Genetic polymorphism of glutathione S-transferase T1 and the risk of rheumatoid arthritis: a meta-analysis

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Abstract

Objective

Reports investigating the association between the genetic polymorphism of glutathione S-transferase T1 (GSTT1) and the risk of rheumatoid arthritis (RA) have revealed conflicting results. To clarify the effect of GSTT1 polymorphism on the risk of developing RA, we carried out a meta-analysis using published data.

Methods

Electronic searches were conducted to select studies. Reports were included if they were observational studies investigating the link between GSTT1 genotype and the risk of RA. The principal outcome measure was the odds ratio (OR) with 95% confidence interval (CI) for the risk of RA with GSTT1 null genotype.

Results

We identified 7 eligible studies including 2652 cases and 4117 controls. The combined results showed that there was not a statistically significant link between GSTT1 null genotype and RA. However, we observed an increased risk in heavy smokers (cigarette consumption >10 pack-years) with GSTT1 null polymorphism compared with never or light smokers (cigarette consumption ≤10 pack-years) with GSTT1 present. Moreover, compared to GSTT1 positive polymorphism with seronegative results, there was an increased risk in GSTT1 null polymorphism with seropositive results.

Conclusion

The results from this meta-analysis suggested that GSTT1 null genotype is not association with an increased susceptibility to RA. However, GSTT1 null polymorphism may increase the risk of RA in relation to heavy smokers or seropositive results. Whether GSTT1 polymorphism may act in synergy with other genes or environmental factors remains to be studied more in depth.

Key words

glutathione S-transferase, GSTT1, polymorphism, rheumatoid arthritis, meta-analysis

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Introduction

Rheumatoid arthritis (RA) is a chronic multi-articular autoimmune disease that affects up to 1% of the general adult population worldwide (1). Although RA is a common autoimmune disease of unclear etiology, it is evidently shown that the expression and development of the disease is due to combination of genetic and environmental factors (2-7). Moreover, the onset of RA is likely to involve multiple genes, including HLA-DRB1, TRAF1/C5, STAT4, REL, PTPN22, TNF-α, IL2/IL21, CTLA4, TNFRII, VDR, etc., which have been identified with the inception and progression of RA by candidate genes and genome studies (8-11). The recent genome-wide association study (GWAS) suggested that 7 new RA risk alleles were identified (12).

Among various candidate genes associated in RA, the glutathione S-transferase (GST) genes are a widely expressed supergene family encoding biotransforming dimeric enzymes that catalyse the conjugation of glutathione and are implicated in the detoxification of free radicals and prostaglandins (13-17). They are divided into two microsomal and various GST-classes, including alpha, mu, kappa, pi, sigma, theta and zeta. GST are involved in the metabolism of many xenobiotics that include an array of environmental carcinogens, polycyclic aromatic hydrocarbons, and reactive oxygen species (ROS), which probably lead to cellular damage by oxidative stress (18). The theta class gene, GSTT1, is located on chromosome 22q 11.2 (19). It has a functional and non-functional allele. Homozygosity for the non-functional allele of GSTT1 (null genotype) causes an absence of enzymatic activity, and therefore may increase susceptibility to the harmful effects of carcinogen exposure and oxidative stress (18).

Several studies on the association between the GSTT1 polymorphism and RA have been published. Some studies have shown that the GSTT1 polymorphism to be associated with increased susceptibility to RA. However, other studies have failed to replicate it. With the aim to clarify the effect of GSTT1 polymorphism on the risk of RA, we carried out a meta-analysis using published data to obtain more accurate estimates of risk. This is, to our knowledge, the first meta-analysis conducted with respect to the association between the polymorphism in the glutathione S-transferase T1 and the risk of RA.

Materials and methods

Identification and relevant studies

We searched the following electronic databases: PubMed, Embase, CNKI database, Wanfang database, and Weipu database (last search was performed on October 1st, 2011). The following search terms were used: 'GSTT1 or glutathione S-transferase T1' and 'rheumatoid arthritis or RA' in combination with 'polymorphism or genetics or variant'. The languages were limited to English and Chinese.

