development of gastric carcinoma and to elucidate whether or not the putative association between IL-6 loci polymorphisms and increased risk of gastric carcinoma varies according to the histologic type of the tumors or the gender of the patients. Material and Methods: The polymorphism IL-6-174 G>C was analysed by Polymerase Chain Reaction-Sequence Specific Primers in 200 biopsies: 50 cases of non-atrophic gastritis, 50 of atrophic gastritis, 50 of gastric carcinoma intestinal-type and 50 of gastric carcinoma diffuse-type. Results: IL-6-174 GC and IL-6-174 CC, low producers, were associated with higher risk of development of gastric carcinoma intestinal-type and diffuse-type respectively while the Il-6-174 GG genotype, high producer, was associated with gastritis and gastric carcinoma absence. The same outcome was observed when comparing the differences between genders. Conclusions: Although the Il-6-174 GG genotype, high producer, was associated with gastritis prevalence it seems to confer protection for the gastric carcinoma development.

**Key words:** Interleukin-6 polymorphism, gastric carcinoma, gastritis

**Resumo**

Introdução e objectivos: A susceptibilidade genética tem sido proposta como uma importante determinante no processo cancerígeno gástrico. Vários polimorfismos humanos demonstraram um aumento do risco de desenvolvimento de carcinoma gástrico. O nosso objectivo foi correlacionar os polimorfismos da IL-6 com as características histopatológicas da gastrite e com o risco de desenvolvimento de carcinoma gástrico e esclarecer se a associação existente entre os polimorfismos de IL-6 e aumento do risco de carcinoma gástrico varia de acordo com o tipo histológico dos tumores ou o sexo dos pacientes. Material e Métodos: O polimorfismo IL-6-174 G> C foi analisado por PCR-SSP em 200 biópsias: 50 casos de gastrite não atrófica, 50 de gastrite atrófica, 50 de carcinoma gástrico do tipo intestinal e 50 de carcinoma gástrico do tipo difuso. Resultados: os genótipos IL-6-174 GC e IL-6-174 CC, baixo produtores, foram associados com maior risco de desenvolvimento de carcinoma gástrico do tipo intestinal e tipo difuso, respectivamente, enquanto o genótipo Il-6-174 GG, alto produtor, foi associada com gastrite e ausência de carcinoma gástrico. O mesmo resultado foi observado quando se compararam as frequências genotípicas na distribuição por géneros masculino e feminino. Conclusões: Embora o genótipo Il-6-174 GG, alto produtor, esteja associado com a prevalência da gastrite parecendo conferir protecção para o desenvolvimento de carcinoma gástrico.

**Palavras-chave:** Polimorfismos citocina IL-6; carcinoma gástrico, gastrite

## Introduction

Although the prevalence of gastric carcinoma has gradually decreased it is yet the fourth most common cancer and second cause of cancer mortality worldwide. In Portugal in 2008 was estimated that 3285 people were dead due to gastric cancer, corresponding to 1786 men and 1499 women.[1] The disease has a geographically varied distribution: the incidence decreases dramatically in USA and in many western European countries but much more slowly in far East (China, Japan, Korea), South America (Kolombia. Puerto Rico), Central Europe (Poland) and in developing countries.[2] [3] However, the early stages are often clinically silent and patients have advanced stages at diagnosis, which leads to poor outcomes. [4] To conquer gastric cancer primary prevention would be the best measure, followed by early detection and subsequent surveillance programs.[5]

The Laurén's classification recognizes two major histological types of gastric carcinoma (intestinal and diffuse) which display different clinicopathologic profiles and often occur in distinct epidemiologic settings. Intestinal type is more prevalent in elderly males, whereas diffuse carcinoma tends to occur in younger individuals, mainly females, and often shows hereditary conditioning. [6]

Correa has proposed a model to explain gastric carcinogenesis: chronic gastritis, associated with *Helicobacter pylori* as a pre-neoplasic condition induces progressive histopathological changes in gastric mucosa, towards chronic atrophic gastritis, intestinal metaplasia, dysplasia and finally, intestinal-type adenocarcinoma. This sequence triggered by *Helicobacter pylori* infection is also influenced by a variety of genetic and environmental factors that may act synergistically.[7].

