Yonsei Medical Journal
Vol. 45, No. 4, pp. 683 - 689, 2004

Glutathione S-Transferase M1 Status and Breast Cancer Risk: A Meta-Analysis

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It is not yet clear whether Glutathione S-transferase M1 (GSTM1) polymorphisms affect the risk of breast cancer. The aim of this study is to provide a comprehensive meta-analysis of all the available, published case-control studies on the extent of the possible association between GSTM1 polymorphisms and susceptibility to breast cancer. Twenty case-control studies on GSTM1 and breast cancer were identified using both PUBMED and a manual search. Meta-analysis was conducted by the Peto method. Subgroup analyses were undertaken, in order to explore the relationship between effect sizes and the study characteristics. The overall odds ratio (OR) was found to be 1.06 (95% CI, 0.99-1.14). The OR for post-menopausal women with GSTM1 deficiency was determined to be 1.19 (95% CI, 1.05-1.34). In populations with a low frequency of GSTM1 deficiency, a greater increase was observed (OR, 1.20; 95% CI, 1.08-1.34). Furthermore, the highest associations were found in post-menopausal women with a low frequency of GSTM1 deficiency (OR, 1.44; 95% CI, 1.20-1.73). The fact that GSTM1 deficiency is not rare in the general population implies that the attributable risk for breast cancer could be sizable. Further studies focusing on the structure of haplotype blocks of GSTM1 are required in order to find a specific haplotype with a predisposing breast cancer susceptibility allele.

Key Words: Breast cancer, GSTM1, polymorphism

INTRODUCTION

Breast cancer is the most common cancer in women, and the leading cause of cancer death in women worldwide.[1] The risk of breast cancer in women probably results from complex interactions between many genetic and environmental factors.[2] Although some of the familial risk is attributable to the shared environment, there may be other, common, low-penetrance genetic variants, which alter predispositions toward breast cancer.

Glutathione S-transferase M1 (GSTM1) is one of these proposed, low-penetrance susceptibility genes. The GSTM1 gene is polymorphic, and at least four alleles exist. Null mutations of this gene have been linked with an increase in a number of cancers, most likely due to increased susceptibilities to environmental toxins and carcinogens.[3]

It is not yet clear whether these GSTM1 polymorphisms affect the risk of breast cancer. Zhong et al.[4] first reported an association between GSTM1 deficiency (null type) and breast cancer. Since the publication of their report in 1993, over 19 studies have appeared in the literature, alternately confirming or refuting this association.[5-23] One of the major problems of the published studies is that most of them have been based on only small samples. Our study focuses on a meta-analysis of all the available published case-control studies, in order to assess the extent of possible association between GSTM1 polymorphisms and susceptibility to breast cancer.

Received March 18, 2004
Accepted July 3, 2004

This study was supported by a grant of the Korea Health 21 R&D Project, Ministry of Health & Welfare, Republic of Korea. (03-PJ1-PG3-21000-0015).

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MATERIALS AND METHODS

Selection of studies

A PUBMED search of the literature was con-

ducted in order to identify studies with information regarding GSTM1 polymorphisms and breast cancer risk, using the search terms 'GSTM1', 'breast', 'cancer' and 'polymorphisms'. We also conducted a manual search of reference lists from original research papers and review articles. Studies that met the following criteria were included in the review: 1) case-control studies of GSTM1 polymorphisms in association with breast cancer susceptibility, and 2) studies written in English with available full-text versions. Among the studies that met the inclusion criteria, we excluded the following: 1) Studies which contained overlapping data, 2) studies in which the number of null and wild genotypes could not be ascertained, and 3) studies in which only family members had been studied. These articles were reviewed independently by two of the authors (J.W.S. and C.M.N.) to determine whether or not the articles met the inclusion criteria of our present study.

Statistical analysis

The odds ratio (OR) of breast cancer associated with GSTM1 deficiency was estimated for each study. Meta-analysis was conducted by the Peto method, which is a modification of the Mantel-Haenszel method. Peto's estimate can be obtained from the weighted average of differences in the observed and expected number of events in a specific group for each study.[24,25] A fixed effect model was used to estimate overall effect size if the effect sizes were homogeneous across studies; otherwise, a random effect model was used. In order to check the homogeneity of effect sizes, we used Q-test statistic, which was the weighted sum of the squared difference between the overall effect size and the effect size from each study. Subgroup analyses were performed to investigate the further effects of GSTM1 according to factors such as: menopausal status, year of publication, and degree of GSTM1 deficiency (%). In order to evaluate potential publication bias, we plotted sample size against effect sizes. Data analysis was performed using Metawin (version 2.0; Sinauer Associates, Inc., Sunderland, MA) and SAS software (version 8.1; SAS Institute, Cary, NC).