Inclusion and exclusion criteria

The criteria for acceptance of the studies were as follows: (i) independent case-control studies for human; (ii) each study should have the similar research purpose with the identical study method; (iii) the main factors of these studies should be involved in GSTT1 polymorphism and risk of RA. Concerning the exclusion criteria, we stipulated the following: (i) studies without raw data we need; (ii) duplicated studies; (iii) abstracts and reviews.
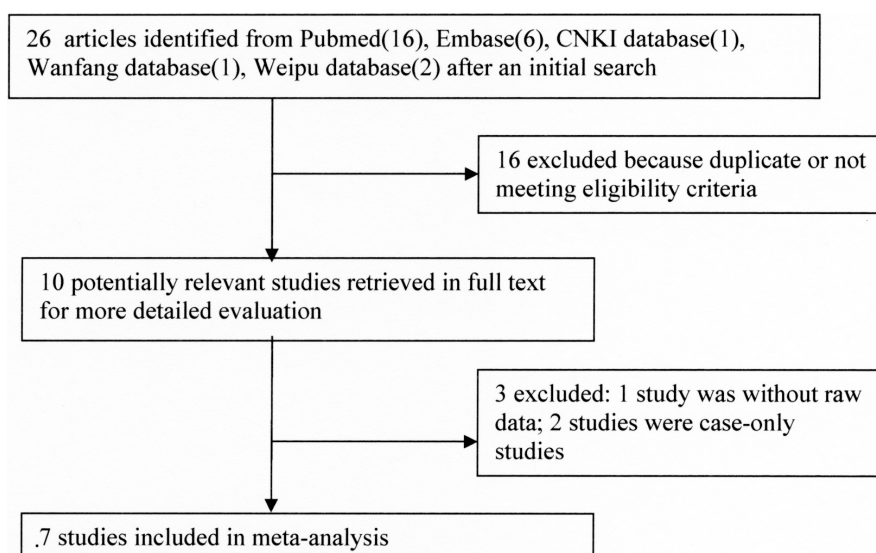
Data extraction

The extraction was made by two researchers independently according to the pre-specified selection criteria. Any disagreement was resolved by discussion and consultation with the third researcher. The information was retrieved from each study as follows: investigators; year of publication; country of study; ethnic origin; study design (categorised as hospital-based case-control studies; population-based case-control studies); the number of cases and controls with GSTT1 null and positive genotype; variables investigated in the studies (sex, smoking history, and serological results).

Statistical analysis

The statistical analysis was performed by RevMan 5.0 provided by Cochrane

26 articles identified from Pubmed(16), Embase(6), CNKI database(1), Wanfang database(1), Weipu database(2) after an initial search

16 excluded because duplicate or not meeting eligibility criteria

10 potentially relevant studies retrieved in full text for more detailed evaluation

3 excluded: 1 study was without raw data; 2 studies were case-only studies

7 studies included in meta-analysis

Fig. 1. Flow diagram of included/excluded studies.

Collaboration. $p<0.05$ was considered statistically significant. Meta-analysis was done with either the random-effect model or the fixed-effect model. Heterogeneity was checked by the χ^2-test. In addition, we used I^2 statistic to quantify the effect of heterogeneity, providing a measure of the degree of inconsistency in the results of the study. If the results of the trials had heterogeneity, we tried to find the sources of heterogeneity with sensitivity analysis. The random-effect model was only applied if we did not identify the source of heterogeneity, but both the random-effect model and the fixed-effect model were applied if the sources of heterogeneity could be found and excluded. On the other hand, we used both random- and fixed-effect model in the absence of heterogeneity. To establish the effect of clinical heterogeneity between studies on meta-analysis conclusions, subgroup analyses were conducted on the basis of sex, ethnic origin, smoking history, and serological results. The results were expressed with odds ratio (OR) for the categorical variables and 95% confidence interval (CI). Publication bias was analysed by using two methods. Visual inspection of asymmetry in funnel plots was carried out. The fail-safe number was also used to statistically assess publication bias.

Results

Study characteristics

Figure 1 shows the selection process of our study. A total of 26 papers were retrieved through initially searching the selected databases. We identified ten relevant studies that described the association between the GSTT1 polymorphism and rheumatoid arthritis, however, after reading the full articles, we excluded two case-only studies (16, 20), and one paper without raw data (21). Hence, only 7 reports met the inclusion crite-ria and were included. All of the studies were based on genotypic methods: polymerase chain reaction (PCR).

Among the eligible studies, 4 studies were involved in Caucasians (23-26), two studies were about Asians (15, 22) and another one contained mixed racial groups, including South Asians and Caucasians (27). 2 case-control studies (23, 26) were population-based studies (PCC), and 5 (15, 22, 24, 25, 27) were hospital-based case-control studies (HCC). History of smoking was verified for cases and controls, with 2 reporting results of the interaction between GSTT1 polymorphism and RA risk in relation to cigarette smoking habits. Sex, serological results were also extracted from the studies respectively. The characteristics of the studies included in this meta-analysis are presented in Table I.

GSTT1 null genotype and RA in total studies

In total, the eligible studies were involved in 2652 cases and 4117 controls. The combined results based on all studies suggested that there was not a statistically significant link between GSTT1 null genotype and RA (OR=1.33, 95% CI:0.86-2.06, χ^2=63.26, d.f.=6, p for heterogeneity <0.00001, I^2=91%) (Fig. 2). Because heterogeneity still existed when any single study was omitted, we used a random-effect model.

GSTT1 null genotype and RA in subgroups
– Ethnic groups

We observed that there was not a significant association between GSTT1 null genotype and RA in Caucasians (Fixed-effect model: OR=1.09, 95%

Table I. Summary of case-control studies of GSTT1 polymorphism and rheumatoid arthritis risk.

