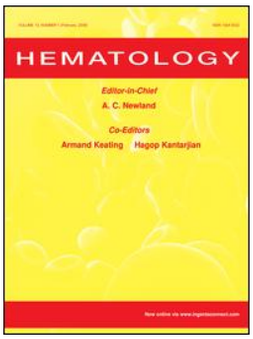

Hematology

ISSN: (Print) 1607-8454 (Online) Journal homepage: www.tandfonline.com/journals/yhem20

Polymorphic variants of *GSTM1*, *GSTT1*, and *GSTP1* genes in childhood acute leukemias: A preliminary study in Argentina

N. Weich, M. C. Nuñez, G. Galimberti, G. Elena, S. Acevedo, I. Larripa & A. F. Fundia

To cite this article: N. Weich, M. C. Nuñez, G. Galimberti, G. Elena, S. Acevedo, I. Larripa & A. F. Fundia (2015) Polymorphic variants of *GSTM1*, *GSTT1*, and *GSTP1* genes in childhood acute leukemias: A preliminary study in Argentina, Hematology, 20:9, 511-516, DOI: 10.1179/1607845415Y.0000000007

To link to this article: https://doi.org/10.1179/1607845415Y.0000000007

Published online: 23 Mar 2015.

Polymorphic variants of *GSTM1*, *GSTT1*, and *GSTP1* genes in childhood acute leukemias: A preliminary study in Argentina

N. Weich[1], M. C. Nuñez[2], G. Galimberti[3], G. Elena[3], S. Acevedo[4], I. Larripa[1], A. F. Fundia[1]

[1]Hematological Genetics Laboratory, Instituto de Medicina Experimental (IMEX), CONICET – Academia Nacional de Medicina, Buenos Aires, Argentina, [2]Mathematics Department, School of Pharmacy and Biochemistry, Universidad de Buenos Aires, UBA, Argentina, [3]Hematology-Oncology Unit, Hospital General de Niños Pedro Elizalde, Buenos Aires, Argentina, [4]Genetics Department, Instituto de Investigaciones Hematológicas (IIHema), Academia Nacional de Medicina, Buenos Aires, Argentina

Background and Aim: Despite recent major advances in leukemia research, the etiopathogenesis of childhood leukemias remains far elusive. Individual predisposing factors, including polymorphisms in detoxification enzymes, have been implicated in the molecular pathogenesis and heterogeneity of the disease. Genetic polymorphisms of glutathione S-transferases (GSTs) that alter enzyme activity could be an additional factor that increases the risk of acute leukemia, but data are lacking in Argentina. We assessed the association of GST polymorphisms and the susceptibility to childhood leukemia in Argentina by conducting an exploratory case-control study and correlated patients' genotype to clinical and biological features.

Methods: Deletion polymorphisms in *GSTM1* and *GSTT1* genes and the single nucleotide polymorphism in *GSTP1* c.313A>G (rs1695; p.105Ile>Val) were genotyped by PCR-RFLP in 36 patients and 133 healthy individuals.

Results: *GSTM1*-null genotype was associated with a lower risk of developing acute leukemia ($P = 0.013$; OR: 0.31; CI: 0.12–0.80), while *GSTP1-GG* variants displayed an increased risk ($P = 0.01$; OR: 3.9; CI: 1.85–8.2). However, no differences were found for *GSTT1* gene.

Conclusion: These preliminary results, to be validated in a larger population from Argentina, suggest that the development of pediatric leukemia may be differentially influenced by polymorphic variants in GST genes.

