RISK FACTORS FOR THE DEVELOPMENT OF GASTRIC CANCER: A SINGLE CENTER EXPERIENCE

Andrija Rančić1, Vesna Brzački1,2, Aleksandar Popović3, Marija Topalović4

Gastric cancer is the fourth most common cancer in the world with over a million new cases per year, and over 750,000 deaths caused by it. The highest incidence is recorded in Japan and China, Eastern Europe and South America, and the lowest is in Africa. The disease is 2 to 3 times more common within male population and people over 60 years of age, but this boundary shifts towards younger age. Five-year survival rate in European countries is 10 to 30%. This is a multifactorial disease and the most important risk factors are: Helicobacter pylori infection, smoking cigarettes, alcohol consumption, male gender, older age, reduced intake of vegetables and fruits, genetic predisposition, diabetes mellitus, etc.

A prospective study was conducted on a selected population of 43 patients diagnosed with the two most common types of stomach cancer: adenocarcinoma and signet ring cell type cancer. Out of the total number of examinees, there were 32 male and 11 female patients, the average age of 68.83±9.26. Among all patients, 39.5% were smokers, 20.9% were alcohol consumers, 62.8% had Helicobacter pylori infection and 16.3% patients had diabetes mellitus. Analysis of the relationship between sex, age, smoking, alcohol intake and diabetes mellitus with the type of cancer did not show statistical significance. The frequency of Helicobacter pylori was statistically significantly different in relation to the type of cancer (p=0.007).

In our study, there was no statistically significant difference between the male gender, the younger population, smokers, chronic alcohol consumers, and patients with diabetes mellitus and a certain type of cancer. Helicobacter pylori infection was found to be a key factor in the development of both types of gastric cancer. This research included a smaller sample of patients and it is necessary to examine a larger number of subjects in the future.

Key words: gastric cancer, Helicobacter pylori, smoking, alcohol, diabetes mellitus

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Introduction

Gastric cancer is a multifactorial disease that is mostly caused by environmental factors and more often by genetic polymorphisms (1). Due to insufficiently known pathogenesis, high aggressiveness and heterogeneous nature, today, this disease represents a major health problem (2).

Epidemiology

Gastric cancer is the fourth most common cancer in the world with an incidence of about 989,500 new cases annually and is the second most common cause of cancer death with about 738,000 cases globally (3). More than 50% of new cases occur in developing countries, such as Europe, East Asia (Japan and China), and parts of Central and South America, while the lowest incidence is in Australia, southern parts of Asia, and North America (4). Gastric cancer occurs 2 to 3 times more often in male population (5). In Japan, five-year survival rate is up to 90% (6), while five-year survival in European countries is only 10 to 30% (7).

Classification of gastric cancer

The first classification of stomach cancer according to Lauren was adopted in 1965, and according to it, all stomach cancers are divided into intestinal and diffuse type (8). The intestinal type includes tubular and glandular elements, with
different degrees of differentiation, and the diffuse type has weakly cohesive individual cells without the formation of glands, which also includes gastric carcinoma with ring cells (8 - 10). In 2010, the World Health Organization suggested a new division of gastric cancer into adenocarcinoma (which has subtypes – tubular, papillary, mucinous, mixed) and poorly cohesive carcinoma (which includes signet ring cell carcinoma and other poorly cohesive types). According to this classification, the most common is tubular gastric adenocarcinoma (11, 12).

Risk factors

One of the important risk factors is the presence of malignant diseases in the family. It is estimated that only 3% of cancers are truly inheritable (13). This form of cancer is known as hereditary diffuse gastric cancer (HDGC) and a mutation in the cadherin-1 (CDH1) gene is thought to be responsible for its development (14). People carrying these mutations have three times higher risk of developing stomach cancer. HDGC occurs more frequently in Asia than in other parts of the world. (15, 16).