Investigators	Year	Study Design	Country	Ethnic Origin	GSTT1 null		GSTT1 present	
					Cases	Controls	Cases	Controls
Yun *et al.*	2005	HCC[§]	Korea	Asians	124	214	134	186
Morinobu *et al.*	2006	HCC	Japan	Asians	45	62	63	81
Padyukov *et al.*	2004	PCC[¥]	Sweden	Caucasians	252	150	878	1481
Mattey *et al.*	1999	HCC	UK	Caucasians	223	451	52	105
Layton *et al.*	1999	HCC	UK	Caucasians	19	105	62	451
Keenan *et al.*	2010	PCC	USA	Caucasians	100	92	434	445
Ghelani *et al.*	2011	HCC	UK	Mixed*	71	40	195	254

[§]HCC: hospital-based case-control; [¥]PCC: population-based case-control; *South Asians and Caucasians.

CI:0.94–1.27, χ^2=5.39, d.f.=4, p for heterogeneity = 0.25, I²=26%; Random-effect model: OR=1.12, 95% CI:0.93–1.35, χ^2=5.39, d.f.=4, p for heterogeneity = 0.25, I²=26%). In Asians group, similarly, there was no statistically significant link between GSTT1 null genotype and RA (OR=1.22, 95% CI:0.63–2.37, χ^2=11.56, d.f.=2, p for heterogeneity = 0.003, I²=83%). Although the result revealed that one study (27) was the main source of the heterogeneity after sensitivity analysis and so we excluded it, we could not find the association between GSTT1 null genotype and RA in the Asian group (Fixed-effect model: OR=0.84, 95% CI:0.64-1.09, χ^2=0.24, d.f.=1, p for heterogeneity = 0.62, I²=0%; Random-effect model: OR=0.84, 95% CI:0.64 – 1.09, χ^2=0.24, d.f.=1, p for heterogeneity = 0.62, I²=0%).

– *The source of controls*

The overall ORs for hospital-based case-control studies were 1.17 (95% CI:0.80–1.71, χ^2=16.37, d.f.=4, p for heterogeneity = 0.003, I²=76%). After sensitivity analysis, the result indicated that one study (27) was the main source of the heterogeneity. Therefore, by excluding the study, the overall OR of rheumatoid arthritis risk associated with the GSTT1 null genotype in hospital-based case-control studies was 0.93 in a fixed-effect model (95% CI:0.76–1.14, χ^2=2.46, d.f.=3, p for heterogeneity = 0.48, I²=0%), and 0.94 in a random-effect model (OR=0.94, 95% CI:0.77–1.15, χ^2=2.46, d.f.=3, p for heterogeneity = 0.48, I²=0%). But in the population-based studies group, a test for heterogeneity showed that there was no heterogeneity (Fixed-effect model: OR=1.03, 95% CI:0.86–1.24, χ^2=0.33, d.f.=1, p for heterogeneity = 0.56, I²=0%; Random-effect model: OR=1.03, 95% CI:0.8 –1.24, χ^2=0.33, d.f.=1, p for heterogeneity = 0.56, I²=0%).

– *Smoking history*

There was no increase in risk for smokers or non-smokers with the GSTT1 null genotype (Smokers: Fixed-effect model: OR=1.05, 95% CI:0.84–1.33, χ^2=1.99, d.f.=1, p for heterogeneity = 0.16, I²=50%; Random-effect model: OR=1.09, 95% CI:0.77–1.54, χ^2=1.99, d.f.=1, p for heterogeneity = 0.16,
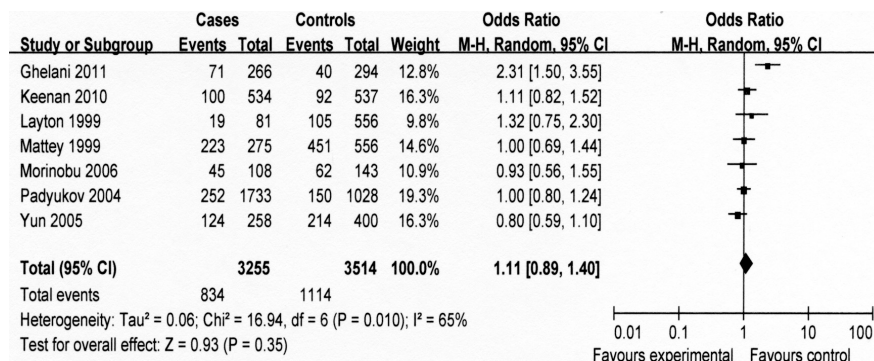