Keywords: Glutathione S-transferases, Childhood acute leukemia, Genetic polymorphisms, Leukemia risk

Introduction

Acute leukemia, the most common cancer in children, is a heterogeneous disease characterized by multiple chromosome and molecular alterations, which are specifically associated with diagnosis and prognosis of both childhood acute lymphoblastic leukemia (ALL) and acute myeloid leukemia (AML). Within all childhood leukemia diagnoses, approximately 80% are ALL, 17% AML, and 3% chronic myeloid leukemias, with some variation in ALL and AML incidence rates worldwide.[1,2] In Argentina, 415 new cases of pediatric ALL and 104 of AML are recorded per year according to national childhood cancer registry.[3] Although treatment outcome of childhood leukemias has improved toward 5-year event-free survival rates of 76–86% for ALL and 49–63% for AML, the mechanisms involved in the etiopathogenesis remain far elusive.[1] Moreover little is known about the factors that influence an individual's susceptibility to *de novo* leukemia.[4] A combination of factors appears to be necessary for leukemogenesis; all involving gene–environment interactions.[5] Recently, the possible role of inherited polymorphisms in the pathogenesis and heterogeneity of the disease has been proposed.[6] Moreover, individual predisposing factors, i.e. genetic polymorphisms in xenobiotic-metabolizing genes have been identified as risk factors for acute leukemia.[7,8]

Glutathione S-transferases (GSTs) are phase II detoxification enzymes involved in cellular protection against xenobiotics, carcinogens, and oxidative stress.[9] As non-enzymatic proteins, GSTs can modulate signaling pathways that control cell proliferation, cell differentiation, cell death, and DNA damage processing.[10] Functional polymorphisms due to complete deletion of *GSTM1* and *GSTT1* genes and a single

DOI 10.1179/1607845415Y.0000000007

nucleotide substitution in *GSTP1* c.313A>G (rs1695; p.105Ile>Val) are common in the general population, and genotype distribution exhibits striking interethnic differences.[11,12] Full or partial loss of GST activity may modify the detoxification capacity, which in turn could cause genome damage leading ultimately to leukemogenesis.[13] Previous studies that evaluated the association between GST genotypes and leukemia risk worldwide produced somewhat contradictory results.[14] To our knowledge, data are still lacking in Argentina. To assess whether GST variants are associated with susceptibility to childhood leukemias in our country, we conducted an exploratory case-control study on 36 patients and 133 healthy controls, both groups of Argentinian origin. GST variants were then correlated to patients' clinical and biological characteristics.

Materials and methods

Population studied

Peripheral blood samples were obtained from 36 pediatric patients (18 females and 18 males; mean age 8.0 years; range 2–16 years) diagnosed with leukemia at the *Hospital General de Niños, 'Pedro Elizalde'*, Buenos Aires, Argentina. Patients were classified according to the criteria of the FAB group in ALL (26) and AML (10). Three patients have relapsed and continue in maintenance and two have died due to disease progression. The clinical–pathological characteristics of patients are shown in Table 1. In addition, two groups of unrelated healthy individuals without medical history of leukemia or cancer were analyzed. The first cohort included 33 pediatric controls with comparable age (mean 10.86 ± 1.09, range 1–18 years) and gender distribution (15 females and 18 males) as patients. A random sample of 100 adult controls (67 males and 33 females; mean age 39.52 ± 1.30, 25–70 years) was further screened as a validation set for genotype analysis. Patients and controls were Argentinians from Buenos Aires city and surrounding urban area, a central region of Argentina, and had the same ethnicity. All individuals provided their informed consent according to institutional guidelines. The study was approved by the Institutional Ethical Committee and complies with the International Declaration of Helsinki.

Table 1 Clinical–pathological characteristics of childhood acute leukemia patients

Clinical characteristics	Pediatric acute leukemia
No. of cases	36
Sex (F/M)	18/18
Mean age (range), years	8.0 (2–16)
Leukemia type	
ALL	26
AML	10
Cytogenetics	
Normal	10
Abnormal	16
Not available	8
	Median (range)
Blasts (%)	87 (35–100)
WBC 10^9/l	13.7 (1.7–79)
Hemoglobin (g/l)	7.3 (3.10–13)
Platelets 10^9/l	28 (6–96)

GSTs genotyping

Genomic DNA was isolated using standard proteinase K/phenol/chloroform or salting out methods. A multiplex PCR assay using previously published primer pairs was used to amplify *GSTT1* (480 bp) and *GSTM1* (273 bp) genes with *β-globin* (680 bp) as an internal positive control.[15] PCR mix, primer concentrations, cycling conditions, and electrophoresis were used as defined earlier.[16] *GSTP1* c.313A>G genotypes were identified by RFLP-PCR at 55°C using previously reported primer pairs (0.4 μM).[17] PCR products were digested overnight with *Alw*261 restriction enzyme and analyzed by electrophoresis on 4% 3:1 NuSieve/agarose gels.[18] All genotypes were independently scored by two reviewers, and 10% of the samples were randomly reanalyzed, yielding identical results.