The influence of smoking and alcohol consumption in the development of gastric cancer is unavoidable. About 60% of male smokers and 20% of female smokers have a higher risk of developing gastric cancer than non-smokers (17, 18). Smoking status and daily number of smoked cigarettes (over 20 cigarettes/day) are also important, and it shows that former smokers are also at risk of developing gastric cancer (18). Increased alcohol intake is especially associated with the development of non-cardiac types of cancer. Studies indicate a typical ALDH2 genotype in those patients. A higher risk of disease was recorded in the group of former and active drinkers, compared to the group of patients who do not consume alcohol (19).

Gastric infections carry an important role in the development of cancer. Helicobacter pylori (H. pylori) was described by the World Health Organization in 1994 as a first-class carcinogen for the development of gastric cancer (19). Most infected patients are asymptomatic, but gastric cancer appears to develop in 1 to 3% of these cases (20). In addition to H. pylori, the Epstein Barr virus (EBV) is increasingly mentioned as an infectious factor present in 10% of gastric cancers. EBV genome and proteins have been identified in gastric tumor cells. This role of EBV is still not very clear (21).

A higher intake of fresh fruit, green and yellow vegetables rich in vitamin C, B, E and folate is recommended for primary prevention. Beta-carotene is considered the main substance in reducing the risk of cancer (22). All these substances have a strong antioxidant effect (22, 23).

Numerous metabolic diseases can be a risk factor for the development of gastric cancer, and one of the diseases that stands out is diabetes mellitus. The reason is its association with a chronic, systemic inflammatory response, which is why it is associated with the development of pancreatic, liver, colon and uterine tumors (24, 25). The association between diabetes and the occurrence of gastric cancer has not been reliably established, although some studies have proven their connection (26).

The objective

The main goal of this research is to determine the relationship between gender, age, smoking cigarettes, alcohol consumption and diabetes mellitus with the appearance of gastric cancer in the observed patient population.

Materials and methods

The prospective study research was conducted at the Clinic for Gastroenterology and Hepatology and at the Institute of Pathology of the University Clinical Center in Niš. The research included 43 patients diagnosed with gastric cancer between January and October 2022. All patients underwent a clinical and physical examination, collection of detailed anamnestic data, analysis of blood count and biochemical parameters, and oesophago-gastro-duodenoscopy examination. Information about concomitant diseases and harmful habits were taken from each patient. During the endoscopic examination, biopsy samples were taken of changes suspected of being tumorous. PH verification was performed at the Institute of Pathology by immunohistochemical staining and determination of specific markers in tumor cells (CK7, CK20, MUC5AC, MUC2, CKAE1/AE3, Ki67 proliferation index, presence of acidic mucins in tumor cells, villin, synaptophysin and chromogranin). Two basic types of cancer were taken into account: adenocarcinoma and signet ring cell type cancer, as well as a group of remaining tumor types (MALT lymphomas, Hodgkin lymphomas and metastatic stomach tumors). After the insight into the final pH diagnoses and the obtained anamnestic data, the statistical processing of the data was performed.

Statistical data processing

Data are presented in the form of arithmetic mean, standard deviation, minimum and maximum values, and in the form of absolute and relative numbers. A comparison of age in relation to gender and type of cancer was performed by t-test and ANOVA. A comparison of the frequency of risk factors was performed using the Chi-square test and Fisher’s test. The hypothesis was tested with a significance threshold of p<0.05. Statistical data processing was performed in the software package R and RStudio.

Results

This study included 43 patients (32 male and 11 female). The average age of the examinees was 68.83 ± 9.26 (Min 45, Max 87 years); there were
39.5% of smokers, 20.9% of patients who consume alcohol, 62.8% of patients with Helicobacter pylori and 16.3% of patients suffering from diabetes mellitus. From all the patients that were examined, 51.2% had adenocarcinoma, 34.9% had signet ring cell type and 14.0% had other types of tumors (MALT lymphomas, Hodgkin lymphomas and metastatic stomach tumors) (Table 1).

Male patients were older, but there was no statistically significant difference compared to female patients ($p = 0.103$). The frequency of smoking and alcohol consumption was higher among male participants, but without a statistically significant difference between genders ($p = 0.154$, respectively $p = 0.407$). The presence of Helicobacter pylori and the incidence of diabetes did not show statistically significant difference in relation to gender ($p = 1.000$, or $p = 0.106$).