Fig. 2. Forest plot for the association between GSTT1 polymorphism and risk of rheumatoid arthritis.
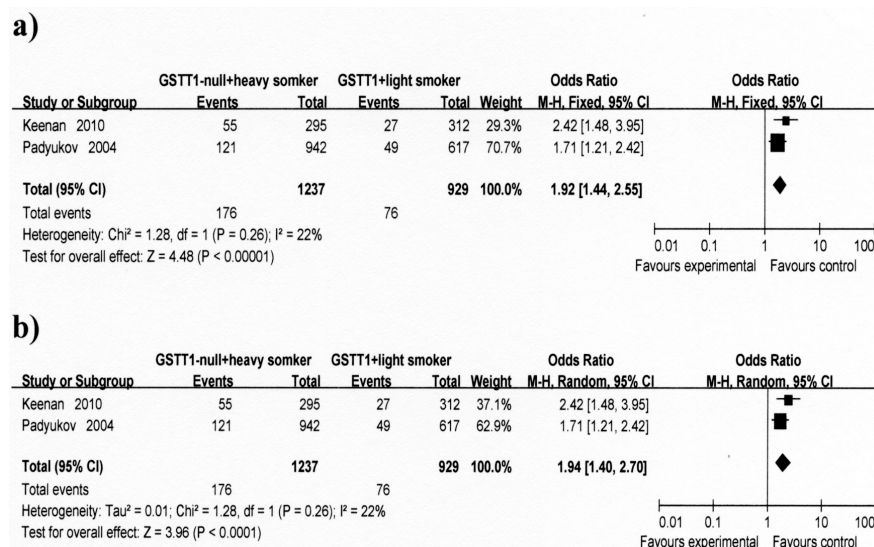

Fig. 3. Forest plot for the association between GSTT1 polymorphism and risk of rheumatoid arthritis in relation to smoking history with (**a**) a fixed-effect model and (**b**) a random-effect model.
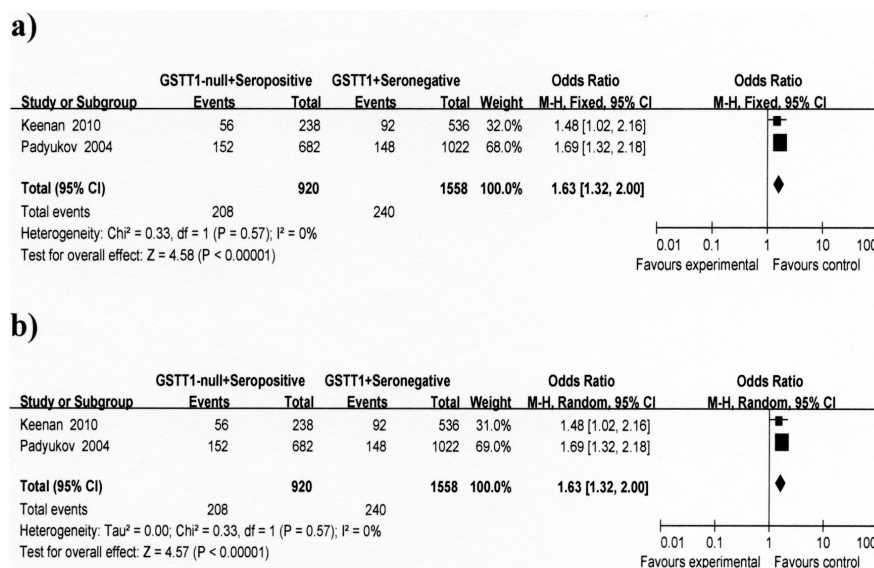

Fig. 4. Forest plot for the association between GSTT1 polymorphism and risk of rheumatoid arthritis in relation to serological results with (**a**) a fixed-effect model and (**b**) a random-effect model.

I^2=50%; Non-smokers: Fixed-effect model: OR=0.94, 95% CI:0.70–1.27, χ^2=1.05, d.f.=1, *p* for heterogeneity = 0.30, I^2=5%; Random-effect model: OR=0.94, 95% CI:0.69–1.29, χ^2=1.05, d.f.=1, *p* for heterogeneity = 0.30, I^2=5%). According to the analysis, we did not detect a statistically difference for smokers with GSTT1 null genotype compared to non-smokers with positive genotype (OR=1.38, 95% CI:0.76–2.52, χ^2=5.32, d.f.=1, *p* for heterogeneity = 0.02, I^2=81%). However, we observed an increased risk in heavy smokers (cigarette consumption >10 pack-years) with GSTT1 null polymorphism compared with never or light smokers (cigarette consumption ≤10 pack-years) with GSTT1 present (Fixed-effect model: OR=1.92, 95% CI:1.44–2.55, χ^2=1.28, d.f.=1, *p* for heterogeneity=0.26, I^2=22%; Random-effect model: OR=1.94, 95% CI:1.40–2.70, χ^2=1.28, d.f.=1, *p* for heterogeneity = 0.26, I^2=22%) (Fig. 3).

