GSTP1 Ile105Val and GPX1 Pro198Leu Polymorphisms and Their Association with Response to Radiotherapy in Nasopharyngeal Carcinoma Patients

Rajaa Benzeid,1,2 Amina Gihbid,3 Nadia Benchekroun,4 Karima Bendahhou,4 Naima El Benna,5 Amal Guensi,6 Abdellatif Benider,4 Nezha Tawfiq,4 Abdelkarim Filali Maltouf,2 Mohammed El Mzibri,1 Mohammed Attaleb,1 Meriem Khyatti,3 Imane Chaoui1

1Biology Unit and Medical Research, National Center of Energy, Sciences and Nuclear Techniques, Rabat, Morocco
2Laboratory of Microbiology and Molecular Biology, Faculty of Sciences, Rabat, Morocco
3Laboratory of Viral Oncology, Pasteur Institute, Casablanca, Morocco
4Department of Radiotherapy-Oncology, Mohammed VI Center for Treatment of Cancer, Ibn Rochd Hospital Center, Casablanca, Morocco
5Department of Radiology, Ibn Rochd University Hospital, Hopital 20 Août, Casablanca, Morocco
6Department of Nuclear Medicine, Ibn Rochd University Hospital, Hassan II University, Casablanca, Morocco

Abstract

Objectives: Radio-resistance poses a major challenge in nasopharyngeal carcinoma (NPC) treatment. Due to individual variations in radio-sensitivity, biomarkers are needed to tailor radiation treatment. Within this frame, the identification of series of genetic signatures mainly SNPs for NPC patients treated with radiotherapy may help to predict treatment outcome and deliver personalized therapy. The aim of this study was to evaluate the possible association between the GSTP1 Ile105Val and GPX1 Pro198Leu polymorphisms and response to radiotherapy in NPC patients.

Methods: From September 2016 to October 2018, a total of 101 patients with confirmed NPC, recruited at Mohammed IV Center for Treatment of Cancer of Casablanca, underwent radiotherapy. DNA was extracted from peripheral blood. Genotyping of the GPX1 Pro198Leu and GPX1 Val105Leu polymorphisms was carried out by PCR amplification and DNA sequencing. SPSS was used to analyse the association of GSTP1 and GPX1 genotypes with clinico-pathological features and response to radiotherapy.

Results: The genotyping data revealed the presence of only two genotypes namely Pro/Pro (57.4%) and Pro/Leu (40.6%) for GPX1 gene. The allelic frequencies of C and T alleles were 78.7% and 21.3% respectively. For GSTP1 gene, the homozygous genotypes Val/Val and Leu/Leu were detected in 35.6% and 12.9% of patients respectively. The heterozygous genotype Val/Leu prevailed (51.5%). Allelic frequencies showed the presence of the two alleles A and G in 57.1% and 42.9% patients respectively. Statistical analysis failed to find any significant association between GSTP1 Val105Ile and GPX1 Pro198Leu genetic polymorphisms and socio-demographic and clinico-pathological features as well as response to radiotherapy (p>0.05).

Conclusion: Further research is warranted on the potential role of SNPs within antioxidant defines genes in radiotherapy response and to identify reliable predictive and non-invasive biomarkers for radio-resistance among NPC patients for personalised therapies.

Keywords: Nasopharyngeal carcinoma, genetic polymorphisms, GSTP1, GPX1, radio-resistance, prognosis

Cite This Article: Benzeid R, Gihbid A, Benchekroun N, Bendahhou K, El Benna N, Guensi A, et al. GSTP1 Ile105Val and GPX1 Pro198Leu Polymorphisms and Their Association with Response to Radiotherapy in Nasopharyngeal Carcinoma Patients. EJMO 2022;6(1):64–72.
asopharyngeal carcinoma (NPC) is a malignant epithelial neoplasm arising from the epithelial lining of the nasopharynx and poses a public health problem in many countries in Southeast Asia and north Africa. In Morocco, like other countries in the Mediterranean basin, the incidence of NPC is moderate, at 3-8 per 100 000 people.\cite{1, 2}

NPC is widely known for its multifactorial aetiology, including Epstein–Barr virus (EBV) infection, environmental factors and genetic susceptibility.\cite{3}

Scientific evidence has shown that NPC is highly sensitive to ionizing radiation. As such, radiotherapy (RT) is the main and the most effective treatment of NPC.\cite{4, 5} However, the main problem faced by clinicians is the growing number of radio-resistance, affecting the treatment outcome and reducing the overall survival (OS) of NPC patients, which suggest the involvement of genetic susceptibility in this radio-resistance.\cite{6} Radio-resistance is a complex biological process associated with abnormal DNA damage response, apoptosis, autophagy, gene mutations, cell cycle checkpoint and deregulated signalling pathways which hampered the success of RT.\cite{7} It's widely accepted that radiotherapy acts directly on cellular components through the ionizing radiations and indirectly through generated reactive oxygen species (ROS) in the cell matrix.\cite{8} Hence, persistence of ROS in the NPC environment will be of a great interest to produce DNA damage and induce apoptosis. Conversely, rapid detoxification and ROS elimination will be associated with a decrease RT efficacy.