There was no statistically significant difference between genders in relation to the type of cancer ($p = 0.664$) (Table 2).

In relation to the average age of the patients, there was no statistically significant difference in the incidence of a certain type of cancer ($p = 0.602$). In relation to the type of cancer, there was no statistically significant difference in the frequency of smoking and alcohol consumption ($p = 0.766$, respectively $p = 0.738$). The frequency of H. pylori was statistically significantly different in relation to the type of cancer ($p = 0.007$). H. pylori was present in 59.1% of patients with adenocarcinoma, 86.7% of patients with a signet ring cell type and 16.7% of patients with other types of cancer (Table 3, Chart 1). The occurrence of any of the two types of gastric cancer in patients with diabetes did not show a statistically significant difference (Table 3).

### Table 1. Demographic and clinical characteristics of the studied population

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Count</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>68.83±9.26</td>
<td>45-87 years</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>32</td>
<td>74.4</td>
</tr>
<tr>
<td>Female</td>
<td>11</td>
<td>25.6</td>
</tr>
<tr>
<td>Smoking</td>
<td>17</td>
<td>39.5</td>
</tr>
<tr>
<td>Alcohol</td>
<td>9</td>
<td>20.9</td>
</tr>
<tr>
<td>H. pylori</td>
<td>27</td>
<td>62.8</td>
</tr>
<tr>
<td>D. mellitus</td>
<td>7</td>
<td>16.3</td>
</tr>
<tr>
<td>Dg.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adenocarcinoma</td>
<td>22</td>
<td>51.2</td>
</tr>
<tr>
<td>Signet ring cell</td>
<td>15</td>
<td>34.9</td>
</tr>
<tr>
<td>Other tumor types</td>
<td>6</td>
<td>14.0</td>
</tr>
</tbody>
</table>

### Table 2. Demographic and clinical characteristics in relation to gender

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Male</th>
<th>Female</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>70.23±8.88</td>
<td>64.91±9.62</td>
<td>0.103&lt;sup&gt;1&lt;/sup&gt;</td>
</tr>
<tr>
<td>Smoking</td>
<td>15 46.9%</td>
<td>2 18.2%</td>
<td>0.154&lt;sup&gt;2&lt;/sup&gt;</td>
</tr>
<tr>
<td>Alcohol</td>
<td>8 25.0%</td>
<td>1 9.1%</td>
<td>0.407&lt;sup&gt;2&lt;/sup&gt;</td>
</tr>
<tr>
<td>H. pylori</td>
<td>20 62.5%</td>
<td>7 63.6%</td>
<td>1.000&lt;sup&gt;2&lt;/sup&gt;</td>
</tr>
<tr>
<td>D. mellitus</td>
<td>3 9.4%</td>
<td>4 36.4%</td>
<td>0.106</td>
</tr>
<tr>
<td>Dg.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adenocarcinoma</td>
<td>17 53.1%</td>
<td>5 45.5%</td>
<td>0.664&lt;sup&gt;3&lt;/sup&gt;</td>
</tr>
<tr>
<td>Signet ring cell</td>
<td>10 31.3%</td>
<td>5 45.5%</td>
<td></td>
</tr>
<tr>
<td>Other tumor types</td>
<td>5 15.6%</td>
<td>1 9.1%</td>
<td></td>
</tr>
</tbody>
</table>

<sup>1</sup> t-test,  <sup>2</sup> Fisher’s test,  <sup>3</sup> Chi-square test
Table 3. Demographic and clinical characteristics in relation to cancer type

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Adenocarcinoma</th>
<th>Signet ring cell</th>
<th>Other types</th>
<th>p^1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>70.14±9.07</td>
<td>66.93±10.19</td>
<td>69.00±8.09</td>
<td>0.602</td>
</tr>
<tr>
<td>Smoking</td>
<td>9</td>
<td>5</td>
<td>3</td>
<td>0.766</td>
</tr>
<tr>
<td>Alcohol</td>
<td>4</td>
<td>3</td>
<td>2</td>
<td>0.738</td>
</tr>
<tr>
<td>H. pylori</td>
<td>13</td>
<td>13</td>
<td>1</td>
<td>0.007</td>
</tr>
<tr>
<td>D. mellitus</td>
<td>3</td>
<td>3</td>
<td>1</td>
<td>0.877</td>
</tr>
</tbody>
</table>