– *Serological results*
We observed that the overall ORs for seropositive studies were 1.33 (95% CI:0.83–2.12, χ^2=11.11, d.f.=2, *p* for heterogeneity = 0.004, I^2=82%). But the main source of heterogeneity led to one study (27) due to the sensitivity analysis. Hence, after excluding the study, there was also no statistically significant link between GSTT1 null genotype and RA (Fixed-effect model: OR=1.03, 95% CI:0.84–1.27, χ^2=0.07, d.f=1, *p* for heterogeneity = 0.79, I^2=0%; Random-effect model: OR=1.03, 95% CI:0.84–1.27, χ^2=0.07, d.f.=1, *p* for heterogeneity = 0.79, I^2=0%). Moreover, in seronegative groups, we did not find a statistically association between GSTT1 null genotype and the risk of RA (OR=0.44, 95% CI:0.07–2.62, χ^2=1.61, d.f.=1, *p* for heterogeneity <0.00001, I^2=97%). However, compared to RA of GSTT1 positive polymorphism with seronegative results, we found that there was an increased risk in GSTT1 null polymorphism with seropositive results, including rheumatoid factor positive or anti-citrullinated protein antibody positive (Fixed-effect model: OR=1.63, 95% CI:1.32–2.00, χ^2=0.33, d.f.=1, *p* for heterogeneity = 0.57, I^2=0%; Random-effect model: OR=1.63, 95% CI:1.32–

2.00, χ^2=0.33, d.f.=1, *p* for heterogeneity = 0.57, I^2=0%) (Fig. 4).

– *Sex*
No link could be detected between GSTT1 null genotype and RA in females (Fixed-effect model: OR=1.03, 95% CI:0.87–1.23, χ^2=0.33, d.f.=2, *p* for heterogeneity = 0.85, I^2=0%; Random-effect model: OR=1.03, 95% CI:0.87–1.23, χ^2=0.33, d.f.=2, *p* for heterogeneity = 0.85, I^2=0%) (Table II).
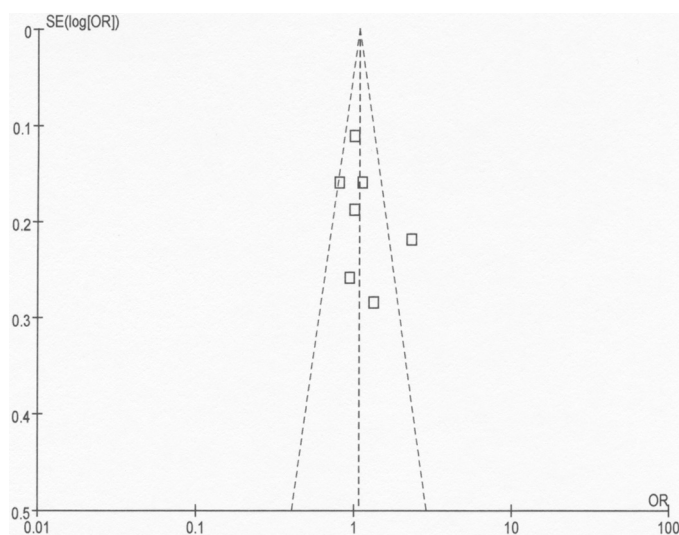
Publication bias
We obtained the evidence of publication bias as shown by the funnel plot (Fig. 5). In our study, according to Rosenthal's mathematical calculation, the fail-safe number (Nfs0.05) was 12 (28).

Discussion
In 1999, Mattey *et al.* (24) first examined the association between the GSTT1 polymorphism and the risk of RA. After that, the case-control studies provided controversial results. Based on cumulated evidence, we performed a meta-analysis on 7 studies with 2652

cases and 4117 controls. To our knowledge, this is the first meta-analysis conducted with respect to the association between the polymorphism in the glutathione S-transferase T1 and the risk of RA. The main observational and analytical results of our meta-analysis suggested that the GSTT1 null polymorphism is not to be a risk factor for susceptibility to RA, but can significantly increase the risk of RA in those who are heavy smokers or with seropositive results.

It is now widely accepted that differences in the distribution of various ethnicities between cases and controls may be a source of confounding when pooling studies (29). In the human population, the frequency of GSTT1 deficiency is 13–26% in Caucasians and 36–52% in Asians (30). However, when analysing the ethnic groups, our report suggested that the pooled OR associated with GSTT1 polymorphism was not significant in Caucasians or in Asians. However, both the Caucasian and the Asian reports in the subgroup analysis

Table II. Summary of meta-analysis of GSTT1 polymorphism and risk of rheumatoid arthritis.