Many genetic studies have been conducted and showed that Pi-class glutathione S-transferase (GSTP1) and glutathione peroxidase1 (GPX1) are the most proteins involved in cell detoxification and xenobiotic elimination. These two proteins play a key role in preventing the accumulation of xenobiotics in cells, reducing their damage and the occurrence of various diseases.

Growing interest was given to investigate the role of GSTP1 and GPX1 and their involvement in cancer development and resistance. GSTP1 enzyme, encoded by the GST pi gene, is an isozyme widely reported to be expressed in human epithelial tissues,\cite{9} that plays an important regulatory role in detoxification and anti-oxidative damage. GSTP1 is also involved in maintaining cell integrity and protecting DNA from genotoxic and cell-damaging molecules by catalysing the detoxification of reactive electrophilic compounds.\cite{10} Large studies were conducted on the association between genetic polymorphisms in GSTP1 gene and cancer development, including NPC, and have focused on some specific polymorphisms like rs1695 (Ile105Val) and rs1138272 (Ala114Val) that alter the catalytic activity of the protein. Indeed, polymorphisms in GSTP1 gene have been reported to be associated with bladder and testicular cancers,\cite{11} head and neck cancers,\cite{12} hepatocarcinoma,\cite{13} lung cancer,\cite{14} oral carcinoma\cite{15} and breast cancer.\cite{16, 17}

GPX1 is a selenium-dependent enzyme highly involved in cell antioxidant defence by protecting cells and their environments from oxidative damage. GPX1, encoded by GPX1 gene, is able to eliminate the hydrogen peroxide ($H_2O_2$) through its reduction into water.\cite{18} In GPX1 gene, a wide interest is given to rs1050450C $>$ T polymorphism, resulting on proline (Pro) to leucine (Leu) amino acid substitution at codon 198 (Pro198Leu), close to the C-terminus of the protein.\cite{19} GPX1 Pro198Leu SNP, has been shown to decrease the enzymatic activity of glutathione peroxidase in the cells expressing the mutant protein, which further can increase the oxidative damage.\cite{20} GPX1 Pro198Leu polymorphism has been shown to be associated with colorectal and gastrointestinal cancers in relationship with exposure at various environmental factors.\cite{21, 22} A recent meta-analysis has reported a positive correlation between GPX1 Pro198Leu SNP and the development of bladder, head and neck and brain cancers.\cite{23}

There are limited data addressing the association of genetic polymorphisms in GSTP1 and GPX1 genes with tumour response to radiotherapy and/or chemotherapy. Accordingly, an association between GSTP1 polymorphism and treatment outcome was reported in breast cancer,\cite{24} bladder cancer\cite{25} and NPC,\cite{26} providing accumulating evidence of the role of genetic variants in predicting response to radio and/or chemotherapy in cancer patients. Data regarding genetic background of radio-resistance-related SNPs in NPC are scarce. So, we designed this study to investigate the association of Pro198Leu and Ile105Val polymorphisms in detoxification genes namely GPX1 and GSTP1 with response to radiotherapy in NPC patients and thus their potential use as biomarkers for better management of NPC in Morocco.

**Methods**

**Study Design**

A total of 101 histologically confirmed NPC patients were recruited at Mohammed IV Center for Treatment of Cancer of Casablanca, between September 2016 and October 2018. Patients with previous cancer and those who had previous chemotherapy or radiotherapy treatments were excluded from this study.

A questionnaire was administrated to each patient to collect information regarding the socio-economic status (age, sex, familial history of cancer, tobacco habits, alcohol consumption and childhood habitat). Clinical data including
histological type and clinical stage were retrieved from their medical records.

Patients were given a median dose of 50 Gy in the nasopharyngeal tumour area and 66 Gy in the involved lymph nodes. A daily fraction of 2 Gy was delivered five times per week and the treatment was completed within 5 to 7 weeks. For patients with an advanced stage of the disease, neoadjuvant chemotherapy was administered using different regimens based on cisplatin, anthracycline and fluorouracil.

Clinical staging of NPC was determined according to the 7th edition of the International Union Against Cancer (IUAC) and American Joint Committee on Cancer (AJCC) staging system. The follow-up was done at 3 month intervals for the first two years and six monthly thereafter. All patients were subjected to peripheral blood sampling before initiation of the treatment for plasmatic cell-free DNA extraction. The study protocol was approved by the Ethics Committee of Ibn Rochd Hospital of Casablanca, Morocco; and a written informed consent was obtained from each patient before its enrolment.