The development of the gastric carcinoma intestinal type after chronic inflammation as response to *Helicobacter pylori* infection is sustained by epithelial cell cycle, increased rates of apoptosis and cell proliferation leading to multifocal mucosal atrophy where intestinal metaplasia and dysplasia arise in either native gastric or "intestinalized" gastric epithelium. Long-standing *Helicobacter pylori*-induced gastric inflammation leads then to atrophic gastritis that is considered the first important condition in gastric cancer histogenesis where dysplasia develops (or not) in the intestinal metaplastic background before the recognized morfhology of gastric carcinoma. [7]

The Laurén's diffuse-type of gastric carcinoma develops in atrophic changes not as severe as in Laurén's intestinal type and was previously considered to have little relation with *Helicobacter pylori* infection. [8]

However, epidemiological and histopathological studies have shown that the development of Laurén's diffuse-type is closely related to *Helicobacter pylori* infection and some reports also identified moderate atrophic changes and pangastritis. [9]

The severity of the mucosal inflammation and host characteristics may directly induce mutagenetic events ultimately leading to Laurén's diffuse type. [10]

Gastric carcinoma arising *de novo* is less frequently associated with intestinal metaplasia or adenoma, even when related with *Helicobacter pylori* infection. [8]

The World Health Organization (WHO) has classified *Helicobacter Pylori* a class I carcinogen since 1994. [11] However, there are high interdividual variations observed in the severity of gastric inflammation and the clinical outcome of the infection. [12] It is believed that a combination of virulent organism, permissive environment and genetically susceptible host

is necessary as only a small proportion of individuals exposed to known environmental risk factors develop gastric carcinoma, in poor socioeconomic hygienic conditions, high salt intake and Helicobacter pylori infection. [13]; [4]

Host risk factors, after the actual state of the art depending on cytokine gene polymorphisms as major determinants in mucosal expression, gastric inflammation and long term development of precancerous lesions. [12]

The host ability to regulate cytokine production has been shown to be influenced by the presence of polymorphisms in the genes coding and promoter regions to regulate the immune and inflammatory response to the resistant infection. [14]

In general, gastric mucosal inflammation in *Helicobacter pylori* infection is exacerbated in pacients with hight producer alleles of pro-inflammatory cytokines and low producer alleles of anti-inflammatory cytokines, inducing higher risk for the development of gastric carcinoma. [15]

Il-6 is a multifunctional cytokine produced by immune and many non-immune cells, including monocytes, lymphocytes, macrophages, endothelial cells, intestinal epithelial cells and osteoclasts. [14] It is known that IL-6 has several immunological activities including induction of several acute phase proteins and regulation of proliferation and differentiation of immunocompetent cells. It also has implications in the pathogenesis and/or prognosis of several tumors, including multiple myeloma, renal cell carcinoma and prostatic carcinoma. [16]

Experimental studies have recently demonstrated that IL-6 plays an important role as a prognostic factor (patients with a high serum level of IL-6 exhibited s significantly poor outcome compared with patients with a low level of IL-6) and it should be used as a tumor marker of advanced gastric cancer and lymph node metastasis.[17]; [16]

The IL-6 gene is located on chromosome 7p21 and the single nucleotide polymorphisms at the 5' flanking region have been identified has IL-6-174,-572 and -597.[18] IL-6-174 G allele carriers produce higher levels of IL-6 than those with the C/C genotype. [19] On combined analyses of several studies IL-6-174 polymorphisms have shown controversial relationship with gastric carcinoma. Some authors have reported that the IL-6-174 G allele carriers account for a significantly higher incidence of gastric carcinoma when compared with G/G genotype IL-6, where lower producer genotype IL-6-174 G/C is also, associated with increased risk. Some studies have shown no significant relationship of IL-6-174 polymorphism with gastric diseases. [15]

A major challenge is to explain why and how a particular infection induce a neoplastic cascade.

Our aim was to correlate human IL-6 susceptibility polymorphisms with the histopathologic features of gastritis and the risk of development of gastric carcinoma and to elucidate whether or not the putative association between IL-6 loci polymorphisms and increased risk of gastric carcinoma varies according to the histologic type of the tumors or the gender of the patients.

## Material and Methods

*Material*

This study included 200 samples obtained from endoscopic biopsies analysed retrospectively.