Groups	subjects	OR (95% CI)	Statistical method	*p*
All studies	6769	1.11 (0.89-1.40)	Random*	0.35
Caucasians	5582	1.09 (0.94-1.27)	Fixed§	0.25
	5582	1.12 (0.93-1.35)	Random	0.23
Asians	1187	1.22 (0.63-2.37)	Random	0.56
	909	0.84 (0.64-1.09)	Fixed	0.19
	909	0.84 (0.64-1.09)	Random	0.19
Hospital-based case-control	2937	1.17 (0.80-1.71)	Random	0.43
	2377	0.93 (0.76-1.14)	Fixed	0.51
	2377	0.94 (0.77-1.15)	Random	0.53
Population-based case-control	3832	1.03 (0.86-1.24)	Fixed	0.72
	3832	1.03 (0.86-1.24)	Random	0.72
Females	3996	1.03 (0.87-1.23)	Fixed	0.71
	3996	1.03 (0.87-1.23)	Random	0.71
Smoking history				
Smokers	2352	1.05 (0.84-1.33)	Fixed	0.65
	2352	1.09 (0.77-1.54)	Random	0.62
Non-smokers	1389	0.94 (0.70-1.27)	Fixed	0.70
	1389	0.94 (0.69-1.29)	Random	0.70
Smokers *vs*. non-smokers	1555	1.38 (0.76-2.52)	Random	0.29
Heavy smokers *vs*. light smokers	2166	1.92 (1.44-2.55)	Fixed	<0.00001
	2166	1.94 (1.40-2.70)	Random	<0.0001
Serological results				
Seropositive	3463	1.33 (0.83-2.12)	Random	0.23
	2903	1.03 (0.84-1.27)	Fixed	0.76
	2903	1.03 (0.84-1.27)	Random	0.76
Seronegative	2326	0.44 (0.07-2.62)	Random	0.37
Seropositive *vs*. seronegative	2478	1.63 (1.32-2.00)	Fixed	<0.00001
	2478	1.63 (1.32-2.00)	Random	<0.00001

*Random-effect model; §Fixed-effect model.

included a mixture of populations from very distant countries, so the result must be interpreted with caution.

As it is conceivable that the GSTT1 gene leads to susceptibility to many cancers and non-cancer diseases, its genotype frequency may differ between the population-based and hospital-based controls. For example, it was suggested that there was a significant association between GSTT1 genotype and various cancers such as lung, oral, gastric, and colorectal cancers (31), as well as other non-cancer diseases such as asthma (32) and cardiovascular disease (33). In our study, however, the use of either hospital or population controls did not produce a significantly stronger association between GSTT1 null polymorphism and RA.

Further analysis regarding smoking history was carried out due to the fact that cigarette smoking is an obvious risk factor for RA, and that GST genes are implicated in the metabolism of enzymes concerning harmful effects of oxidative stress. After grouping according to smoking history, the GSTT1 null genotype was not corelated with an increased risk of RA in either smokers or non-smokers. Meanwhile, there was also not a statistically difference for smokers with GSTT1 null genotype compared to non-smokers with positive genotype. However, we observed an increased risk in heavy smokers (cigarette consumption >10 pack-years) with GSTT1 null polymorphism compared with never or light smokers (cigarette

consumption ≤10 pack-years) with GSTT1 present.

Since serological results is an indispensable contributor to diagnosis and evaluation of activity and prognosis of autoimmune diseases including RA, after analysing the relationship between the GSTT1 null genotype and the RA risk in relation to the serological results, we suggested that there was no association between GSTT1 null genotype and the risk of RA in relation to either seropositive or seronegative results.

However, compared to RA of GSTT1 positive polymorphism with seronegative results, we found that there was an increased risk in GSTT1 null polymorphism with seropositive results, including rheumatoid factor positive or anti-citrullinated protein antibody positive.

We then determined the GSTT1 polymorphism in relation to patients' sex. However, the sex research included only a female subgroup analysis due to unclear raw data of male subgroup. Our data revealed that no association between the GSTT1 null genotype and female patients with RA. Therefore, the result must be interpreted with caution. RA is a consequence of multiple risk factors, and the interaction between environmental and genetic factors is generally accepted. GSTs represent a family of detoxifying enzymes that play an important role in protecting cells against the injuring effects of RA (34-35). GSTT1, a member of GSTs super-family, is investigated commonly. However, we found no significant as-

sociations between GSTT1 null polymorphism and the risk of RA. The lack of an association with GSTT1 may be due to differences in tissue-specific genetic expression or to a different role of certain substrates of GST in the pathogenesis of RA (23).

As in most meta-analysis, we have to carefully consider the possible limitations of the study in interpreting the results. First, only published studies in English and Chinese were included in the meta-analysis; therefore a publication bias may have occurred. The funnel plot shows significant evidence of the bias (Fig. 5). It is widely accepted that positive results usually have a greater probability of being published, and an overestimation of the GSTT1 null effect may appear if unpublished studies are not included, even though they are of more superior quality than published ones. Second, the results of an observational study may be influenced by unmeasured confounders such as physical activity, infective factors, and environmental effect; these may be related to RA but not evaluated in our study. Third, in our data, moreover, the description of cigarette-smoking condition and serological results was showed in reports, suggesting the association between GSTT1 null polymorphism and the risk of RA in relation to pathogenic factors. However, the criteria of subgroup analysis varied in reports, leading to the conclusion that further analysis could not be made. Fourth, there is probably another potential source of heterogeneity, but due to the fact that certain factors were determined in only one study, and some studies did not display the raw data, we were unable to explore them further in subgroup analysis. If the authors of all the published studies could share their data, a more precise meta-analysis could be performed.

It is well known that using hospital-based controls could generate some source of bias (36). Therefore, the utilisation of population-based controls is more appropriate.