DNA Extraction

Genomic DNA was extracted from leukocyte nuclei using phenol chloroform DNA extraction method. DNA concentration and purity were assayed using the NanoDrop 2000 UV-Vis Spectrophotometer (Thermo Fisher Scientific). DNA was used immediately for PCR amplification or stored at -20°C until use.

**GPX1 Pro198Leu and GSTP1 Ile105Val Genotyping**

Pro198Leu and Ile105Val polymorphisms screening were carried out by PCR amplification and DNA sequencing. For GPX1Pro198Leu, primers flanking a region in exon 2 of GPX1 gene containing the SNP rs1050450 C>T were used. For GSTP1 Ile105Val, primers flanking a region in exon 6 of GSTP1 gene harboring the SNP rs1695 A>G were used. The primers sequences of the two SNPs and their characteristics are shown in Table 1.

PCR amplification was performed in a total volume of 25μl, containing 1X PCR buffer, 200µM of each dNTP, 200nM of each primer, 1.5 mM MgCl2, 0.5U Platinum Taq DNA polymerase (Invitrogen) and 50ng of genomic DNA. The mixtures were first denatured at 94°C for 7 min. Then, 40 cycles of PCR were performed with denaturation at 94°C for 30 s, primer annealing at 60°C for 30 s and primer extension for 30 s at 72°C. At the end of the last cycle, amplicons were incubated at 72°C for 7 min. For each PCR run, a negative control, in which DNA template was omitted from the amplification mixture, was included. GSTP1 and GPX1 fragments’ amplification was checked by DNA size separating by electrophoresis on 2% agarose gel and visualization under ultraviolet light.

PCR products were then purified using ExoSAP-IT® (Applied Biosystems) to eliminate the primers and unincorporated remaining dNTPs. Sequencing of purified PCR products was done with BigDye Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems). Sequencing reactions were performed in a final volume of 10μl, containing 1μl of 2.5X Big Dye ready reaction mix v3.1, 10 pmol of forward or reverse primer and 100ng of purified PCR product. Sequencing mixtures were first incubated at 96°C during 1 minute, then followed 25 cycles of denaturation at 96°C for 10s, primer annealing at 50°C for 5s and extension at 60°C for 4 min. The sequencing reaction products were then purified using Sephadex G-50 gel-exclusion chromatography (GE Healthcare Life Sciences) to eliminate the excess of ddNTPs. Direct sequencing of amplified DNA was performed by capillary electrophoresis on an ABI 3130xL Genetic Analyzer (Applied Biosystems). The sequences were analyzed using Sequencing Analysis v5.4 software (Applied Biosystems).

**Statistical Analysis**

Statistical analyses were performed using Software Package for Social Sciences (SPSS) version 23.0. The differences were considered significant if the p value did not exceed 0.05. Allelic frequency and genotypic frequencies were also calculated using the corresponding formula.

**Results**

The socio-demographic, clinical and pathological characteristics of all cases are summarized in Table 2. The median age of the recruited NPC patients was 54 years old,
ranging from 12 to 80 years old. In this cohort, males’ patients prevailed (60%) with sex ratio of 1.5. Smoking and alcoholic habits concerned 31.68% and 9.9% of the patients respectively. The clinico-pathological tumor characteristics revealed that 87.13% of patients were at advanced stages (III and IV), and most patients exhibited the undifferentiated type (89.1%). TNM staging revealed that 75% of patients were classified in T3-T4 categories and up to 34.6% of patients presented distant metastasis (M1). No involvement of regional lymph nodes was observed in 7.9% of patients.

In this study, we identified SNPs (rs1050450 and rs1695) by PCR-Sequencing within NPC patients.

### Table 2. Socio-demographic, clinical and pathological characteristics of NPC patients

| Parameter                  | Total cases | Percentage (%) |
|---------------------------|-------------|----------------|
| Gender                    |             |                |
| Male                      | 61          | 60.39          |
| Female                    | 40          | 39.61          |
| Age                       |             |                |
| ≤30                       | 23          | 23.76          |
| >30                       | 78          | 76.24          |
| Smoking habit             |             |                |
| Smoke                     | 32          | 31.68          |
| Non-smoke                 | 69          | 68.32          |
| Alcoholic habit           |             |                |
| Alcoholic                 | 10          | 9.9            |
| Non-Alcoholic             | 91          | 90.1           |
| Tumour stage              |             |                |
| I-II                      | 13          | 12.87          |
| III-IV                    | 88          | 87.13          |
| Histological type         |             |                |
| Type I                    | 1           | 0.99           |
| Type II                   | 7           | 6.93           |
| Undifferentiated type     | 90          | 89.1           |
| Other types               | 3           | 2.97           |
| TNM                       |             |                |
| Tumoral extension         |             |                |
| T1-T2                     | 24          | 23.8           |
| T3-T4                     | 77          | 76.2           |
| Regional lymph node involvement  |  |                |
| N=0                       | 8           | 7.9            |
| N≥1                       | 93          | 92.4           |
| Metastasis                |             |                |
| M0                        | 65          | 75.2           |
| M1                        | 36          | 24.8           |

