Association Between Sulfotransferase 1A1 Genotype and Survival of Breast Cancer Patients Receiving Tamoxifen Therapy

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Background: **Human sulfotransferase 1A1 (SULT1A1) catalyzes the sulfation of a variety of phenolic and estrogenic compounds, including 4-hydroxytamoxifen (4-OH TAM), the active metabolite of tamoxifen. A functional polymorphism in exon 7 of the SULT1A1 gene (SULT1A1*2) has been described that generates an enzyme that has approximately twofold lower activity and is less thermostable than that of the common allele SULT1A1*1. We investigated the hypothesis that that high sulfation activity would increase the elimination of 4-OH TAM by examining whether the presence of this polymorphism affects the efficacy of tamoxifen therapy.** *Methods:* **We examined the relationship between the SULT1A1*2 allele and survival in a cohort of 337 women with breast cancer who received tamoxifen (n = 160) or who did not (n = 177). SULT1A1 genotype was determined by restriction fragment polymorphism analysis. Patient survival was evaluated according to SULT1A1 genotype using Kaplan–Meier survival functions. Hazard ratios (HRs) were calculated from adjusted Cox proportional hazards modeling. All statistical tests were two-sided.** *Results:* **Among tamoxifen-treated patients, those who were homozygous for the SULT1A1*2 low-activity allele had approximately three times the risk of death (HR = 2.9, 95% confidence interval [CI] = 1.1 to 7.6) as those who were homozygous for the common allele or those who were heterozygous (SULT1A1*1/*2). Among patients who did not receive tamoxifen, there was no association between survival and SULT1A1 genotype (HR = 0.7, 95% CI = 0.3 to 1.5).** *Conclusions:* **Sulfation of 4-OH TAM provides a previously unanticipated benefit, possibly due to alterations in the bioavailability of the active metabolite or to undefined estrogen receptor-mediated events. These data alternatively suggest that variability in the metabolism of tamoxifen may affect its efficacy. [J Natl Cancer Inst 2002;94:1635–40]**

Antiestrogen therapy is a widely used method of treatment for hormone-responsive breast cancers, and tamoxifen is the most common antiestrogen administered in the United States. Analysis performed by the Early Breast Cancer Trialist's Collaborative Group in 1998 demonstrates that tamoxifen prolongs survival in breast cancer patients. Tamoxifen treatment was also associated with a 47% reduction in the occurrence of contralateral breast cancer in patients receiving tamoxifen for approximately 5 years *(1)*. However, some patients experience recurrence despite tamoxifen therapy. The pharmacology and metabolism of tamoxifen are complex, and differences in patient outcomes could be due to individual variation in the metabolism of tamoxifen.

Human sulfotransferase 1A1 (SULT1A1) enzyme catalyzes the sulfation of a variety of phenolic and estrogenic compounds, including endogenous and environmental estrogens and the 4-hydroxy metabolite of tamoxifen, 4-OH TAM *(2)*. Sulfation of 4-OH TAM, catalyzed by SULT1A1, may have a substantial impact on the efficacy of tamoxifen therapy, because 4-OH TAM is capable of binding to the estrogen receptor with much greater affinity than the parent compound. Consequently, SULT1A1 activity may be an important determinant in the efficacy of tamoxifen treatment, and modulation of SULT1A1 activity by genetic and/or environmental factors could influence therapeutic outcome.

A genetic polymorphism in exon 7 of the SULT1A1 gene results in an amino acid change from arginine to histidine at the conserved position 213 *(3,4)*. This polymorphism has functional consequences for the translated protein in that the variant allele (His213, SULT1A1*2) has an approximately twofold lower catalytic activity and decreased thermostability. When we assayed SULT1A1 enzymatic activity in platelets from 279 human subjects, individuals with the homozygous SULT1A1*2/*2 genotype had statistically significantly lower activity than individuals who were heterozygous (SULT1A1*1/*2) or homozygous (SULT1A1*1/*1) for the high-activity SULT1A1*1 common allele *(5)*. The SULT1A1 genotype has also been investigated in relation to risk of breast cancer. In a cohort of postmenopausal women, investigators found a statistically significant association between the variant SULT1A1*2 allele and increased risk of breast cancer *(6)*. Another study *(7)* found no association between the SULT1A1 genotype and risk of breast cancer at younger ages but did find a positive association between genotype and age of onset of breast cancer. Although findings of these studies are not entirely consistent, they suggest a role for SULT1A1 in breast cancer risk. Because SULT1A1 has a role in the metabolism of 4-OH TAM, the lower catalytic activity associated with the SULT1A1*2 allele can be hypothesized to affect prognosis in women treated with tamoxifen for breast cancer.