RESULTS

Using the key words 'GSTM1', 'breast', 'cancer' and 'polymorphisms', we found a total of 50 publications in PUBMED. Of the 29 papers that met inclusion criteria 1 and 2 described above, 9 were excluded due to the exclusion criteria described above. Ten articles were actually divided into 20 independent studies according to different subpopulations inherent in the studies. Therefore, a total of 30 case-control studies were included in the present analysis. The characteristics of the studies included in this meta-analysis are shown in Table 1. Ten studies involved post-menopausal women as study populations, and nine involved pre-menopausal women.

The sizes of the case-control studies varied substantially, from 118 to 2341 subjects. The frequencies of GSTM1 deficiency ranged from 28.6 to 64.0% in the control groups. The OR with respect to an association between GSTM1 deficiency and breast cancer risk also varied substantially, from 0.64 to 2.58 (Table 2). A Q-test was performed to assess homogeneity of ORs across the studies, but it provided no statistically significant evidence for heterogeneity (p=0.06). The overall OR (Peto estimate) was 1.06 (95% CI, 0.99-1.14) according to the fixed effect model.

Table 3 summarizes the overall effect sizes in subgroups of studies according to year of publication (<July 2000, ≥July 2000), frequencies of GSTM1 deficiency in the controls (<50.4%, ≥ 50.4%), and menopausal status. The OR for pre-menopausal women with GSTM1 deficiency was 1.02 (95% CI, 0.88 - 1.18), and for post-menopausal women with GSTM1 deficiency was 1.19 (95% CI, 1.05 - 1.34). A greater increase was observed in populations with a low frequency of GSTM1 deficiency (<50.4%).

DISCUSSION

Dunning et al.[26] pooled 6 of the available case-control studies, and concluded that GSTM1 deficiency conferred a 1.14-fold increase in the risk of breast cancer (95% CI, 0.97-1.35) and a 1.33-fold increase in the risk of post-menopausal breast cancer (95% CI, 1.01-1.76). In contrast to

Table 1. Characteristics of Case-Control Studies Included in the Meta-analyses

Study No.	Author, Year of publication	Place of Study	Sample Ethnicity	Menopausal-Status
1	Zhong et al., 1993	UK	Caucasian	Pre, Post
2	Ambrosone et al., 1995	USA	Caucasian	Post
3	Kelsey et al., 1997	USA	Caucasian	Pre, Post
4	Helzlsouer et al., 1998 a*	USA	Caucasian	Post
5	Helzlsouer et al., 1998 b	USA	Caucasian	Pre
6	Charrier et al., 1999 a	France	French	Post
7	Charrier et al., 1999 b	France	French	Pre
8	Garcia-Closas et al., 1999 a	USA	Caucasian	Post
9	Garcia-Closas et al., 1999 b	USA	Caucasian	Pre
10	Curran et al, 2000	Australia	Caucasian	Pre, Post
11	Millikan et al., 2000 a	USA	African-American Whites	Post
12	Millikan et al., 2000 b	USA	African-American Whites	Pre
13	Park et al., 2000 a	Korea	Korean	Post
14	Park et al., 2000 b	Korea	Korean	Pre
15	Krajinovic et al., 2001	Canada	French-Canadian	Pre, Post
16	Mitrunen et al., 2001 a	Finnish	Finnish	Post
17	Mitrunen et al., 2001 b	Finnish	Finnish	Pre
18	Gudmundsdottir et al., 2001	Iceland	Icelandic	Pre, Post
19	Zheng et al., 2001	USA	NA	Post
20	Amorim et al., 2002 a	Brazil	White	Pre, Post
21	Amorim et al., 2002 b	Brazil	Non-white	Pre, Post
22	Zheng et al., 2002 a	USA	NA	Post
23	Zheng et al., 2002 b	USA	NA	Pre
24	Matheson et al, 2002	Australia	NA	Pre
25	Van der Hel et al., 2003 a	Netherlands	Dutch	Post
26	Van der Hel et al., 2003 b	Netherlands	Dutch	Pre
27	Khedheier et al., 2003	Tunisia	Tunisian	Pre, Post
28	Egan et al., 2004	China	Chinese	Pre, Post
29	Roodi et al., 2004 a	USA	Caucasian	Pre, Post
30	Roodi et al., 2004 b	USA	African-American	Pre, Post