^1 ANOVA, ^2 Fisher’s test, ^3 Chi-square test

![Chart 1. Cancer type in relation to H.pylori]

Discussion

Gastric cancer is a major problem today despite attempts of changing lifestyles, eating habits and treatment of Helicobacter pylori infection. There are still many unknown genetic and epigenetic factors that precede the development of this disease (27). The Japanese Association for Gastric Cancer as well as Nakamura and colleagues suggest a division into 5 histological subtypes: tubular adenocarcinoma, papillary adenocarcinoma, poorly cohesive (signet ring cell and other subtypes), mucinous and mixed adenocarcinoma (28). According to ESMO guidelines from July 2022, more than 1 million new cases of this disease were registered during 2020, out of which 768,800 resulted in deaths (29). Despite the large number of patients on a global level, some studies have determined a decrease in the incidence of the disease in the last few decades (27). A large study by Morgan Eileen and colleagues, based on data from the GLOBOCAN database from 185 countries of the world from 2020, indicates that the incidence of the disease is twice higher in male than in female population (15.8 male and 7.0 female per 100,000 cases), with variations between individual countries. The highest incidence is recorded in East Asia (32.5 male and 13.2 female per 100,000), in the male population in Japan (48.1 per 100,000), Mongolia (47.2) and Korea (39.7). The lowest incidence is in Africa with less than 5 developed cancers in 100,000 cases (30). In our observed population of 43 patients, there was no statistically significant difference in morbidity in relation to gender, although world studies record a higher incidence of morbidity in males. Arnold M and colleagues in their study deny the decline in incidence and indicate an increase in the number of patients under 50 years of age. This is probably due to the expansion of obesity and bad eating habits (31). A large cohort study by Wong Martin and colleagues examined the global incidence and mortality of gastric cancer and showed a decrease in the average of patients from 59.1 years (in 1980.) to 56.8 years (in 2018) (32). This supports the previous research conducted by Arnold, which indicated an increase in incidence in younger patients. Also, Wong Martin and colleagues point to a drastically faster death outcome and shorter survival rate, ranging from 1.3 to 25.8 years (in 1980.) to only 1.5 to 18.5 years (in 2018.). This study indicates an increase in the number of
patients under the age of 40 in certain countries, primarily Sweden, Great Britain and Ecuador (32). In the group of patients we examined, the recorded average age of developing cancer ranged from $70.14 \pm 9.07$ for gastric adenocarcinoma, to $66.93 \pm 10.19$ for signet ring cell carcinoma. Although we recorded a slightly younger age for developing signet ring cell type cancer, the results do not follow the trend of boundary moving towards a younger age of life, which is currently recorded in some countries. Although the incidence of the disease is different around the world, the population of Japan and Korea still lead in the number of developed gastric cancers. The reasons are probably eating habits, hygiene and higher prevalence of the East Asian form of CagA (cytotoxicity-associated genes) compared to the American countries where the western variant of CagA Helicobacter pylori is usually present (33). It is considered that Helicobacter pylori is an unavoidable factor in the development of gastric cancer. In 1982, Warren and Marshall discovered the association of Helicobacter pylori with the development of chronic gastritis (34). It is estimated that 85% of patients with developed gastric cancer also have a positive test for Helicobacter pylori (20). A large meta-analysis of 7 randomized studies by Ford and colleagues showed that eradication of Helicobacter pylori reduces the risk of disease by about 30.5% (35). This bacterium usually acts in two main ways: indirect effect on the gastric mucosa and potential inflammation, but also a direct action in the form of changing the function of epithelial cells, dominantly through the bacterial antigen CagA. Probably, both of these pathways are involved in the development of gastric cancer (36). Research in Shandong indicates that the use of proton pump inhibitors (PPIs) and amoxicillin significantly reduces the incidence of gastric cancer by 39% (in follow-up studies of 14.7 years). This confirms the effectiveness of antibiotic therapy against Helicobacter pylori (37). The recommendations of the last Maastricht VI from 2022 suggest that Helicobacter pylori is a key etiological factor for the development of proximal adenocarcinoma and carcinoma of the gastroesophageal junction (38). In our selected examined population, the presence of this bacterium was found in 59.1% of patients with gastric adenocarcinoma and in 86.7% patients with signet ring cell carcinoma. These results were statistically significant in relation to a certain type of cancer, which confirms the importance of this bacterium in the development of the disease. The eradication of H. pylori reduces the chance of further progression of atrophic gastritis towards metaplasia and dysplasia up to gastric cancer (38). In addition to H. pylori infection, the role of smoking and increased alcohol consumption in the development of cancer