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To test the hypothesis that the SULT1A1 genotype would affect the outcome of tamoxifen treatment, we conducted a retrospective study among a cohort of women treated for primary breast cancer. We evaluated survival according to SULT1A1 genotype among a group of women who were treated with tamoxifen. To test the specificity of the association, we also examined SULT1A1 genotype and survival among breast cancer patients who did not receive tamoxifen therapy.

Subjects and Methods

Study Subjects

Patients who received their first course of adjuvant treatment for primary invasive breast cancer at the Arkansas Cancer Research Center, University of Arkansas for Medical Sciences (UAMS), from 1985 through 1996 were identified from hospital tumor registry records. Patients with a prior history of cancer were excluded. Hospital tumor registry records were used to obtain information concerning age, stage at diagnosis, tumor size, tumor grade, hormone receptor status, race, and date and type of therapy received (chemotherapy, radiotherapy, surgery, and hormonal treatment). The hospital tumor registry was also the source of follow-up information; the registry conducts active follow-up for each patient, contacting the physician or the patient annually and recording the date last contacted and vital status. The study protocol was approved by the Institutional Review Board of UAMS.

Archived tissues, stored in paraffin blocks in pathology department storage, were used as a source of DNA for genotyping. Therefore, patients had to have a pathology report and blocks available at UAMS to be included in the study. Pathology reports were reviewed to identify paraffin blocks that contained normal tissue. A block containing normal lymph nodes was selected if available; if normal lymph nodes were not available, a block containing other tissue noted as histologically normal was used. Pathology reports were also reviewed to confirm registry information on tumor characteristics.

Hospital tumor registry records identified 815 patients with invasive breast cancer who had been treated at UAMS from 1985 through 1996. One hundred twenty-four (15%) of these patients had received no adjuvant therapy (chemotherapy, radiation therapy, or tamoxifen) and were excluded from the study, as were five (1%) patients with missing adjuvant therapy information. Most of the excluded patients who did not receive adjuvant therapy had lymph node-negative disease. During a pilot phase of data collection, we obtained pathology reports and blocks for a sample of patients. We found that 1) pathology records could more easily be obtained for more recent diagnoses and 2) poor DNA quality was sometimes a problem for older tissue samples. Therefore, for the main phase of data collection we excluded 198 patients with earlier years of diagnosis. We attempted to obtain pathology reports and blocks for 488 eligible women diagnosed from 1990 through 1996. Tissue blocks were unavailable for 18 of the eligible patients because they did not have surgery, and no normal tissue was available for 23 patients. Pathology reports and/or blocks were unavailable at UAMS for an additional 133 patients; in most cases, this was because the patient had had surgery at another institution before being referred to UAMS for adjuvant therapy. Blocks containing normal tissue were obtained for 314 (64%) of the 488 eligible patients diagnosed from 1990 through 1996. Some patient characteristics were different among subjects for whom blocks were unavailable compared with those for whom blocks were available. All the patients who did not have surgery had advanced disease at diagnosis. The patients who were referred to UAMS after diagnosis and surgery elsewhere included a higher proportion of women who were under age 40 years and who had adverse prognostic characteristics, i.e., positive lymph nodes and negative estrogen receptor status. Despite these differences between registry-identified patients with and without samples, the case group with samples can be considered representative of women who received surgery and adjuvant therapy at the institution where the study was based. The normal tissue sample set included the 314 normal tissue samples from women diagnosed from 1990 through 1996, plus an additional 46 samples obtained during the pilot data collection phase for patients diagnosed from 1985 through 1989, a total of 360 samples. Sixteen subjects for whom samples were obtained were excluded because hormone receptor status was missing, and one subject was excluded because of missing race information, so 343 samples from eligible subjects were available for analysis. No polymerase chain reaction (PCR) product was obtained for six (2%) of the 343 samples. These patients with missing genotype were excluded from the data analysis. Thus, a total of 337 patients/samples were included in the final analysis.