*Lowercase letters (a, b) represent different subpopulations in the same study.
NA: non-available.

these findings, however, several other studies showed no association between the GSTM1 genotype and breast cancer risk. There are several potential reasons for these inconsistencies in the outcomes of the above studies; they may arise from population stratification or admixture, un-

Table 2. Odds Ratio and 95% Confidence Intervals of Each Study

Study No.	Author	Case		Control		Ln (OR)	95% CI of ln (OR)
		N	GSTM1 Deficiency (%)	N	GSTM1 Deficiency (%)		
1	Zhong	197	47.7	225	41.8	0.239	(-0.145, 0.624)
2	Ambrosone	177	52.5	233	50.2	0.093	(-0.298, 0.483)
3	Kelsey	244	57.8	245	51.4	0.256	(-0.100, 0.612)
4	Helzlsouer a*	86	65.1	87	41.4	0.948	(0.352, 1.543)
5	Helzlsouer b	24	62.5	25	64.0	-0.063	(-1.213, 1.087)
6	Charrier a	135	60.0	107	43.0	0.679	(0.172, 1.186)
7	Charrier b	226	53.1	330	53.9	-0.034	(-0.373, 0.305)
8	Garcia-Closas a	357	49.9	346	48.8	0.041	(-0.255, 0.336)
9	Garcia-Closas b	78	52.6	86	52.3	0.010	(-0.602, 0.621)
10	Curran	129	56.6	129	55.8	0.031	(-0.460, 0.522)
11	Millikan a	322	40.1	334	39.2	0.035	(-0.278, 0.348)
12	Millikan b	324	40.5	294	45.2	-0.196	(-0.515, 0.123)
13	Park a	74	52.7	80	57.5	-0.193	(-0.826, 0.441)
14	Park b	114	62.3	97	49.5	0.518	(-0.027, 1.063)
15	Krajinovic	147	54.4	207	52.7	0.071	(-0.352, 0.494)
16	Mitrunen a	317	46.1	277	37.5	0.349	(0.022, 0.675)
17	Mitrunen b	164	45.7	201	47.8	-0.081	(-0.494, 0.331)
18	Gudmundsdottir	500	54.6	395	54.2	0.017	(-0.248, 0.282)
19	Zheng	202	49.5	481	51.8	-0.090	(-0.419, 0.238)
20	Amorim a	79	41.8	123	52.8	-0.441	(-1.001, 0.123)
21	Amorim b	49	34.7	133	28.6	0.289	(-0.423,1.000)
22	Zheng a	233	53.6	209	50.7	0.117	(-0.256, 0.491)
23	Zheng b	84	47.6	124	54.0	-0.256	(-0.808, 0.297)
24	Matheson	157	58.0	157	49.0	0.357	(-0.086, 0.800)
25	Van der Hel a	102	61.8	128	47.7	0.565	(0.045, 1.086)
26	Van der Hel b	127	55.1	135	50.4	0.190	(-0.295, 0.674)
27	Khedheier a	309	53.7	242	57.0	-0.133	(-0.471, 0.205)
28	Egan	1135	56.2	1206	56.6	-0.017	(-0.181, 0.146)
29	Roodi a	203	57.6	203	61.6	-0.163	(-0.559, 0.233)
30	Roodi b	54	37.0	59	40.7	-0.152	(-0.905, 0.602)
	Overall effect size of ln(OR)					0.060	(-0.010, 0.129)
	Overall effect size of OR					1.06	(0.99, 1.14)

*Lowercase letters (a, b) represent different subpopulations in the same study.