The samples correspond to 29 male and 21 female with diffuse type gastric carcinoma (mean age 62.0 ±14.3 years); 33 male and 17 female had intestinal type gastric carcinoma (mean age 73.5 ±11.0 years), 27 male and 23 female had atrophic gastritis (mean age 54.4 ±12.8 years) and 15 male and 35 female had non-atrophic gastritis (mean age 46.6 ±17.2 years).

The specimens were formalin fixed, paraffin embedded, and stained with hematoxylin-eosin. Histological classifications were applied according with the criteria of Laurén's classification for gastric carcinomas and update Sydney System for gastritis inflammatory level grading. The cases correspond to 50 diffuse type gastric carcinomas, 50 intestinal type gastric carcinoma, 50 cases of atrophic gastritis and 50 cases of non-atrophic gastritis. Histological slides were examined by experienced pathologists (L. Carvalho and V. Sousa).

Figure I and II show representative images of the biopsy samples.

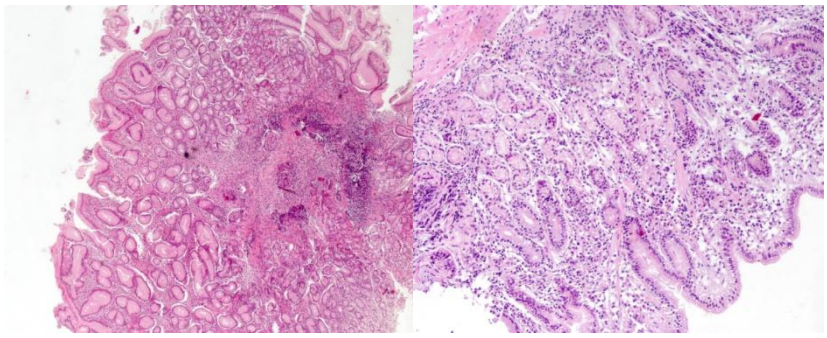

**Figure 1: A- Non atrophic gastritis (H&E, x 40). B- atrophic gastritis (H&E, x 100)**

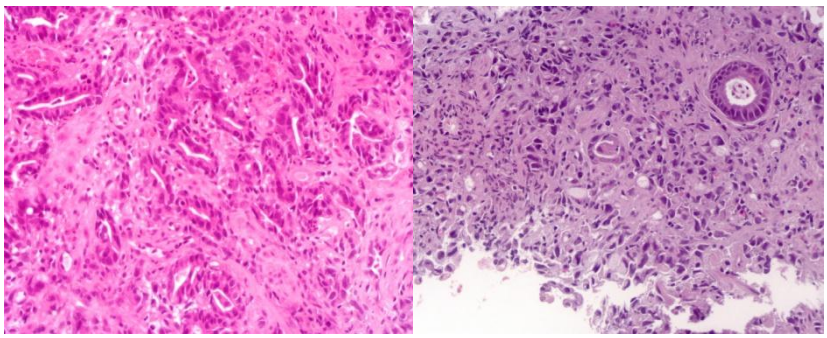

**Figure 2: A- Gastric cancer intestinal-type (H&E, x 200). B- gastric cancer diffuse-type (H&E, x 200)**

*DNA extraction*

Genomic DNA was extracted from the gastric biopsies by using a commercial DNA extraction kit *QIA-amp DNA mini Kit* (Qiagen, Hilden, Germany). Samples were processed for isolation of DNA as described by the manufacturer by FFPE samples.

*Genotyping of cytokine gene polymorphisms*

IL-6-174 cytokine gene polymorphism was genotyped by Polymerase Chain Reaction-Sequence Specific Primers (PCR-SSP) in the promoter region (-174). Briefly, 100 ng of extracted DNA were amplified in a 50 µl final reaction volume under the following conditions: 1×DNA polymerase gold buffer (Applied Biosystems), 1.5 mM $MgCl_2$, 200 µM each dNTPs, 500 nM each primer, and 2.5 U AmpliTaq gold (Applied Biosystems). PCR conditions were: 94°C for seven minutes, then 40 cycles at 94°C for 30 seconds, 60°C for 30 seconds, 72°C for 60 seconds, and finally 72°C for 20 minutes in a termocycler MyCycler Personal (BioRad, Califórnia, EUA). Separation of PCR product on a 2% gel agarose Seakem ME (Biowhittaker Molecular Aplications, Rockland, EUA), and stained with ethidium bromide.

