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Repair gene Polymorphism XRCC2 and the Risk of Gastric Cancer in the Brazilian Amazon

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Abstract

Cancer occurs due to a malfunction in cell growth regulation. Gastric cancer is one the most prevalent types of cancer worldwide and is characterized by abnormal cell growth in the stomach. The XRCC2 gene is associated with DNA repair by homologous recombination and a faulty mechanism can lead to the development of several genetic diseases, such as cancer. Several types of cancer have been associated with polymorphisms in DNA repair genes, such as Arg188His of the XRCC2 gene. This study was aimed at examining the possible involvement of this polymorphism in the development of gastric cancer in patients in the municipality of Macapá-AP. PCR-RFLP analysis was carried out with the HphI enzyme (Haemophilus parahaemolyticus) to detect polymorphism in 30 gastric cancer patients and 55 control individuals. Our results revealed that 43.3% of patients had this polymorphism compared to 18.1% of the control group. Our findings demonstrated that the Arg188His polymorphism was statistically significant and therefore it was associated with gastric cancer in the study population, similar to other polymorphisms of repair genes already linked with this disease in the same population.

Key words: *Repair gene XRCC2; Arg188His; Gastric cancer; Macapa; Brazil*

Introduction

Cancer occurs due to a malfunction in cell growth regulation, initially causing a neoplasia or tumor [1]. In gastric cancer, abnormal cell growth may be present throughout the gastroenteric system, usually giving rise to lesions [2]. Studies have examined whether the large number of genetic polymorphisms found in the human genome can be associated with differences in protein expression capable of altering the cellular phenotype, increasing or not the risk of developing several disorders such as benign and malignant neoplasias [3].

During DNA replication, mechanisms of DNA repair act to prevent unintentional changes in the genome. One of them is performed by enzymes produced from genes such as XRCC2 [4,5] located in the long arm of chromosome 7, position 36.1, a member of family of the RAD51 gene [6]. The enzymes synthesized from this gene prevent DNA damage, decrease mutations, replication errors, and genomic instability.

Some polymorphisms occur due to variations in the DNA sequence of a single nucleotide (SNPs - single nucleotide polymorphisms) [7]. The SNP Arg188His in the XRCC2 gene is relatively rare and is located at the nucleotide 31479 in exon 3, where the wild-type G allele is

replaced by the mutant A allele, resulting in the substitution of the amino acid arginine (Arg) for histidine (His) [8].

National Cancer Institute (INCA) [9] estimates that 12,920 new cases of gastric cancer in men and 7,600 in women will be diagnosed, indicating the importance of molecular studies on this disease. This study was aimed at examining the possible involvement of the Arg188His polymorphism in the development of gastric cancer in the municipality of Macapa-AP, Brazil.

Materials and Methods

Biological Samples

The study population was represented by 85 DNA samples, of which 55 comprised the control group from the Institute of Hematology and Hemotherapy of Amapa (HEMOAP) and 30 from gastric cancer patients from the Dr. Alberto Lima Hospital (HCAL).

After individuals signed the Informed Consent Term (ICF), blood samples were collected and sent to the Laboratory of Molecular Biology and Biotechnology (BIOMOL) of the Federal University of Amapá (UNIFAP).

Identification of the XRCC2 Arg188His polymorphism

The protocol developed by Gök et al. [10] was used to identify the Arg188His polymorphism of the XRCC2 gene. PCR was performed with the primer pairs F5'-TGTAGTCACCCATCTCTCTGC-3' and R5'-AGTTGCTGCCATGCCTTACA-3'. The initial denaturation was carried out for 3 min at 95°C, followed by 35 cycles at 94°C for 30s, 57°C for 30s, 72°C for 45s and a final extension of 1 cycle at 72°C for 7 minutes.