Genotypes Distribution and Allelic Frequencies

For GPX1 gene, only two genotypes were detected namely Pro/Pro (57.4%) and Pro/leu (40.6%). The homozygous genotype (Leu/Leu) of the GPX1 variant was not found. The allelic frequencies of C and T alleles were 78.7% and 21.3% respectively.

For GSTP gene, the heterozygous genotype Val/Leu prevailed (51.5%). The homozygous genotypes Val/Val and Leu/Leu were detected in 35.6% and 12.9% of patients respectively. Allelic frequencies showed the presence of the two alleles A and G in 57.1% and 42.9% patients respectively (Table 3).

### Association between SNPs and Socio-Demographic & Clinico-Pathological Parameters

The genotypes distributions of GPX1 Pro198Leu and GSTP1 Val105Ile polymorphisms according to socio-demographic and clinico-pathological characteristics of NPC patients are summarized in Tables IV and V respectively. The expressions of genotypes were unrelated to gender, age, and clinical stage. Overall, no association was detected between GPX1 Pro198Leu genetic variants and any of the studied parameter. Also, Statistical analysis failed to find any significant association between GSTP Val105Ile polymorphism and socio-demographic and clinico-pathologic features.

Among the 101 diagnosed NPC patients, 67.3% (68/101) underwent radiotherapy and among them, 86% had a complete remission. Although C allele was four times more prevalent than T allele for GPX1 Pro198Leu polymorphism, statistical analysis failed to find any significant association between this polymorphism and radiotherapy response (p>0.05) (Table 4).

For GSTP1 Ile105Val polymorphism, we noted that both alleles had same frequencies within radio-resistant patients. Despite the prevalence of heterozygous genotypes AG within radiosensitive patients, no significant association was found between this polymorphism and response to radiotherapy (p>0.05) (Table 5).

### Table 3. Genotypes distribution and allelic frequencies within NPC patients

| Gene/ Genotype | No. Of patients | % | Allele | % |
|----------------|-----------------|---|--------|---|
| GPX1           |                 |   |        |   |
| CC             | 58              | 57.4 | C     | 78.7 |
| CT             | 43              | 42.6 | T     | 21.3 |
| TT             | 0               | 0   |       |    |
| GSTP1          |                 |   |        |   |
| AA             | 36              | 35.6 | A     | 57.1 |
| AG             | 52              | 51.5 | G     | 42.9 |
| GG             | 13              | 12.9 |       |    |
Discussion

NPC is a malignant disease with skewed ethnic and geographic distribution. The deep-seated anatomical position of NPC made this malignity not accessible by surgery and thus the principal treatment approach is radiotherapy. Although NPC is highly radio-sensitive, patients are prone to recur loco-regionally and to develop distant metastasis.

Radio-resistance hampers the success of treatment of NPC and multiple signalling pathways are involved in this phenomenon. Hence, deciphering new molecular targets and pathways is essential for enhancing and promoting the radio-sensitivity of NPC patients since clinical TNM staging has limited accuracy in NPC response prediction. Despite progress towards molecular mechanisms underlying radiotherapy response, there are no effective biomarkers for NPC radio-sensitivity/radio-resistance. Up to date, there is no standard test available that may reliably predict radio-sensitivity for cancer patients. Due to individual variations in radio-sensitivity, biomarkers are selected to tailor radiation treatment to NPC. Hence, there is a need to identify genes involved in radio-resistance along with the biological process contributing to the development of radio-resistance.

Extensive studies reported that some genetic polymorphisms have been implicated in increased susceptibility to NPC. Of interest, several studies have evaluated the association between GSTP1 c.105A>G and GPX1 c.198C>T polymorphisms and risk of cancer development with type specific effects, however, these studies yielded conflicting results.