Statistical analysis

SPSS statistical package (version 15.0) was used for data analysis (IBM, SPSS Inc., Chicago, USA). Differences in genotype distribution between patients and controls and Hardy–Weinberg equilibrium were determined by Chi^2 or Fisher exact tests. The influence of each polymorphism on acute leukemia risk was evaluated by applying univariate analysis, unconditional logistic regression method, Mantel–Haenszel test, and estimating the odds ratio (OR) with the 95% confidence interval (CI). Kolmogorov Smirnov test was used to evaluate normal distribution and homogeneity was determined by Breslow–Day test. Patients with particular GST genotypes were tested for various clinico-pathological parameters using Student's *t*-test for continuous variables and the Chi^2 test for discrete variables and those without normal distribution. Values of $P < 0.05$ were considered statistically significant.

Results

To determine the genetic profile of GST genes in Argentina, *GSTM1*, *GSTT1*, and *GSTP1* c.313A>G polymorphisms were first examined in pediatric and adult healthy controls. No deviation from Hardy–Weinberg equilibrium was demonstrated for *GSTP1* gene. The distribution of genotype frequencies in controls showed no significant differences when stratified by gender or comparing pediatric vs. adult individuals (all $P > 0.07$) (Fig. 1). Therefore, GST

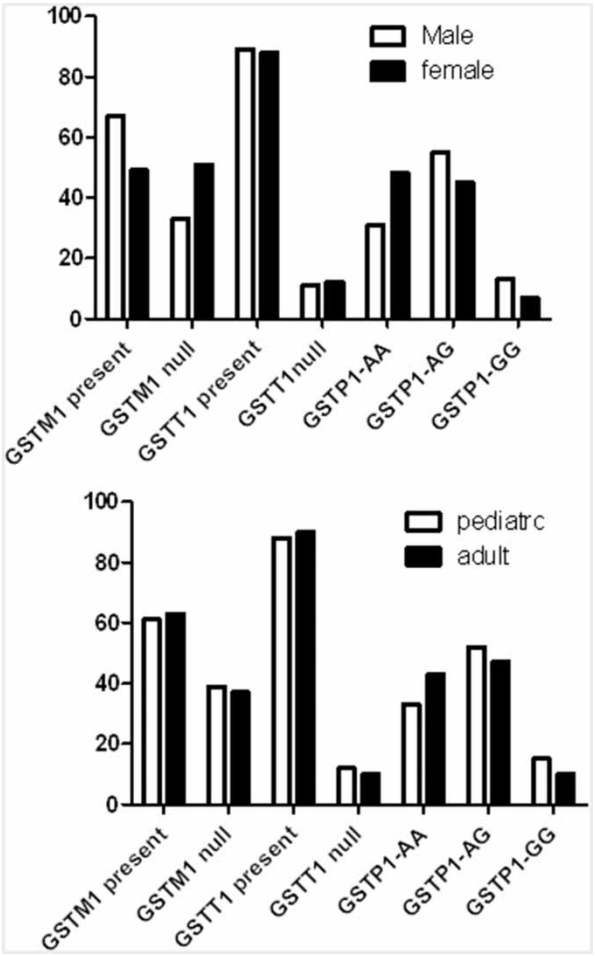

Figure 1 Distribution of GST genotype frequencies in 133 healthy Argentinians stratified by gender and age. (A) The distribution of *GSTM1*, *GSTT1*, and *GSTP1* genotypes according to gender. (B) Stratification of *GSTM1*, *GSTT1*, and *GSTP1* genotypes according to age grouped as pediatric (1–18 years) and adults (25–70 years). The analysis comparing the percentage of cases with different genotypes among gender or age groups showed no significant difference.

frequencies from both control cohorts were combined and referred to as the control group for comparative analysis with patients.