We obtained the evidence of publication bias as shown by the funnel plot (Fig. 5). The shape of the funnel plots seemed symmetrical, suggesting the

absence of publication bias. In addition, Rosenthal's fail-safe-number (FSN) is probably one of the best known statistics in the context of meta-analysis aimed to estimate the number of unpublished studies in meta-analyses required to bring the meta-analytic mean effect size down to a statistically insignificant level (28). In our study, the fail-safe number (Nfs 0.05=12) is large, suggesting that publication bias probably has little effect on summary estimates.

Finally, meta-analysis remains retrospective research that is subject to the methodological deficiencies of the included studies. We minimised the likelihood of bias before initiating the study by developing a detailed protocol, including a meticulous search for published studies and using explicit methods for study selection, data extraction and data analysis.

In conclusion, the results from this meta-analysis suggested that GSTT1 null genotype is not associated with an increased susceptibility to RA. However, GSTT1 null polymorphism may increase the risk of RA in relation to heavy smokers or seropositive results. On the other hand, whether GSTT1 polymorphism acts in synergy with other genes or environmental factors needs to be studied more in depth.

References

1. SILMAN AJ, PEARSON JE: Epidemiology and genetics of rheumatoid arthritis. *Arthritis Res* 2002; 4 (Suppl. 3): S265-72.
2. JAWAHEER D, LUM RF, AMOS CI, GREGERSEN PK, CRISWELL LA: Clustering of disease features within 512 multicase rheumatoid arthritis families. *Arthritis Rheum*, 2004; 50: 736-41.
3. BARTON A, BOWES J, EYRE S, SYMMONS D, WORTHINGTON J, SILMAN A: Investigation of polymorphisms in the PADI4 gene in determining severity of inflammatory polyarthritis. *Ann Rheum Dis* 2005; 64: 1311-5.
4. TABERNER M, SCOTT KF, WEININGER L, MACKAY CR, ROLPH MS: Overlapping gene expression profiles in rheumatoid fibroblast-like synoviocytes induced by the proinflammatory cytokines interleukin-1 beta and tumor necrosis factor. *Inflamm Res* 2005; 54: 10-6.
5. PALUCKA AK, BLANCK JP, BENNETT L, PASCUAL V, BANCHEREAU J: Cross-regulation of TNF and IFN-α in autoimmune diseases. *Proc Natl Acad Sci USA* 2005; 102: 3372-7.
6. LI X, SUNDQUIST J, SUNDQUIST K: Risks of rheumatic diseases in first- and second-generation immigrants in Sweden .A nationwide follow-up study. *Arthritis Rheum* 2009; 60: 1588-96.
7. MIDWOOD K, SACRE S, PICCININI AM *et al.*: Tenascin-C is an endogenous activator of Toll-like receptor 4 that is essential for maintaining inflammation in arthritic joint disease. *Nat Med* 2009; 15: 774-80.
8. BOWES J, BARTON A: Recent advances in the genetics of RA susceptibility. *Rheumatology* 2008; 47: 399-402.
9. GREGERSEN PK, AMOS CI, LEE AT *et al.*: REL, encoding a member of the NF-kappaB family of transcription factors, is a newly defined risk locus for rheumatoid arthritis. *Nat Genet* 2009; 41: 820-3.
10. PRAHALAD S, HANSEN S, WHITING A *et al.*: Variants in TNFAIP3, STAT4, and C12orf30 loci associated with multiple autoimmune diseases are also associated with juvenile idiopathic arthritis. *Arthritis Rheum* 2009; 60: 2124-30.
11. DAHA NA, KURREEMAN FAS, MARQUES RB *et al.*: Confirmation of STAT4, IL2/IL21, and CTLA4 polymorphisms in rheumatoid arthritis. *Arthritis Rheum* 2009; 60: 1255-60.
12. STAHL EA, RAYCHAUDHURI S, REMMERS EF *et al.*: Genome-wide association study meta-analysis identifies seven new rheumatoid arthritis risk loci. *Nat Genet* 2010; 42: 508-14.
13. SEIDEGARD J, PERO RW, MILLER DG, BEATTIE EJ: A glutathione transferase in human leukocytes as a marker for the susceptibility to lung cancer. *Carcinogenesis* 1986; 7: 751-3.
14. MATTEY DL, HUTCHINSON D, DAWES PT *et al.*: Smoking and disease severity in rheumatoid arthritis: association with polymorphism at the glutathione-S-transferase M1 locus. *Arthritis Rheum* 2002; 46: 640–6.
15. YUN BR, EL-SOHEMY A, CORNELIS MC, BAE SC: Glutathione S-transferase M1, T1, and P1 genotypes and rheumatoid arthritis. *J Rheumatol* 2005; 32: 992-7.