By establishing a radio-resistant cell line, Guo et al. identified a panel of candidate's genes including GSTP1 whom...
might become a potential biomarker in predicting NPC response to radiotherapy.\[^{31}\] In this study, we identified c.105A>G SNP in GSTP1 gene within NPC patients and the results clearly showed that this polymorphism had no relevant effect neither on clinic-pathological features nor with radiotherapy response. Likewise, in a recent study, no association was detected between this polymorphism and primary tumour site, clinicopathological characteristics, treatment (chemotherapy and/or radiotherapy) and survival time of the head and neck cancer (HNC) patients.\[^{42}\] Moreover, a meta-analysis performed by Lang et al. failed to qualify c.105A>G SNP in GSTP1 gene as risk for developing HNC in many studies.\[^{43}\]

To the best of our knowledge, no study has investigated the association between this SNP and RT response in NPC. The variant genotype was reported to be linked with CT and RT responses and side effects in different malignancies. Pasqualetti et al. have shown that this SNP was associated with different OS in glioblastoma patients treated with RT and temozolamide.\[^{44}\] Also, it was reported that this SNP might be a useful biomarker for prediction of clinical benefit and toxicities of platinum-based chemotherapy in Thai epithelial ovarian cancer.\[^{45}\] In contrast, Deng et al. found that patients carrying GSTP1 variant genotypes were rather at lower risk of recurrence following an intravesical therapy and thus, suggested that c.105A>G SNP may predict chemotheraphy outcome in patients with bladder cancer (BC) and may, therefore, serve as molecular marker to monitor BC recurrence.\[^{46}\] Similarly, in a meta-analysis performed by Shen et al. GSTP1 Ile105Val polymorphism was associated with tumour response, progression free survival, and OS in gastric and colorectal cancers after CT.\[^{47}\]

Accumulating evidence implicates that GPX1 polymorphism (rs1050450) contributes to tumour susceptibility to cancer; individuals carrying variant Leu allele (Pro/Leu and Leu/Leu) were associated with an increased risk of cancer.\[^{47, 48}\] In NPC, the association between became evidenced; Laribi et al. found strong association between GPX1 Pro198Leu genetic variant and NPC risk. Moreover, this variant increased the risk of progression to regional lymph node metastasis.\[^{49}\] Dequanter et al. found that GPX1 low expressing HNSCC were at T3-T4 clinical stages and that GPX1 expression does not influence neither the response to radiotherapy nor the survival.\[^{50}\] In contrast, in the present study, the expressions of GPX1 Pro198Leu genotypes in patients with NPC were irrelevant to gender, age, clinical stage and response to RT.

Indeed, studies on the relationships between genetic polymorphisms in radio-resistance related genes and radiotherapy sensitivity in NPC have been performed.\[^{31, 49}\] Ban et al. suggest Wnt/beta-catenin as a novel prognostic factor for NPC patients treated with RT.\[^{51}\] More specifically, rs18880481 and rs3864004 polymorphisms in the catenin beta 1 gene (CTNNB1) and rs3755557 polymorphism in the glycogene synthase kinase 3 beta gene (GSK3beta), were significantly associated with a poorer efficacy of RT in NPC patients.\[^{52}\] Wang et al. have investigated the relationship between SNPs (G2677T exon 21 and C3435T in exon 26) in human multidrug resistance 1 gene (MDR) and radio-sensitivity and found that patients with 2677G-3435C haplotype had better response to radiotherapy compared to other haplotypes.\[^{53}\]

Very recently, inflammation-associated gene SNPs were found to be associated with increased radiotherapy sensitivity of HNSCC patients.\[^{54}\] Likewise, Seibold et al. reported that variants in oxidative stress-related genes may affect radiotherapy response.\[^{55}\] However, in NPC, the relationship between SNPs in detoxification related genes and the response of NPC patients to radiotherapy has not been reported.

Plenty of studies were performed to access the potential role of GPX1 and GSTP1 SNPs in the risk of Cardiovascular diseases\[^{56}\] and solid tumours\[^{57}\] but studies on their contribution to predict (chemo)radiotherapy treatment outcomes in NPC patients are very scarce. Treatment outcome is difficult to set; it is hampered by heterogeneity of tumours. Indeed, patients with same clinical stages, according to the TMN staging system, have different outcomes albeit receiving similar treatments. Recently, accumulated knowledge on tumour behaviour as well as response to cancer treatment has led to emergence of radiogenomics which links genetic variation with response to radiotherapy to improve decision making and thus patient outcomes.\[^{58}\]

Overall, the present study failed to statistically establish the associations between the well characterized functional GSTP1 c.105A>G and GPX1 c.198C>T polymorphisms within NPC patients and response to radiotherapy. Further studies with larger sample size and targeting additional polymorphisms may better demonstrate the interaction of GSTP1 & GPX1 germline polymorphisms as well different haplotypes and response to the radiotherapy and then select potential candidate biomarkers to predict NPC response to radiotherapy for personalized therapeutic strategies in the future.

This study was funded by the Institut de Recherche sur le Cancer (IRC) Ref: 201932.