Allelic and genotype frequencies from patients and controls are shown in Table 2. *GSTM1*-null genotype was significantly underrepresented in patients (16.7%) compared to controls (37.6%) and was associated with a lower risk of acute leukemia ($P = 0.013$; OR: 0.31; CI: 0.12–0.80). No significant differences were observed for *GSTT1* genotypes. These associations persisted similar after adjustment in multivariate analysis (Table 2). For the overall analysis, significant association between the risk of leukemia and the variant genotypes of *GSTP1* c.313A>G polymorphism was found in homozygote comparison (AA vs. GG: $P = 0.001$; OR: 6.17; CI: 2.06–18.43). No significant association was found in heterozygote comparison (AA vs. AG). Moreover, comparison between *GSTP1* homozygous variant (GG) vs. heterozygous

Table 2 Analysis of GST genotypes in Argentinian patients and controls

Genes	Genotype/alleles	Controls $n = 133$(%)	Patients $n = 36$(%)	Univariate analysis			Multivariate analysis		
				OR	CI (95%)	*P*	OR*	CI (95%)	*P*
GSTM1	Present	75 (62.4)	30 (83.3)	1			1		
	Null	48 (37.6)	6 (16.7)	0.31	0.12–0.80	0.013	0.29	0.11–0.74	0.01
GSTT1	Present	109 (89.5)	33 (91.6)	1			1		
	Null	14 (10.5)	3 (8.4)	0.71	0.20–2.6	NS	0.72	0.20–2.7	NS
GSTP1	AA	53 (39.8)	7 (19.4)	1			1		
	AG	65 (48.9)	17 (47.2)	1.93	0.74–5.03	NS	1.76	0.69–4.51	NS
	GG[a]	15 (11.3)	12 (33.3)	6.17	2.06–18.43	0.001	1.67	0.57–4.94	NS
	GG[b]			3.01	1.2–7.6	0.02	3.91	1.43–10.7	0.008
	AA+AG vs. GG	118 vs. 15	24 vs. 12	3.9	1.85–8.2	0.01	4.52	1.8–11.36	0.001
	A	171 (64)	31 (43)	1			1		
	G	95 (36)	41 (57)	2.4	1.4–4.2	0.001	1.34	0.83–2.15	NS

*Adjusted OR: adjusted in multivariate logistic regression models.
[a]Compared to wild type genotype.
[b]Compared to heterozygous genotypes; NS = not significant.

(AG) genotypes was also statistically significant ($P = 0.02$; OR: 3.01; CI: 1.2–7.6) (Table 2). As no significant differences were observed between individuals carrying homozygous wild type (AA) or heterozygous (AG) genotypes, we considered a recessive model (AA+AG vs. GG: $P = 0.01$; OR: 3.9; CI: 1.85–8.2), showing that homozygosity for G allele is a significant risk factor for childhood leukemia. This association remained statistically significant after adjustment for age and sex in multivariate analysis ($P = 0.001$; OR: 4.52; CI: 1.8–11.36).

Taking into account that different GSTs are involved in the detoxification of similar activated carcinogens, we investigated the joined effect of GST genotypes on acute leukemia risk. No cumulative effect of more than one variant genotype was observed, but it should be noted that the number of children with two or three variant alleles was small. Regarding the clinico-pathological characteristics of these patients, there was significant correlation between white blood count and *GSTP1*-GG genotype ($P = 0.028$). No correlation was found for leukemic type, age, sex, cytogenetics, platelets, blasts, and hemoglobin.