Table 3. Differences of Odds Ratios in Subgroups of Studies According to the Study Characteristics

Characteristics	Number of studies	Case		Control		OR	95%CI
		N	GSTM1 Deficiency (%)	N	GSTM1 Deficiency (%)		
Year of publication*							
< July 2000	14	2299	50.1	2441	47.6	1.12	0.99-1.25
≥ July 2000	16	3605	53.2	4018	51.6	1.04	0.95-1.14
GSTM1 deficiency(%)*							
< 50.4	15	2682	49.0	2813	44.2	1.20	1.08-1.34
≥ 50.4	15	3667	54.6	4085	54.8	0.98	0.89-1.07
Menopausalstatus							
Pre	9	1298	50.4	1449	50.2	1.02	0.88-1.18
Post	10	2005	50.4	2282	46.7	1.19	1.05-1.34

*Median value was used to dichotomize the characteristics.

known menopausal status, or lack of information regarding the risk factors known to confer breast cancer risk.[14] Our result, which analyzed 30 studies, showed a slightly higher risk among women with GSTM1 deficiency type than among those with wild type. Our conclusions are consistent with another recent meta-analysis.[22]

Age appears to be relevant in the determination of probable exposure to carcinogens.[27] A difference in age between cases and controls is, therefore, a potential source of bias. When summarizing the results of 16 studies which adjusted for age and other potential risk factors of breast cancer, the overall odds ratio of GSTM1 deficiency was 1.03 (95% CI, 0.94-1.12) which was compatible with our main findings. Our results also suggest that GSTM1 deficiency is more strongly associated with late onset breast cancer. Ambrosone et al.[5] suggested elevated risk among the youngest post-menopausal women. Previous other studies also found a significantly increased risk in the subgroup of post-menopausal breast cancer.[7,14,20] With regard to the early onset of breast cancer, BRCA1 and BRCA2 are known to play the most important role in breast cancer susceptibility.[28] But, these results do not suggest any role of GSTM1 in the early onset of breast cancer.

GSTM1 deficiency differed according to ethnicity. Frequencies were higher among Caucasians (50.2%) and Asians (56.3%) than among African-Americans (40.7%). Frequencies differed significantly even within an ethnicity. For example, GSTM1 deficiency ranged from 37.5 - 64.0% in Caucasians (Table 2). We found an increased risk in populations with a low frequency of GSTM1 deficiency. Also, the highest association was found in post-menopausal women with a low frequency of GSTM1 deficiency (OR, 1.44; 95% CI, 1.20-1.73; data not shown).

This meta-analysis was not without its limitations. Firstly, because most of the studies included

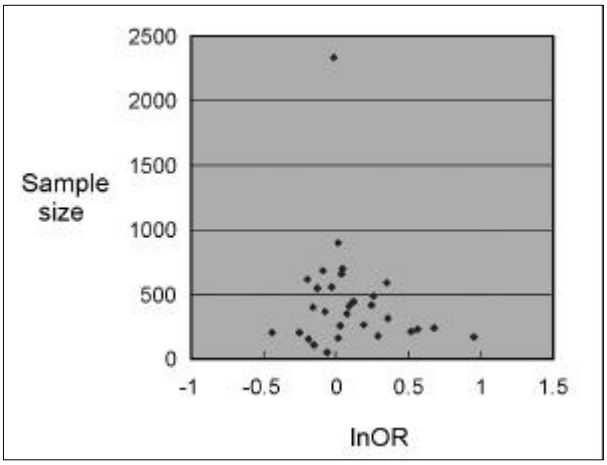

Fig. 1. Funnel Plot of Sample Size by Effect Size for Published Studies.

in this review were done in Western countries, the results obtained have limited relevance in Asian countries such as Korea, due to the ethnic variations noted above. Secondly, a possible publication bias might have been introduced as we included only published studies. However, an examination of funnel plots showed no evidence of a strong publication bias (Fig. 1).

In conclusion, there was a slightly higher breast cancer risk among women with GSTM1 deficiency than among those with wild type. Most notably, this association was higher in post-menopausal women and populations with a low frequency of GSTM1 deficiency. The fact that GSTM1 deficiency is not rare in the general population implies that the attributable risk for breast cancer could be sizable. Further studies focusing on the interaction between GSTM1 and environmental risk factors, and the structure of haplotype blocks of GSTM1 and haplotype pathway, are required in order to find a specific genomic region with a predisposing breast cancer susceptibility allele.

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