*Statistical analysis*

The results are presented as genotypes and allele frequencies for the IL-6-174 polymorphism. $X^2$test and Fisher's exact test were performed using the STATISTICA 9.1 software (StatSoft, Inc., 2009), p-values < 0.05 were considered significant.

**Results**

The alleles and genotypes of the polymorphism at the promoter region of the IL-6 gene (position -174) are shown at Table 1.

We determined that the G allele is more frequent in non-atrophic gastritis, atrophic gastritis and intestinal-type gastric carcinoma samples (54%, 56% and 58%, respectively) compared to 32% found in diffuse-type gastric carcinoma samples.

Genotype analyses showed that the homozigotic GG genotype is more frequent in non-atrophic gastritis and atrophic gastritis samples (46%, 50%, respectively) compared to the frequency found in the two types of gastric carcinoma (16% in intestinal-type and 8 % in diffuse-type) samples.

The homozigotic CC genotype was more frequent in diffuse-type gastric carcinoma samples (70%) compared to 34 % in intestinal-type gastric carcinoma samples, which had a similar frequency of the two types of gastritis (36% in non-atrophic gastritis and 38% in atrophic gastritis) samples.

The heterozigotic CG genotype had highest levels in the intestinal-type gastric carcinoma samples (50%) compared to lower frequencies in diffuse-type gastric carcinoma samples (22%) and with gastritis samples (18% in non-atrophic gastritis and 12% in atrophic gastritis).

**Table 1: Allele and genotype distribution of IL-6- 174 polymorphism according to disease**

| | NAG | | AG | | GC intestinal | | GC diffuse | |
|---|---|---|---|---|---|---|---|---|
| **IL- 174** | **n** | **Freq.** | **n.** | **Freq.** | **n.** | **Freq.** | **n.** | **Freq.** |
| **Allele** | | | | | | | | |
| **G** | 55 | 55% | 56 | 56% | 58 | 58% | 32 | 32% |
| **C** | 45 | 45% | 44 | 44% | 42 | 42% | 68 | 68% |
| **Genotype** | | | | | | | | |
| **GG** | 23 | 46% | 25 | 50% | 8 | 16% | 4 | 8% |
| **GC** | 9 | 18% | 6 | 12% | 25 | 50% | 11 | 22% |
| **CC** | 18 | 36% | 19 | 38% | 17 | 34% | 35 | 70% |

NAG: non-atrophic gastritis; AG: atrophic gastritis; GC: gastric cancer

Diffuse-type gastric carcinoma demonstrated significant differences compared to intestinal-type gastric carcinoma in the allele C (p= 0, 0002), genotype CG (p=0.0035) and genotype CC (p=0.0003).

Comparing diffuse-type gastric carcinoma to non-atrophic gastritis we found significant differences in the allele C (p=0.0006), genotype CC (p=0.0013) and highly significant differences in genotype GG (p<0.0001).

No significant differences were found when comparing gastric carcinoma with atrophic gastritis and intestinal type gastric carcinoma with non-atrophic gastritis.

**Table II: Comparison of the prevalence of gastric cancer (diffuse and intestinal-type) and gastritis (atrophic and non-atrophic)**

| | GC vs AG (p value)[a] | GC intestinal vs NAG (p value)[a] | GC intestinal vs GC diffuse (p value)[a] | GC diffuse vs NAG (p value)[a] |
|---|---|---|---|---|
| **Allele** | | | | |
| **G** | NS | NS | NS | NS |
| **C** | NS | NS | 0.0002 | 0.0006 |
| **Genotype** | | | | |
| **GG** | NS | NS | NS | < 0.0001 |
| **CG** | NS | NS | 0.0035 | NS |
| **CC** | NS | NS | 0.0003 | 0.0013 |

NAG: non-atrophic gastritis; AG: atrophic gastritis; GC: gastric cancer
[a] p values were calculated with the qui-square test or Fisher's exact test
NS: not significant (p > 0.05)

When cases were divided according to the gender of the patients, in males, genotype analyses showed that the homozigotic G/G genotype is more frequent in non-atrophic gastritis and

atrophic gastritis samples (60%, 74%, respectively) compared to the frequency found in the two types of gastric carcinoma (18% in intestinal-type and 3 % in diffuse-type) samples.