Discussion

Genetic variability of GST genes was assessed in leukemic and healthy Argentinians because ethnicity has been linked to disparities in the incidence of acute leukemia,[2] and remarkable differences in the pattern of GST frequencies were described worldwide.[11,12] In this study, we found that *GSTM1*-null genotype was associated with a reduced risk of leukemia; *GSTP1*-GG variant genotype was found to increase risk, while *GSTT1* did not alter the risk. These data indicate that GST polymorphic variants may have a different involvement in the susceptibility to acute leukemia in Argentina children.

GSTs are a well-known family of multifunctional enzymes involved in the detoxification of a wide variety of carcinogen. Previous studies demonstrated striking ethnic differences in the distribution of GST genotype frequencies, indicating that 30–50% of the Caucasian population presents the *GSTM1*-null genotype, 10–20% has *GSTT1*-null, and 7–12% carries the *GSTP1*-GG variant.[11,12,19] Our data demonstrate that GST frequencies determined in the general Argentinian population resembles Caucasian findings, which is consistent with the large European immigrant flow arriving mainly from Spain and Italy. Although the population of Argentina is the result of a long-standing process of admixture between several ethnic groups, the genetic structure within Buenos Aires city and surrounding urban areas is predominantly composed by European ancestry.[20]

GST polymorphisms have been related to various cancers because some of these variants are directly associated with an inefficient carcinogen detoxification and an increased rate of DNA mutation, genomic instability, and cancer.[21] Studies of GST genotypes in relation to the susceptibility to acute leukemia have shown conflicting results.[14] Several groups showed that *GSTM1*-null genotype is related to increased risk of acute leukemia in various populations.[13,22–30] However, we found that *GSTM1*-null variant displays a reduced risk for leukemia in Argentinian children, suggesting that the presence of GSTM1 protein has in fact a role in leukemogenesis. This finding is in agreement with those reported by Barnette *et al.*,[22] who demonstrated that the presence of one or two alleles of *GSTM1* gene implies a higher risk for acute leukemia. The role of an efficient GSTM1 enzyme activity on leukemia development is difficult to understand. It has been suggested that the effectiveness of GSTs activity is dependent on the supply of glutathione (GSH).[31] Therefore, despite an efficient enzyme activity, when the levels of GSH decrease, the conjugation with phase II detoxifying enzymes may produce active metabolites and increased toxicity.[32] Results obtained from our study support that the presence of GSTM1 protein may be involved in leukemia etiology, probably coupled with alterations in other key cellular mechanisms.

Many epidemiological studies have tested associations between *GSTT1* polymorphisms and acute leukemia risk, but conflicting results have been achieved. Our study has not elucidated any particular association for this gene, as was seen in previous replicated studies.[13,24–26,28,33] While significant involvement of *GSTT1* deletion was reported,[4,29,34–36] other group showed a significantly lower frequency of *GSTT1*-null genotype among AML patients compared to controls, suggesting that may play protective roles in leukemia.[37] Moreover, an increased risk of ALL for patients carrying nonnull alleles of *GSTT1* was also reported.[22] These inconclusive findings warrant further studies to confirm the role of *GSTT1* polymorphisms in the susceptibility to acute leukemia in the Argentinian population.

The *GSTP1* c.313A>G polymorphism was found to fit a recessive model, in which two copies of the allele G is required for an increased leukemia risk, complying with the tumor suppressive role of this gene. GSTP1 has been shown to function not only as a phase II metabolizing enzyme, but it is also involved in the regulation of cell cycle and apoptosis.[38] Variation in the expression and activity of GSTP1 has been associated with a variety of human cancers and has been linked to functional genetic polymorphisms. The SNP c.313A>G affects the substrate binding site and results in reduced catalytic activity and detoxification capacity of the enzyme,[17] leading to the accumulation of genotoxic compounds which may be

involved in the carcinogenic process.[35] The results obtained in our study indicating that *GSTP1*-GG genotype is a risk factor for childhood leukemia are in agreement with earlier publications,[29,33] but, again, opposite results were found in other epidemiological studies.[27,34,39,40]