Disclosures

Aknowledgements: The authors are grateful to Mohammed VI Center for Cancer Treatment staff for patients’ recruitment, blood collection and follow up of patients.
References

1. Benider A, Harif M, Karkouri M, Quessar A, Sahraoui S, Sqalli S. Registre des Cancers de la région du Grand Casablanca pour la période 2008–2012. Lalla Salma Foundation; 2016. Available at: https://www.contrelecancer.ma/site_media/upload_files/RCRGC.pdf. Accessed Mar 1, 2022.

2. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 2018;68:394–424.

3. Shannon-Lowe C, Rickinson A. The global landscape of EBV-associated tumors. Front Oncol 2019;9:713.

4. Lee HM, Okuda KS, González FE, Patel V. Current perspectives on nasopharyngeal carcinoma. Adv Exp Med Biol 2019;1164:11–34.

5. Aksoy A, Etkiran ET, Harputluoglu H, Dagli AF, Isikdogan A, Urakci Z. Is excision repair cross-complementation Group1 expression a biological marker in nasopharynx carcinoma? J Cancer Res Ther 2019;15:550–5.

6. Li W, Chen J, Liu C. Glutathione S-transferase P1 Ile105Val polymorphism and oral cancer risk: a meta-analysis. Int J Med Sci 2013;10:392–8.

7. Ryczaj K, Tang DG. Cancer stem cells and radioresistance. Int J Radiat Biol 2014;90:615–21.

8. Ameziane-El-Hassani R, Boufraqech M, Lagente-Chevallier O, Weyemi U, Talbot M, Mètivier D, et al. Role of H2O2 in RET/PtC1 chromosomal rearrangement produced by ionizing radiation in human thyroid cells. Cancer Res 2010;70:4123–32.

9. Baroudi O, Benammar-Elgaaied A. Involvement of genetic factors and lifestyle on the occurrence of colorectal and gastric cancer. Crit Rev Oncol Hematol 2012;13:264–71.

10. Al-Eitan LN, Rababa'h DM, Alghamdi MA, Khasawneh RH. Association Of GSTM1, GSTT1 And GSTP1 Polymorphisms With Breast Cancer Among Jordanian Women. Onco Targets Ther 2019;12:7757–65.

11. Morita S, Yano M, Tsujinaka T, Akiyama Y, Taniguchi M, Kanciko K, et al. Genetic polymorphisms of drug-metabolizing enzymes and susceptibility to head-and-neck squamous-cell carcinoma. Int J Cancer 1999;80:685–8.

12. Chen YL, Tseng HS, Kuo WH, Yang SF, Chen DR, Tsai HT. Glutathione S-Transferase P1 (GSTP1) gene polymorphism increases age-related susceptibility to hepatocellular carcinoma. BMC Med Genet 2010;11:46.

13. Ding F, Li JP, Zhang Y, Qi GH, Song ZC, Yu YH. Comprehensive Analysis of the Association Between the rs1138272 Polymorphism of the GSTP1 Gene and Cancer Susceptibility. Front Physiol 2019;9:1897.

14. Yadav P, Banerjee A, Boruah N, Singh CS, Chatterjee P, Mukherjee S, et al. Glutathione S-transferasesP1 AA (105Ile) allele increases oral cancer risk, interacts strongly with c-jun kinase and weakly detoxifies areca-nut metabolites. Sci Rep 2020;10:6032.

15. Fardmohammadi A, Arab-Yarmohammadi V, Ramzanpour R. Association analysis of rs1695 and rs1138272 variations in GSTP1 gene and breast cancer susceptibility. Asian Pac J Cancer Prev 2020;21:1167–72.

16. Valko M, Rhodes CJ, Moncol J, Izakovic M, Mazur M. Free radicals, metals and antioxidants in oxidative stress-induced cancer. Chem Biol Interact 2006;160:1–40.

17. Wang C, Zhang R, Chen N, Yang L, Wang Y, Sun Y, et al. Association between glutathione peroxidase-1 (GPX1) and 8-oxoguanine glycosylase 1 (hOGG1) on lung cancer. J Prev Med Public Health 2006;39:130–4.

18. Ji M, Tang J, Zhao J, Xu B, Qin J, Lu J. Polymorphisms in genes involved in drug detoxification and clinical outcomes of anthracycline-based chemotherapy in Chinese Han breast cancer patients. Cancer Biol Ther 2012;13:264–71.

19. Baroudi O, Benammar-Elgaaied A. Involvement of genetic factors and lifestyle on the occurrence of colorectal and gastric cancer. Crit Rev Oncol Hematol 2016;107:72–81.

20. Pérez S, Taléns-Visconti R, Rius-Pérez S, et al. Redox signaling in the gastrointestinal tract. Free Radic Biol Med 2017;104:75–103.