DNA Extraction and Genotyping

Sections (50-μm thick) were cut from archived paraffin normal tissue blocks, the tissue was deparaffinized, and DNA was extracted using a commercially available kit (Qiagen Inc., Valencia, CA). SULT1A1 genotyping was performed as previously described *(4,5)*. Briefly, specific primers were used to amplify a 281-base pair fragment of the SULT1A1 gene encompassing the polymorphic base. The PCR product was exposed to the restriction endonuclease *Hae*II; the resulting fragments were resolved on a 3% Metaphor agarose gel (FMC BioProducts, Rockland, ME) and visualized by ethidium bromide staining and UV transillumination. The polymorphism consists of a G→A transition at nucleotide 638 in exon 7, which abolishes the recognition site for the restriction endonuclease. Therefore, the PCR product from patients homozygous for the SULT1A1*2 variant allele will exhibit a band of 281 base pairs, corresponding to the original PCR product. Digestion of the PCR product of the homozygous common allele SULT1A1*1 generates two fragments of 181 and 100 base pairs; heterozygotes exhibit all three bands.

Statistical Analysis

Stata software (Stata Corp., College Station, TX) was used for statistical analysis. The SULT1A1 genotype in relation to study subject characteristics was evaluated by Pearson's chi-square test and Fisher's exact test. Survival analysis according to SULT1A1 genotype was conducted separately for the two groups of patients (those who received tamoxifen and those who did not). Phenotype–genotype correlation studies from our laboratory *(5)* have demonstrated that platelet phenol sulfotransferase activity for individuals with either one or two common SULT1A1*1 alleles are similar (1.2 and 1.4 nmol/min/mg protein, respectively), whereas the activity in individuals homozygous for the SULT1A1*2 allele conferred by the homozygous SULT1A1*2 genotype is statistically significantly lower (0.74 nmol/min/mg protein) than that in the other two groups. Therefore, we combined the SULT1A1*1/*1 and SULT1A1*1/*2

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genotypes for survival analysis. Overall survival was calculated from time of diagnosis to time of death or the end of follow-up (October 1999, for most subjects), and survival by genotype was compared using Kaplan–Meier survival functions and log-rank tests for equality of survival functions. The relative risk of death in women who were homozygous for the variant SULT1A1*2 allele compared with women who possessed at least one SULT1A1*1 common allele was estimated by calculating the hazard ratio from a Cox proportional hazards model. We first used a univariate analysis and then a multivariate model with adjustment for prognostic factors for breast cancer survival. In the adjusted model, stage of disease was included as a stratifying variable, and age, race, and hormone receptor status were included as covariates. Proportionality of hazards over time for the SULT1A1*2/*2 group compared with the combined SULT1A1*1/*1 and SULT1A1*1/*2 reference group was assessed qualitatively, by visual assessment of the survival functions, and quantitatively, by a test of scaled Schoenfeld residuals from the Cox proportional hazards models. Interaction between treatment and genotype was evaluated using the likelihood ratio test.

Results

We obtained normal tissue samples and complete baseline information for 337 eligible study subjects, 160 of whom had received adjuvant tamoxifen therapy and 177 of whom had not been treated with tamoxifen. Tamoxifen therapy is normally indicated for patients with estrogen receptor-positive (ER+) tumors; among ER+ patients in the present study, several other patient characteristics were associated with tamoxifen therapy, including later year of diagnosis. One hundred six (80%) of 132 ER+ patients diagnosed from 1993 through 1996 received tamoxifen, compared with 32 (36%) of 90 ER+ patients diagnosed in 1992 or earlier (P<.001). Ninety-five (70%) of 135 of ER+ patients aged 50 years or older received tamoxifen, compared with 43 (49%) of 87 ER+ patients younger than 50 years (P = .001). Eighty-six (69%) of 125 ER+, lymph node-negative patients received tamoxifen, compared with 52 (54%) of 97 ER+ patients with lymph node-positive disease (P = .02).