Initiation of leukemogenesis is likely caused by multiple factors, nevertheless, the exact mechanisms underlying remains poorly understood.[41] Multiple pathways and genetic lesions and all of them are necessary to develop a fully established leukemia. Accumulating studies suggest that inherited genetic factors affect the risk of developing leukemia. Hereditary differences in the expression and activity of human GSTs have been reported and altered GST enzymatic activity was associated with leukemogenesis. Polymorphisms in the *GSTM1*, *GSTT1*, and *GSTP1* loci have been investigated in several case-control studies from different regions in the world, but the association to acute leukemia risk was largely inconsistent. A potential explanation for the discrepancy between studies was thought to be related to differences in ethnicity and age of patients, treatment, and follow-up periods.[14]

We recognize that the limitation of our work is related to the small number of patients evaluated. This study, however, still contributes to the preliminary understanding of the involvement of GSTs in the development of childhood acute leukemia in our population. To the best our knowledge; we have determined for the first time the genetic profile of GST polymorphisms in Argentinian children. In addition, we observed that susceptibility to the disease could be related to an active GSTM1 enzyme and a lower GSTP1 protein activity. These preliminary results, to be validated in a larger population from Argentina, suggest that the development of childhood leukemia in our country may be differentially influenced by genotypic variants in GST genes. The extent to which phase II enzymes are coordinately regulated in networks that favor leukemic development should be deeply explored.

Disclaimer statements

Contributors NW: designed and performed research, analyzed data, and wrote the paper; MN: analyzed data, made the statistical analysis and final approval of the manuscript; GG and GE: provided patients samples, evaluated clinical data and final approval of the manuscript; SA and IL: designed research discussed results and final approval of the manuscript; AF: designed and performed research, analyzed data, and wrote the paper.

Funding This study was supported in part by grants from the National Research Council (CONICET-PIP0946), the National Agency of Scientific and Technical Promotion (SECyT-PICT 0330), and the Academia Nacional de Medicina from Buenos Aires, Argentina.

Conflicts of interest None.

Ethics approval The study was approved by the Institutional Ethical Committee and complies with the International Declaration of Helsinki.