is huge (39). Cigarettes smoking is a known carcinogen responsible for the development of tumors in more than 20 locations. From all of 5200 known components of smoke, over 60 of them have direct carcinogenic effects tested in rodents and for over 10 components, there is strong evidence of carcinogenesis in humans (40). A study of the etiology of gastric cancer conducted in Shanghai by Cheng Xiao found an 80% higher risk of developing gastric cancer in smokers compared to a group of non-smokers (39). A meta-analysis of 32 studies conducted by Lopes-Ladeiras and co-workers indicates a 60% higher risk of disease in smokers compared to non-smokers (40). Ramos Marcus Fernando and colleagues studied the importance of smoking experience in the development of cancer and found that the risk of the disease was higher if the smoking status was longer and more cigarettes were smoked throughout the years (when the index number of cigarettes smoked-years is greater than 38) (41). Lindblad and co-workers noted that the former smoking status was important only in female smokers, while they did not establish such an association with gastric cancer in male population (30). In our examined patient population, a higher frequency of smoking was recorded in male patients (46.9%) compared to females (18.2%). Although there were more sick male smokers, there was no statistically significant difference in developing a certain type of cancer in these patients. Female smokers were equally likely to develop any type of gastric cancer. Nishino and colleagues confirm the hypothesis that smoking status increases the risk of the disease in both male and female, which was also the case with our examinees (42). In addition to smoking, alcohol consumption is an inevitable factor in the development of gastric cancer. A large cohort study done in Europe on a sample of 444 cases of gastric cancer found a positive association of frequent alcohol consumption with the occurrence of cancer, but this risk was significantly lower in people who drank less than 60 grams of alcohol per day. Alcohol is considered a risk factor for the development of primary non-cardiac gastric cancer (43). According to a survey conducted in Brazil by the World Health Organization (WHO), 20.4% of cancer patients were former alcohol users, while 58% of patients consumed alcohol at the time of diagnosis (44). A meta-analysis by Ma Ke and colleagues supports the hypothesis of an increase in gastric cancer incidence with increased alcohol intake (45). However, an increase in stomach and oesophageal cancer has been reported in the first two years after stopping alcohol consumption. The reason is probably the quitting of alcohol consumption only at the time of diagnosis or during the appearance of symptoms, which is why patients are identified as former drinkers (46). Out of a total of 43 of our respondents, 8 were male and only 1 was a female. The incidence of a certain type of cancer in these 9 patients was uniform and there was no statistically significant difference in relation to the type of developed cancer. In addition to the mentioned factors, some systemic diseases can be related to gastric cancer. Diabetes can be a predisposition to the development of cancer in several ways. Hyperglycemia can lead to direct DNA damage or the production of free radicals (47). This activates oxidative stress, which can result in mutations in oncogenes and tumor-suppressor genes. This kind of damage can be explained by the synergistic effect of diabetes and H. pylori infection.
A large cohort study conducted in 2020 by Yang Hyo-Joon and colleagues showed that diabetes can lead to mucosal atrophy and the development of intestinal metaplasia, which can be the basis for the development of cancer. This study also suggests more frequent endoscopic check-ups in those patients. In our observed population, there were only 7 patients (16.3%) suffering from diabetes mellitus, 3 male and 4 female patients. Although all these patients had a diagnosis of gastric cancer, it was not statistically significantly different according to the type of cancer. This indicates that diabetes can be a risk factor, but the number of such subjects is relatively small. Tseng Chin and colleagues, in their study of the potential relationship between diabetes and gastric carcinoma, indicate a positive correlation of this disease with the occurrence of cancer, more often in females in the Asian population. Research on this topic conducted in 2021 by Dabo Bashir and colleagues did not establish the relationship between diabetes and the occurrence of gastric cancer. These studies and the obtained connections between diabetes and the occurrence of gastric cancer are inconsistent, which is also the case with our patient population, so further research on this topic is required. Other risk factors and protective factors include: poor socioeconomic status (associated with a higher mortality rate), frequent use of non-steroidal anti-inflammatory drugs (NSAIDs, especially aspirin usage), use of statins (they reduce the risk of disease by about 30%), high body-mass index (BMI of 30-35, the risk of disease is 2 times higher, BMI over 40, the risk of disease is more than 3 times higher), increased physical activity (reduces the risk of developing cancer by 21%), and gastroesophageal reflux disease (2 to 4 times more common gastric cardia malignancy).