21. Wang C, Zhang R, Chen N, Yang L, Wang Y, Sun Y, et al. Association between glutathione peroxidase-1 (GPX1) Rs1050450 polymorphisms and cancer risk. Int J Clin Exp Pathol 2017;10:9527–40.

22. Tulsyan S, Chaturvedi P, Agarwal G, Lal P, Agrawal S, Mittal RD, et al. Pharmacogenetic influence of GST polymorphisms on anthracycline-based chemotherapy responses and toxicity in breast cancer patients: a multi-analytical approach. Mol Diagn...
25. Deng X, Yang X, Cheng Y, Liu X, Li X, Zhao R, et al. GSTP1 and GSTO1 single nucleotide polymorphisms and the response of bladder cancer patients to intravesical chemotherapy. Sci Rep 2015;5:14000.

26. Pincinato EC, Costa EFD, Lopes-Aguiar L, Nogueira GAS, Lima TRP, Visacri MB, et al. GSTM1, GSTT1 and GSTP1 Ile105Val polymorphisms in outcomes of head and neck squamous cell carcinoma patients treated with cisplatin chemoradiation. Sci Rep 2019;9:9312.

27. Edge SB, Compton CC. The American Joint Committee on Cancer: the 7th edition of the AJCC cancer staging manual and the future of TNM. Ann Surg Oncol 2010;17:1471–4.

28. Petersson F. Nasopharyngeal carcinoma: a review. Semin Diagn Pathol 2015;32:54–73.

29. Holleczek B, Stegmaier C, Radosa JC, Solomayer EF, Brenner H. Brenner H. Risk of loco-regional recurrence and distant metastases of patients with invasive breast cancer up to ten years after diagnosis - results from a registry-based study from Germany. BMC Cancer 2019;19:520.

30. Zhan Y, Fan S. Multiple mechanisms involving in radioresistance of nasopharyngeal carcinoma. J Cancer 2020;11:4193–204.

31. Guo Y, Zhu XD, Qu S, Li L, Su F, Li Y, et al. Identification of genes involved in radioresistance of nasopharyngeal carcinoma by integrating gene ontology and protein-protein interaction networks. Int J Oncol 2012;40:85–92.

32. Drobin K, Marczyk M, Halle M, Danielsson D, Papiez A, Sangsusan T, et al. Molecular profiling for predictors of radiosensitivity in patients with breast or head-and-neck cancer. Cancers (Basel) 2020;12:753.

33. Jiang Y, Li N, Dong P, Zhang N, Sun Y, Han M, et al. Polymorphisms in GSTM1, GSTT1 and GSTP1 and nasopharyngeal carcinoma in the East of China: a case-control study. Asia Pac J Cancer Prev 2011;12:3097–100.

34. Cao M, Mu X, Jiang C, Yang G, Chen H, Xue W. Single-nucleotide polymorphisms of GPX1 and MnSOD and susceptibility to bladder cancer: a systematic review and meta-analysis. Tumour Biol 2014;35:759–64.

35. Ratnasinghe D, Tangrea JA, Andersen MR, Barrett MJ, Virtamo J, Taylor PR, et al. Glutathione peroxidase codon 198 polymorphism variant increases lung cancer risk. Cancer Res 2000;60:6381–3.

36. Ichimura Y, Habuchi T, Tsuchiya N, Wang L, Oyama C, Sato K, et al. Increased risk of bladder cancer associated with a glutathione peroxidase 1 codon 198 variant. J Urol 2004;172:728–32.

37. Hu J, Zhou GW, Wang N, Wang YJ. GPX1 Pro198Leu polymorphism and breast cancer risk: a meta-analysis. Breast Cancer Res Treat 2010;124:425–31.

38. Hansen RD, Krath BN, Frederiksen K, Tjønneland A, Overvad K, Roswall N, et al. GPX1 Pro(198)Leu polymorphism, erythrocyte GPX activity, interaction with alcohol consumption and smoking, and risk of colorectal cancer. Mutat Res 2009;664:13–9.

39. Erdem O, Eken A, Akay C, Arsova-Sarafinovska Z, Matevska N, Suturkova L, et al. Association of GPX1 polymorphism, GPX activity and prostate cancer risk. Hum Exp Toxicol 2012;31:24–31.

40. Yu Z, Li Z, Cai B, Wang Z, Gan W, Chen H, et al. Association between the GSTP1 Ile105Val polymorphism and prostate cancer risk: a systematic review and meta-analysis. Tumour Biol 2013;34:1855–63.