References

1 Pui CH, Carroll WL, Meshinchi S, Arceci RJ. Biology, risk stratification, and therapy of pediatric acute leukemias: an update. J Clin Oncol. 2011;29:551–65.
2 Metayer C, Milne E, Clavel J, Infante-Rivard C, Petridou E, Taylor M, *et al.* The Childhood Leukemia International Consortium. Cancer Epidemiol. 2013;37:336–47.
3 Moreno F, Loria D, Abriata G, Terracini B, ROHA network. Childhood cancer: incidence and early deaths in Argentina, 2000–2008. Eur J Cancer. 2013;49:465–73.
4 Bolufer P1, Collado M, Barragán E, Cervera J, Calasanz MJ, Colomer D, *et al.* The potential effect of gender in combination with common genetic polymorphisms of drug-metabolizing enzymes on the risk of developing acute leukemia. Haematologica. 2007;92:308–14.
5 Eden T. Aetiology of childhood leukaemia. Cancer Treat Rev. 2010;36:286–97.
6 Stavropoulou V, Brault L, Schwaller J. Insights into molecular pathways for targeted therapeutics in acute leukaemia. Swiss Med Wkly 2010;140:w13068.
7 Vijayakrishnan J, Houlston RS. Candidate gene association studies and risk of childhood acute lymphoblastic leukemia: a systematic review and meta-analysis. Haematologica. 2010;95:1405–14.
8 Puumala SE, Ross JA, Aplenc R, Spector LG. Epidemiology of childhood acute myeloid leukemia. Pediatr Blood Cancer 2013;60:728–33.
9 Hayes JD, Flanagan JU, Jowsey IR. Glutathione transferases. Annu Rev Pharmacol Toxicol. 2005;45:51–88.
10 Dusinska M, Staruchova M, Horska A, Smolkova B, Collins A, Bonassi S, *et al.* Are glutathione S transferases involved in DNA damage signalling? Interactions with DNA damage and repair revealed from molecular epidemiology studies. Mutat Res. 2012;736:130–7.
11 Garte S, Gaspari L, Alexandrie AK, Ambrosone C, Autrup H, Autrup JL, *et al.* Metabolic gene polymorphism frequencies in control populations. Cancer Epidemiol Biomarkers Prev. 2001;10:1239–45.
12 Piacentini S, Polimanti R, Porreca F, Martinez-Labarga C, De Stefano, Fuciarelli M. GSTT1 and GSM1 gene polymorphisms in European and African populations. Mol Biol Rep. 2011;38:1225–30.
13 Pakakasama S, Mukda E, Sasanakul W, Kadegasem P, Udomsubpayakul U, Thithapandha A, *et al.* Polymorphisms of drug-metabolizing enzymes and risk of childhood acute lymphoblastic leukemia. Am J Hematol. 2005;79:202–5.
14 Luo W, Kinsey M, Schiffman JD, Lessnick SL. Glutathione s-transferases in pediatric cancer. Front Oncol. 2011;24:1–39.
15 Morari EC, Leite JL, Granja F, da Assumpção LV, Ward LS. The null genotype of glutathione s-transferase M1 and T1 locus increases the risk for thyroid cancer. Cancer Epidemiol Biomarkers Prev. 2002;11:1485–8.
16 Fundia AF, Weich N, Crivelli A, La Motta G, Larripa IB, Slavutsky I. Glutathione S-transferase gene polymorphisms in celiac disease and their correlation with genomic instability phenotype. Clin Res Hepatol Gastroenterol. 2014;38:379–84.
17 Harries LW, Stubbins MJ, Forman D, Howard GC, Wolf CR. Identification of genetic polymorphisms at the glutathione S-transferase Pi locus and association with susceptibility to bladder, testicular and prostate cancer. Carcinogenesis 1997;18:641–4.
18 Stella F, Weich N, Panero J, Fantl DB, Schutz N, Fundia AF, *et al.* Glutathione S-transferase P1 mRNA expression in plasma cell disorders and its correlation with polymorphic variants and clinical outcome. Cancer Epidemiol. 2013;37:671–4.