Conclusion

The purpose of this study of the selected population of patients was to determine the relationship between gender, age, Helicobacter pylori infection, smoking cigarettes, alcohol consumption and diabetes mellitus with the occurrence of two basic types of gastric cancer - adenocarcinoma and signet ring cell type. Although numerous worldwide studies indicate higher incidences of gastric cancer in male and more often in younger population, in smokers, chronic alcohol consumers and patients with diabetes mellitus, our study did not show statistically significant difference in relation to the type of cancer. Helicobacter pylori infection is one of the key factors in the development of both types of gastric cancer. Further research on a larger population of patients should aim at discovering important genetic factors, obtaining more precise data on the intake of vegetables, fruits and vitamins, analyzing the role of obesity and the role of systemic and inflammatory diseases. This would be necessary in order to clarify the etiology of gastric cancer in patients where the most common risk factors are not present.
References


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FAKTORI RIZIKA ZA RAZVOJ KARCINOMA ŽELUCA – ISKUSTVO JEDNOG CENTRA

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Karcinom želuca je četvrti najčešći karcinom na svetu, sa preko milion novih slučajeva godišnje i preko 750 000 smrtnih ishoda od karcinoma. Njegova incidencija beleži se u Japanu i Kini, istočnoj Evropi i Južnoj Americi, a najniža u Africi. Bolest je dva do tri puta češća kod muškaraca i kod osoba starih do 60 godina, ali se granica pomera ka mlađem uzrastu. Petogodišnje preživljavanje u evropskim zemljama je od 10% do 30%. Ovo je multifaktorijska bolest, a najvažniji faktori rizika su: infekcija bakterijom Helicobacter pylori, pušenje, konzumiranje alkohola, muški bol, starije životno doba, smanjen unos povrća i voća, genetska predispozicija, šećerna bolest i dr.

Prospektivno istraživanje sprovedeno je na odabranoj populaciji od 43 bolesnika sa dijagnozom dva najčešća tipa karcinoma želuca: adenokarcinom i signet ring cell tip karcinom. Od ukupnog broja ispitanika bilo je 32 muškarca i 11 žena, prosečnih godina starosti 68,83 godine ± 9,26 godina. Među obolelima bilo je 39,5% pušača, 20,9% konzumenata alkohola, 62,8% ispitanika sa nalazom Helicobacter pylori i 16,3% obolelih od šećerne bolesti. Analiza povezanosti pola, godina, pušenja, konzumiranja alkohola i šećerne bolesti sa tipom karcinoma nije pokazala statističku značajnost. Učestalost H. pylori statistički se značajno razlikovala u odnosu na tip karcinoma (p = 0,007).


Ključne reči: karcinom želuca, Helicobacter pylori, pušenje, alkohol, diabetes mellitus

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