The PCR amplification product of 290 bp was visualized in 1.5% agarose gel. The amplified fragment was digested with the restriction enzyme HphI, under the following conditions: 10 ul of the product, 2ul of H_2O, 2u of buffer, and 1ul of enzyme in a final volume of 15ul, at 37ºC for 16 hours. After the electrophoresis, the genotypes observed were wild homozygote (290 bp - Arg / Arg), heterozygote (148 bp + 142 bp - Arg / His) or the mutant homozygote (290 bp + 148 bp + 142 bp - His / His).

Statistical Analysis

Allele and genotype frequencies were analyzed with the Hardy-Weinberg equilibrium and the chi-square test (X2) using the software Bio Estat (Ayres, M. Pará, Brazil).

Results and Discussion

Of the 30 analyzed samples of gastric cancer patients, 43.3% (13) had the Arg188His and 56.6% had theArg188Arg genotypes, while in the 55 controls they accounted for 18.1% and 81.8% of samples, respectively (Table 1).

Regarding genotype frequency, 20% of the patients and 10.9% of the controls had the Arg188His genotype (2.6471 (0.7496-9.3478) p=0.2277). The frequency of the His188His genotype was 23.3% and 7.2% in patients and controls, respectively (4.6324 (1.2018-17.8560) p = 0.0446) (Table 2). Several studies have examined the association of Arg188His polymorphism and the development of several types of cancer. No association has been found with laryngeal cancer11 and variations in the coding region of the XRCC2 gene did not show any susceptibility to thyroid cancer [12,13].

Gastric cancer patients (n=30)					Control group (n=55)				
Gene	with SNP	%	no SNP	%	with SNP	%	no SNP	%	P value
XRCC2	13	43.3	17	56.6	10	18.1	45	81.8	0.0134

Table 1: *Frequency of the XRCC2 Arg188His polymorphism in gastric cancer patients and controls.*

Genotype	Patients	%	Control	%	OR (CI 95%)
XRCC2 (G188A)					Reference
Arg/Arg (GG)	17	56.6	45	81.8	
Arg/His (GA)	06	20	06	10.9	2.6471 (0.7496-9.3478) p=0.2277
His/His (AA)	07	23.3	04	7.2	4.6324 (1.2018-17.8560) p=0.0446

Table 2: *Genotype frequency of the XRCC2 Arg188His polymorphism in gastric cancer patients and controls.*

Studies on the association with the development of ovarian cancer reported contrasting results, partly due to the small effect of the polymorphism on the disease and the insufficient amount of samples analyzed [14,15]. On the other hand, studies involving the Arg188His polymorphism and cancer of head and neck in a population of India in 2014 found a highly significant association in individuals with the heterozygous genotype (Arg/His), which was then considered a risk factor for the disease [16]. Studies in France also showed an association between polymorphism and the development of oral and pharyngeal cancer in carriers of the His allele [11]. In patientswith gastric cancer in Turkey, the Arg188His polymorphism was observed in 39% of samples [10].

Our results were statistically significant, although this polymorphism is not directly associated to gastric cancer but to breast cancer [17]. Gastric cancer is one of the leading causes of death in the state of Amapá, with patients developing one of the most aggressive forms of the disease. External factors associated with the onset of this disease in the state are still poorly known. However, the study population has a genetic predisposition for the development of gastric cancer, as the Arg188His polymorphism is associated with this disease. Other DNA repair gene polymorphisms have also been significantly correlated with gastric cancer in this population.

The sample size in the present study was small, but our findings regarding the association of Arg188His polymorphisms with gastric cancer in the study population may still be relevant.

Consent

All authors declare that written informed consent was obtained from all the patient.

Ethical Approval

All authors hereby declare that all experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

Competing Interests

Authors have declared that no competing interests exist.