The homozigotic CC genotype was the most frequent in diffuse type gastric carcinoma samples (76%) and in intestinal type gastric carcinoma samples the genotype with higher frequencies was the GC (46%).

The genotype frequencies according to disease among males are summarized in Table III.

**Table III: Genotype distribution of IL-6- 174 polymorphism according to disease in males**

| | NAG | | AG | | GC intestinal-type | | GC diffuse-type | |
|---|---|---|---|---|---|---|---|---|
| **IL- 174** | **n. (15)** | **Freq.** | **n.(27)** | **Freq.** | **n.(33)** | **Freq.** | **n.(29)** | **Freq.** |
| **Genotype** | | | | | | | | |
| **GG** | 9 | 60% | 20 | 74% | 6 | 18% | 1 | 3% |
| **GC** | 3 | 20% | 2 | 7% | 15 | 46% | 6 | 21% |
| **CC** | 3 | 20% | 5 | 19% | 12 | 36% | 22 | 76% |

NAG: non-atrophic gastritis; AG: atrophic gastritis; GC: gastric cancer

Results from IL-6 genotype on females showed a similar distribution to the one in males. Again the CC genotype was more frequent in the gastric carcinoma diffuse-type (62%), the GC genotype was the most prevalent in gastric carcinoma intestinal-type (59%). The only difference was related to the genotype most frequent in both forms of gastritis, which in females was the CC genotype (43% in non-atrophic gastritis and 61% in atrophic gastritis).

The genotype frequencies according to disease among females are summarized in Table IV.

**Table IV: Genotype distribution of IL-6- 174 polymorphism according to disease in females**

| | NAG | | AG | | GC intestinal | | GC diffuse | |
|---|---|---|---|---|---|---|---|---|
| **IL- 174** | **n. (35)** | **Freq.** | **n.(23)** | **Freq.** | **n.(17)** | **Freq.** | **n.(21)** | **Freq.** |
| **Genotype** | | | | | | | | |
| **GG** | 14 | 40% | 5 | 22% | 2 | 12% | 3 | 14% |
| **GC** | 6 | 17% | 4 | 17% | 10 | 59% | 5 | 24% |
| **CC** | 15 | 43% | 14 | 61% | 5 | 29% | 13 | 62% |

NAG: non-atrophic gastritis; AG: atrophic gastritis; GC: gastric cancer

**Table V: Comparison of the prevalence of gastric cancer (diffuse and intestinal-type) and gastritis (atrophic and non-atrophic) in both genders**

| | NAG vs AG | | GC intestinal vs diffuse | | CG vs Gastritis | |
|---|---|---|---|---|---|---|
| | **Male (p value)[a]** | **Female (p value)[a]** | **Male (p value)[a]** | **Female (p value)[a]** | **Male (p value)[a]** | **Female (p value)[a]** |
| **Genotype** | | | | | | |
| **GG** | NS | NS | NS | NS | < 0.0001 | 0.02 |
| **GC** | NS | NS | 0.05 | 0.03 | 0.02 | 0.01 |
| **CC** | NS | NS | 0.002 | 0.04 | 0.0005 | NS |

NAG: non-atrophic gastritis; AG: atrophic gastritis; GC: gastric cancer
[a] p values were calculated with the qui-square test or Fisher's exact test
NS: not significant ($p > 0.05$)

When cases were divided according to the gender of the patients, a significant difference in genotype frequency was observed between gastric carcinoma biopsies and gastritis (Table V). The presence of GG genotype was significantly higher in gastritis than in gastric carcinoma ($p< 0.0001$ in males and $p = 0.02$ in females) biopsies.

Diffuse-type gastric carcinoma demonstrated significant differences compared to intestinal-type gastric carcinoma in the genotype CG (p=0.05 in males and p=0.03 in females) and genotype CC (p=0.002 in males and p=0.04 in females).

There was no significant difference in genotype frequency between both forms of gastritis (non-atrophic and atrophic).

**Discussion**

The development of gastric carcinoma is an unpredictable process and disease progression varies considerably in between patients, recognizing the presence and extent of gastritis with associated atrophy as a marker to identify patients with different carcinogenic risk.