41. Chatterjee A, Gupta S. The multifaceted role of glutathione S-transferases in cancer. Cancer Lett 2018;433:33–42.

42. Maniglia MP, Russo A, Biselli-Chicote PM, Oliveira-Cucolo JG, Rodrigues-Fleming GH, Maniglia JV, et al. Glutathione S-transferase polymorphisms in head and neck squamous cell carcinoma treated with chemotherapy and/or radiotherapy. Asian Pac J Cancer Prev 2020;21:1637–44.

43. Lang J, Song X, Cheng J, Zhao S, Fan J. Association of GSTP1 Ile105Val polymorphism and risk of head and neck cancers: a meta-analysis of 28 case-control studies. PLoS One 2012;7:e48132.

44. Pasqualetti F, Gonnelli A, Cantarella M, Delishaj D, Molinari A, Ortenzi V, et al. Association of Glutathione S-Transferase P-1 (GSTP-1) rs1695 polymorphism with overall survival in glioblastoma patients treated with combined radio-chemotherapy. Invest New Drugs 2018;36:340–5.

45. Liblab S, Vusuratana A, Areepium N. ERCC1, XRCC1, and GSTP1 Polymorphisms and treatment outcomes of advanced epithelial ovarian cancer patients treated with platinum-based chemotherapy. Asian Pac J Cancer Prev 2020;21:1925–9.

46. Shen X, Wang J, Yan X, Ren X, Wang F, Chen X, et al. Predictive value of GSTP1 Ile105Val polymorphism in clinical outcomes of chemotherapy in gastric and colorectal cancers: a systematic review and meta-analysis. Cancer Chemother Pharmacol 2016;77:1285–302.

47. Chen J, Cao Q, Qin C, Shao P, Wu Y, Wang M, et al. GPx-1 polymorphism (rs1050450) contributes to tumor susceptibility: evidence from meta-analysis. J Cancer Res Clin Oncol 2011;137:1553–61.

48. Janicka A, Szymańska-Pasternak J, Bober J. Polymorphisms in the oxidative stress-related genes and cancer risk. Ann Acad Med Stetin 2013;59:18–28.

49. Laribi A, Aouf S, Gabbouj S, Bouaouinaa N, Zakhama A, Harizi E. Human glutathione peroxidase codon 198 polymorphism and risk of head and neck cancers: evidence from meta-analysis. J Cancer Res Clin Oncol 2011;137:1553–61.

50. Dequanter D, Dok R, Koolen L, Vander Poorten V, Nuyts S. Prognostic significance of glutathione peroxidase levels (GPx1) in head and neck cancers. Front Oncol 2017;7:84.

51. Ban S, Konomi C, Iwakawa M, Yamada S, Ohno T, Tsuji H, et al. Radiosensitivity of peripheral blood lymphocytes obtained...
from patients with cancers of the breast, head and neck or cervix as determined with a micronucleus assay. J Radiat Res 2004;45:535–41.

52. Yu J, Huang Y, Liu L, Wang J, Yin J, Huang L, et al. Genetic polymorphisms of Wnt/β-catenin pathway genes are associated with the efficacy and toxicities of radiotherapy in patients with nasopharyngeal carcinoma. Oncotarget 2016;7:82528–37.

53. Wang ZY, Chen LH, Fan Q, Yan WP, Chen YQ, Li QS, et al. Relationship between radiosensitivity of nasopharyngeal carcinoma and MDR1 gene polymorphism. Nan Fang Yi Ke Da Xue Xue Bao 2007;27:580–3.

54. Li Y, Zhu L, Yao H, Zhang Y, Kong X, Chen L, et al. Association of inflammation-related gene polymorphisms with susceptibility and radiotherapy sensitivity in head and neck squamous cell carcinoma patients in Northeast China. Front Oncol 2021;11:651632.

55. Seibold P, Hall P, Schoof N, Nevanlinna H, Heikkinen T, Benner A, et al. Polymorphisms in oxidative stress-related genes and mortality in breast cancer patients—potential differential effects by radiotherapy? Breast 2013;22:817–23.

56. Zhang JX, Wang ZM, Zhang JJ. Association of glutathione peroxidase-1 (GPx-1) rs1050450 Pro198Leu and Pro197Leu polymorphisms with cardiovascular risk: a meta-analysis of observational studies. J Geriatr Cardiol 2014;11:141–50.

57. Zmorzyński S, Świderska-Kołacz G, Koczkodaj D, Filip AA. Significance of polymorphisms and expression of enzyme-encoding genes related to glutathione in hematopoietic cancers and solid tumors. Biomed Res Int 2015;2015:853573.

58. Zhang CD, Yang Y, Chen HH, Zhang T, Wang Q, Liang Y, et al. RTPDB: a database providing associations between genetic variation or expression and cancer prognosis with radiotherapy-based treatment. Database 2018;2018:bay118.