References

1. Borges-Osório MR, Robinson WM. (2013). Genética humana 3: 385-407.
2. Almeida VLD, Leitão A, Reina LDCB, Montanari CA, Donnici CL, Lopes MTP. (2005). Câncer e agentes antineoplásicos ciclo-celular específicos e ciclo-celular não específicos que interagem com o DNA: uma introdução. Química Nova 28(1), 118-129.
3. Xuan G, Hui Y, Fang H. (2015). The association of XRCC3 Thr241Met genetic variant with risk of prostate cancer: A meta-analysis. African Health Sciences. 117–122 p.
4. Varki A, Geschwind DH, Eichler EE. (2008). Human uniqueness: Genome interactions with environment, behaviour and culture. Nat Rev Genet 9: 749–63.
5. Zhao Y, Deng X, Wang Z, Wang Q, Liu Y. (2012). Genetic polymorphisms of DNA repair genes XRCC1 and XRCC3 and risk of colorectal cancer in chinese population. Asian Pacific J Cancer Prev 13: 665–9.
6. Tambini CE, George AM, Rommens JM, Tsui LC, Scherer SW, Thacker J. (1997). The XRCC2 DNA repair gene: Identification of a positional candidate. Genomics 41: 84–92.
7. Costa APP, Garcia AHC, Copreski B, Brunoni D. (2007). Estudo de Polimorfismos de DNA associados aos disturbios do desenvolvimento/ Studies of DNA polymorphism and developmental disorders. : 112–31.
8. Jiao L, Hassan MM, Bondy ML, Wolff RA, Evans DB, Abbruzzese JL, et al. (2008). XRCC2 and XRCC3 gene polymorphismand risk of pancreatic cancer. Am J Gastroenterol 103: 360–7.
9. Instituto Nacional De Câncer José Alencar Gomes Da Silva (Inca). Estimativa (2016): incidência de câncer no Brasil. Dísponivel em: http://www.inca.gov.br/estimativa/2016/estimativa-2016-v11.pdf (Acessada em 21/06/2017).
10. Gök İ, Baday M, Çetinkünar S, Kiliç K, Bilgin BÇ. (2014). Polymorphisms in DNA repair genes XRCC2 and XRCC3 risk of gastric cancer in Turkey. Bosn J Basic Med Sci 14: 214–8.
11. Benhamou S, Tuimala J, Bouchardy C, Dayer P, Sarasin A, Hirvonen A. (2004). DNA repair gene XRCC2 and XRCC3 polymorphisms and susceptibility to cancers of the upper aerodigestive tract. Int J Cancer 112: 901–4.
12. Fayaz S, Fard-Esfahani P, Fard-Esfahani A, Mostafavi E, Meshkani R, Mirmiranpour H, et al. (2012). Assessment of genetic mutations in the XRCC2 coding region by high resolution melting curve analysis and the risk of differentiated thyroid carcinoma in Iran. Genet Mol Biol 35: 32–7.
13. Yan L, Li Q, Li X, Ji H, Zhang L. (2016). Association studies between XRCC1, XRCC2, XRCC3 polymorphisms and differentiated thyroid carcinoma. Cell Physiol Biochem 38: 1075–84.

14. Du J, Lu C, Cui G, Chen Y, He J. (2013). DNA repair gene XRCC1 polymorphisms and susceptibility to childhood acute lymphoblastic leukemia: A meta-analysis. Chinese J Cancer Res 25: 405–15.
15. Zhu H, Cao S, Liu Y, Ding X, Wu Q, Ma H. (2013). Genetic polymorphisms of xeroderma pigmentosum group D and prostate cancer risk: A meta-analysis. J Cancer Res Ther 9: 187–92.
16. Saeed S, Mahjabeen I, Sarwar R, Bashir K, Kayani MA. (2017). Haplotype analysis of XRCC2 gene polymorphisms and association with increased risk of head and neck cancer. Sci Rep Springer US 7: 1–10.
17. Sirisena ND, Adeyemo A, Kuruppu AI, Samaranayake N, Dissanayake VHW. (2018). Genetic variants associated with clinicopathological profiles in sporadic breast cancer in Sri Lankan women. J Breast Cancer BMC Cancer 21: 165–72.