Gastritis is characterized by increased infiltration of the lamina propria by mononuclear leukocytes (chronic inflammation) and polymorphonuclear neutrophils (acute inflammation) The most frequent cause of gastritis is *Helicobacter pylori* infection and polymorphonuclear neutrophille infiltration is usually associated with it in humans. Both mononuclear leukocytes and neutrophils infiltrate the epithelium, and marked acute inflammation is frequently accompanied with microabscesses (collections of neutrophils in the glandular or foveolar lumen). Another characteristic of chronic gastritis *Helicobacter pylori* associated is the presence of lymphoid aggregates with germinal centers. [20]

The inflammatory changes may persist throughout the whole precancerous process, but their intensity tends to decrease as the process advances. Since the initial *Helicobacter pylori* infection targets a normal mucosa with well-preserved gastric glands, by definition such gastritis is non-atrophic. [21]

The outcome of such lesion follows the epidemiological model of causation, modulated by

the interplay of three sets of etiological factors: those linked to the infectious agent, the host's genetic susceptibility and those related to the external environment. The non-atrophic gastritis may be cured by clearing H*elicobacter pylori* infection. Otherwise, it may evolve in two ways: either it remains as non-atrophic or it progresses in severity, leading to damage to the gastric glands, which may eventually disappear. This dichotomy determines whether the gastritis enters in the precancerous process or not. Infection with *Helicobacter pylori* strains possessing recognized virulence factors is significantly associated with a higher risk for progression to gastric pre-neoplastic lesions. Individuals with a duodenal ulcer, who classically have antral predominant non-atrophic gastritis lasting for decades, do not have elevated gastric carcinoma risk, despite being typically infected with virulent strains. [21]

Mucosal gastric atrophy is the first specific recognizable step in the precancerous cascade. It is diagnosed on the basis of the presence chronic inflammatory cells, including lymphocytes and plasma cells that expand the lamina propria, and the disappearance of the normal glands which leads to a reduction of gastric secretory functions. Usually is multifocal and the foci of atrophy are present in the mucosa of gastric antrum and body, and their extension progresses with time. [22]

The current definition of gastric mucosal atrophy includes two different phenotypes: loss (shrinkage or disappearance) of glands, which are replaced by fibrotic expansion of the lamina propria; and metaplastic replacement of native glands by intestinalized and/or pseudopyloric glands. [23]

The risk for subsequent carcinoma is low among patients with chronic gastritis alone, this is a relatively common condition. However, the risk arises with the acquisition of atrophy,

metaplasia and dysplasia, and, therein, carcinoma has been observed to develop in 9% of patients with atrophic gastritis during long-term follow-up. Those carcinomas are usually of the intestinal type. The precursor lesion for sporadic diffuse GC is yet not known. The hereditary diffuse GC is due to the E-cadherine (CDH1) germline mutation. [24]

The histological classification of gastric carcinoma into the intestinal type and diffuse type is based on the criteria proposed by Laurén. The intestinal type is characterized by cohesive neoplastic cells forming gland like tubular structures, whereas in diffuse type cell cohesion is absent, so that individual cells infiltrate and thicken the stomach wall without forming a discrete mass. This difference in microscopic growth pattern is also reflected in the different macroscopic appearance of the two histological subtypes. Whereas for intestinal type the macroscopic margins correspond approximately to the microscopic spread, the diffuse type as a poorly differentiated cancer can extend submucosally far beyond its macroscopic borders. [6]

In 2011 a meta-analyses assessed the association of interleukin gene polymorphisms (IL-1β, IL-1RN, IL-8, IL-10 and TNF-α) with gastric carcinoma risk and revealed an increased risk for IL-1RN*2 carriers. This association was specific for non-Asian populations and was independent from Laurén's type and location of the carcinoma. [25]

The individual's genotype at polymorphic sites in IL-6 (specially -174 site) is thought to determine the IL-6 response to stimuli and predisposes to development of diseases where IL-6 has been implicated. It has been demonstrated that the C allele at position -174 compared with the G allele was associated with lower levels of plasma IL- 6 in normal subjects.[19]

Other studies showed that gastric carcinoma tissues contain high levels of the pro-inflammatory cytokine IL-6 and that the levels of these cytokine are associated with the

growth pattern of the tumor. [16] it was also suggested that IL-6 may be involved in angiogenesis in gastric carcinoma or accelerating the activity and spread of cancer cells and that there was a tendency toward shorter survival in patients expressing a high level of IL-6. [17]

Our results show that the carriers of the low producer polymorphisms (such as IL-6-174 C allele and IL-6-174 CC genotype) have high prevalence of diffuse type gastric carcinoma.