16. GRABAR PB, LOGAR D, TOMSIC M, ROZMAN B, DOLZAN V: Genetic polymorphisms of glutathione S-transferases and disease activity of rheumatoid arthritis. *Clin Exp Rheumatol* 2009; 27: 229-36.
17. HAYES JD, PULFORD DJ: The glutathione S-transferase supergene family: regulation of GST and the contribution of the isoenzymes to cancer chemoprotection and drug resistance. *Crit Rev Biochem Mol Biol* 1995; 30: 445-600.
18. EATON DL, BAMMLER TK: Concise review of the glutathione S-transferases and their significance to toxicology. *Toxicol Sci* 1999; 49: 156-64.
19. WEBB G, VASKA V, COGGAN M, BOARD P: Chromosomal localization of the gene for the human theta class glutathione transferase (GSTT1). *Genomics* 1996; 33: 121-3.
20. PARK JH, EL-SOHEMY A, CORNELIS MC, KIM HA, KIM SY, BAE SC: Glutathione S-transferase M1, T1, and P1 gene polymorphisms and carotid atherosclerosis in Korean patients with rheumatoid arthritis. *Rheumatol Int* 2004; 24: 157-63.
21. MATTEY DL, HASSELL AB, DAWES PT *et al.*: Influence of polymorphism in the manganese superoxide dismutase locus on disease outcome in rheumatoid arthritis: evidence for interaction with glutathione S-transferase genes. *Arthritis Rheum* 2000; 43: 859-64.
22. MORINOBU S, MORINOBU A, KANAGAWA S, HAYASHI N, NISHIMURA K, KUMAGAI S: Glutathione S-transferase gene polymorphisms in Japanese patients with rheumatoid arthritis. *Clin Exp Rheumatol* 2006; 24: 268-73.
23. KEENAN BT, CHIBNIK LB, CUI J *et al.*: Effect of interactions of glutathione S-transferase T1, M1, and P1 and HMOX1 gene promoter polymorphisms with heavy smoking on the risk of rheumatoid arthritis. *Arthritis Rheum* 2010; 62: 3196-210.
24. MATTEY DL, HASSELL AB, PLANT M *et al.*: Association of polymorphism in glutathione S-transferase loci with susceptibility and outcome in rheumatoid arthritis: comparison with the shared epitope. *Ann Rheum Dis* 1999; 58: 164-8.
25. LAYTON MA, JONES PW, ALLDERSEA JE *et al.*: The therapeutic response to D-penicillamine in rheumatoid arthritis: influence of glutathione S-transferase polymorphisms. *Rheumatol* 1999; 38: 43-7.
26. PADYUKOV L, SILVA C, STOLT P, ALFREDSSON L, KLARESKOG L, FOR THE EPIDEMIOLOGICAL INVESTIGATION OF ARTHRITIS STUDY GROUP: A gene–environment interaction between smoking and shared epitope genes in HLA–DR provides a high risk of seropositive rheumatoid arthritis. *Arthritis Rheum* 2004; 50: 3085-92.
27. GHELANI AM, SAMANTA A, JONES AC, MASTANA SS: Association analysis of TNFR2, VDR, A2M, GSTT1, GSTM1, and ACE genes with rheumatoid arthritis in South Asians and Caucasians of East Midlands in the United Kingdom. *Rheumatol Int* 2011; 31: 1355-61.
28. ROBERT G. ORWIN: A Fail-Safe N for effect size in meta-analysis. *J Educational Statistics* 1983, 8: 157-9.
29. GARTE S: The role of ethnicity in cancer susceptibility gene polymorphisms: the example of CYP1A1. *Carcinogenesis* 1998; 19: 1329-32.
30. GARTE S, GASPARI L, ALEXANDRIE AK *et al.*: Metabolic gene polymorphism frequencies in control populations. *Cancer Epidemiol Biomarkers Prev* 2001; 10: 1239-48.
31. DEAKIN M, ELDER J, HENDRICKSE C *et al.*: Glutathione S-transferase GSTT1 genotypes and susceptibility to cancer: studies of interactions with GSTM1 in lung, oral, gastric and colorectal cancers. *Carcinogenesis* 1996; 17: 881-4.
32. SAADAT M, SAADAT I, SABOORI Z, EMAD A: Combination of CC16, GSTM1, and GSTT1 genetic polymorphisms is associated with asthma. *J Allergy Clin Immunol* 2004; 113: 996-8.
33. DONEY AS, LEE S, LEESE GP, MORRIS AD, PALMER CNA: Increased cardiovascular morbidity and mortality in type 2 diabetes in associated with the glutathione S-transferase theta-null genotype: a Go-DARTS study. *Circulation* 2005; 111: 2927-34.
34. BAKER MS, FEIGAN J, LOWTHER DA: Chondrocyte antioxidant defences: the roles of catalase and glutathione peroxidase in protection against H2O2 dependent inhibition and proteoglycan biosynthesis. *J Rheumatol* 1988; 15: 670-7.
35. STEVENS CR, BENBOUBETRA M, HARRISON R, SAHINOGLU T, SMITH EC, BLAKE DR: Localisation of xanthine oxidase to synovial endothelium. *Ann Rheum Dis* 1991; 50: 760-2.
36. SADETZKI S, BENSAL D, NOVIKOV I, MODAN B: The limitations of using hospital controls in cancer etiology-one more example for Berkson's bias. *Eur J Epidemiol* 2003; 18: 1127-31.