19 Avena S, Via M, Ziv E, Pérez-Stable EJ, Gignoux CR, Dejean C, *et al.* Heterogeneity in genetic admixture across different regions of Argentina. PLoS One 2012;7(4):e34695.
20 HapMap. Available from http://hapmap.ncbi.nlm.nih.gov/thehapmap.html.
21 Nørskov MS, Frikke-Schmidt R, Bojesen SE, Nordestgaard BG, Loft S, Tybjærg-Hansen A. Copy number variation in glutathione-S-transferase T1 and M1 predicts incidence and 5-year survival from prostate and bladder cancer, and incidence of corpus uteri cancer in the general population. Pharmacogenomics J. 2011;11:292–9.
22 Barnette P, Scholl R, Blandford M, Ballard L, Tsodikov A, Magee J, *et al.* Detection of glutathione S-transferase polymorphic alleles in pediatric cancer population. Cancer Epidemiol Biomarkers Prev. 2004;13:304–13.
23 Krajinovic M, Labuda D, Richer C, Karimi S, Sinnett D. Susceptibility to childhood acute lymphoblastic leukemia; influence of CYP1A1, CYP2D6, GSTTM1 and GSTT1 genetic polymorphisms. Blood 1999;93:1496–501.
24 Davies SM, Robison LL, Buckley JD, Radloff GA, Ross JA, Perentesis JP. Glutathione S-transferase polymorphisms in children with myeloid leukemia: a children's cancer group study. Cancer Epidemiol Biomarkers Prev. 2000;9:563–6.
25 Alves S, Amorim A, Ferreira F, Norton L, Prata MJ. The GSTM1 and GSTT1 genetic polymorphisms and susceptibility to acute lymphoblastic leukemia in children from north Portugal. Leukemia 2002;16:1565–7.
26 Aydin-Sayitoglu M1, Hatirnaz O, Erensoy N, Ozbek U. Role of CYP2D6, CYP1A1, CYP2E1, GSTT1, and GSTM1 genes in the susceptibility to acute leukemias. Am J Hematol. 2006;81:162–70.
27 Sunnetha KJ, Nancy KN, Rajalekshmy KR, Sagar TG, Rajkumar T. Role of GSTM1 (present/null) and GSTP1 (Ile105Val) polymorphisms in susceptibility to acute lymphoblastic leukemia among the South Indian population. Asian Pac J Cancer Prev. 2008;9:733–6.
28 Das P, Shaik AP, Bammidi VK. Meta-analysis study of glutathione-S-transferases (GSTM1, GSTP1, and GSTT1) gene polymorphisms and risk of acute myeloid leukemia. Leuk Lymphoma 2009;50:1345–51.
29 Dunna NR, Vure S, Sailaja K, Surekha D, Raghunadharao D, Rajappa S, *et al.* Deletion of GSTM1 and T1 genes as a risk factor for development of acute leukemia. Asian Pac J Cancer Prev. 2013;14:2221–4.
30 Joseph T, Kusumakumary P, Chacko P, Abraham A, Pillai MR. Genetic polymorphism of CYP1A1, CYP2D6, GSTM1 and GSTT1 and susceptibility to acute lymphoblastic leukaemia in Indian children. Pediatr Blood Cancer. 2004;43:560–7.
31 Meijerman I, Beijnen JH, Schellens JH. Combined action and regulation of phase II enzymes and multidrug resistance proteins in multidrug resistance in cancer. Cancer Treat Rev. 2008;34:505–20.
32 Leone G, Pagano L, Yehuda DB, Voso MT. Therapy-related leukemia and myelodysplasia: susceptibility and incidence. Haematologica. 2007;92:1389–98.
33 Krajinovic M, Labuda D, Sinnett D. Glutathione S-transferase P1 genetic polymorphisms and susceptibility to childhood acute lymphoblastic leukaemia. Pharmacogenetics 2002;12:655–8.
34 Rollinson S, Roddam P, Kane E, Roman E, Cartwright R, Jack A, *et al.* Polymorphic variation with the glutathione S-transferase genes and risk of adult acute leukemia. Carcinogenesis 2000;21:43–7.
35 D'alo F, Voso MT, Guidi F, Massini G, Scardocci A, Sica S, *et al.* Polymorphisms of CYP1A1 and glutathione S-transferase and susceptibility to adult acute myeloid leukemia. Haematologica 2004;89:664–70.
36 Kim HN, Kim NY, Yu L, Tran HT, Kim YK, Lee IK, *et al.* Association of GSTT1 polymorphism with acute myeloid leukemia risk is dependent on smoking status. Leuk Lymphoma 2012;53:681–7.
37 Balta G, Yuksek N, Ozyurek E, Ertem U, Hicsonmez G, Altay C, *et al.* Characterization of MTHFR, GSTM1, GSTT1, GSTP1, and CYP1A1 genotypes in childhood acute leukemia. Am J Hematol. 2003;73:154–60.
38 Moyer AM, Salavaggione OE, Wu TY, Moon I, Eckloff BW, Hildebrandt MA, *et al.* Glutathione s-transferase p1: gene sequence variation and functional genomic studies. Cancer Res. 2008;68:4791–801.
39 Voso MT, Fabiani E, D'Alo' F, Guidi F, Di Ruscio A, Sica S, *et al.* Increased risk of acute myeloid leukaemia due to polymorphisms in detoxification and DNA repair enzymes. Ann Oncol. 2007;18:1523–8.
40 Gatedee J, Pakakassama S, Muangman S, Pongstaporn W. Glutathione S-transferase P1 genotypes, genetic susceptibility and outcome of therapy in Thai childhood acute lymphoblastic leukemia. Asian Pac J Cancer Prev. 2007;8:294–6.
41 Dai YE, Tang L, Healy J, Sinnett D. Contribution of polymorphisms in IKZF1 gene to childhood acute leukemia: a meta-analysis of 33 case-control studies. PLoS One. 2014;259(11):e11374.