However, the IL-6-174 G allele and the IL-6-174 GG genotype, associated with high production of IL-6, have demonstrated correlation with gastritis prevalence. These results provide indirect evidence of the increased pro-inflammatory effect associated with the IL-6-174 G allele. In this case (GG genotype) when comparing diffuse type gastric carcinoma and non-atrophic gastritis group we found high significant statistical difference ($p<0.0001$).

Altogether, these findings indicated that the odds of developing diffuse-type gastric carcinoma were greatest in those individuals with low producer polymorphisms and that the high production of IL-6 apparently provides some protection against the development of diffuse-type gastric carcinoma.

Although our findings shown that the G allele was the most prevalent in the group with intestinal type gastric carcinoma (58%) when comparing the frequency of genotypes we find that GC genotype was the most prevalent (50%).

So, as we were able to see in the previous case, the low producer genotype CG has high prevalence of intestinal type gastric carcinoma.

The genotype GG (high producer) is associated with low prevalence of the intestinal type gastric carcinoma.

Altogether, these findings indicated that the odds of developing intestinal type gastric carcinoma were greatest in those individuals with low producer polymorphisms and that the high production of IL-6 apparently provides some protection against the development of intestinal type gastric carcinoma.

However, although both gastric carcinoma histological types have increased risk of developing disease with low producer genotypes, the diffuse type gastric carcinoma prevalence is highest with IL-6-174 CC genotype and the intestinal type gastric carcinoma is more frequent in the carriers of the IL-6-174 CG genotype.

In the end our results support the previously reported by Kamangar et al, who have demonstrated that, compared with IL-6-174 GG genotype, the low producer genotype IL-6-174 GC has an increased risk of gastric carcinoma. [26]

In contrast, several other studies have shown no significant relationship of IL-6- 174 polymorphisms with gastric disease, while Gatti et al have shown that the IL-6-174 G allele carriers account for a significantly higher incidence of gastric carcinoma, correlation that has not been observed in this study. [15]

As suggested by Hu et al, although previous molecular studies of potentially functional polymorphisms in candidate genes and gastric cancer susceptibility lack consistency, this heterogeneity may arise from the disease itself due to different histological types and anatomic localizations that may involve different etiologies and genetic predispositions or due to diverse environmental exposure in the early stages such as H. pylori infection or dietary factors. Controversy, they have advanced our knowledge of the role of genetic susceptibility in the etiology of gastric cancer. [27]

The discovery of markers for increased risk, whether causative or not, or markers of protection may facilitate screening for high-risk individuals.

A recent study suggested a new method of screening the degree of atrophy. A good indicator is the blood level of pepsinogen I (PGI). It is secreted most prominently by the oxyntic mucosa and therefore blood levels become progressively lower as the loss of oxyntic glands advances. Pepsinogen II (PGII) is secreted by foveolar glands in the mucosa of gastric antrum and body. Its secretion is stimulated by inflammation (such as *H. pylori* infection) and cellular proliferation, both hyperplastic and neoplastic. PGI/PGII is a good indicator of atrophy and to some extent of precancerous lesions. [21]

When cases were divided according to the gender of the patients the relationships between genotype frequencies and diseases where similar to the ones found in the whole group: both gastric carcinoma histological types have increased risk of developing disease with low producer genotypes (GC genotype in intestinal-type and CC genotype in diffuse-type) and IL-6-174 GG genotype, high producer, apparently provides some protection against the development of gastric carcinoma.

In conclusion, although the Il-6-174 GG genotype, high producer, was associated with gastritis prevalence it seems to confer protection for the gastric carcinoma development.

It is too early to generalize from this work. Nevertheless, we believe that the outcomes of this type of study (i.e., identification of risk factors and a better definition of risk) are important to provide means to identify individuals who are at greatest risk of developing gastric carcinoma. As a consequence, it may become possible to target such individuals with selective interventions designed to prevent and/or reduce the incidence of gastric carcinoma in the general population.

Even more, clarifying the etiology of gastric cancer could throw light into the pathogenesis of other cancers, especially those where chronic active inflammation is suspected to play a role, such as cervical carcinoma, hepatic and colo-rectal and perhaps, even prostatic carcinoma. [28]

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