Registry follow-up information was obtained in October 1999. Eligible patients were diagnosed over a broad time interval, from 1985 through 1996, so the opportunity to follow subjects ranged from 3 to 14 years. Censoring of survival for living patients was therefore primarily related to year of diagnosis. Follow-up of patients by the registry was almost complete, with few subjects lost to follow-up; 97% of living subjects had last contact dates in 1998 or 1999. The median follow-up time for patients who were alive at the end of the observation period was 65 months. There were 34 deaths reported during the follow-up period in the tamoxifen-treated group and 67 deaths in the group receiving therapies that did not include tamoxifen.

Study subject characteristics are presented in Table 1. The

Table 1. Characteristics of breast cancer patients stratified by sulfotransferase 1A1 (SULT1A1) genotype†

Characteristics	All subjects (n = 337)	SULT1A1 genotype			
		*1/*1 (n = 145)	*1/*2 (n = 147)	*2/*2 (n = 45)	*P*‡
Race					
Caucasian	275 (81.6)	118 (81.4)	118 (80.3)	39 (86.7)	.62
African-American	62 (18.4)	27 (18.6)	29 (19.7)	6 (13.3)	
Age at diagnosis, y					
⩽39	33 (9.8)	15 (10.3)	14 (9.5)	4 (8.9)	.41
40–49	108 (32.0)	45 (31.0)	50 (34.0)	13 (28.9)	
50–59	92 (27.3)	43 (29.7)	38 (25.9)	11 (24.4)	
60–69	65 (19.3)	25 (17.2)	33 (22.5)	7 (15.6)	
⩾70	39 (11.6)	17 (11.7)	12 (8.2)	10 (22.2)	
Stage (AJCC)§					
I	108 (32.1)	49 (33.8)	41 (27.9)	18 (40.0)	.26
II	162 (48.1)	72 (49.7)	74 (50.3)	16 (35.6)	
III	50 (14.8)	20 (13.8)	21 (14.3)	9 (20.0)	
IV	17 (5.0)	4 (2.8)	11 (7.5)	2 (4.4)	
Estrogen receptor status					
Positive	222 (65.9)	89 (61.4)	100 (68.0)	33 (73.3)	.26
Negative	115 (34.1)	56 (38.6)	47 (32.0)	12 (26.7)	
Progesterone receptor status					
Positive	172 (51.0)	66 (45.5)	84 (57.1)	22 (48.9)	.13
Negative	165 (49.0)	79 (54.5)	63 (42.9)	23 (51.1)	
Tumor grade‖					
I	48 (17.1)	21 (18.1)	22 (17.6)	5 (12.5)	.90
II	109 (38.8)	44 (37.9)	47 (37.6)	18 (45.0)	
III	124 (44.1)	51 (44.0)	56 (44.8)	17 (42.5)	
Diagnosis‖					
IDC	281 (92.1)	115 (91.3)	126 (91.3)	40 (97.6)	.38
ILC	24 (7.9)	11 (8.7)	12 (8.7)	1 (2.4)	
Tumor size, cm‖					
⩽2	156 (46.9)	71 (49.3)	63 (43.5)	22 (50.0)	.69
2.1–3	86 (25.8)	34 (23.6)	39 (26.9)	13 (29.6)	
>3	91 (27.3)	39 (27.1)	43 (29.7)	9 (20.5)	

†Values in the table are number of patients (%).

‡Statistical significance was determined using the chi-square test.

§AJCC = American Joint Committee on Cancer.

‖IDC = infiltrating ductal carcinoma; ILC = infiltrating lobular carcinoma. The total is less than 337 because, for some subjects, information on tumor grade, diagnosis, or tumor size was missing.

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distribution of SULT1A1 alleles was 43.0% for SULT1A1*1/*1, 43.6% for SULT1A1*1/*2, and 13.4 % for SULT1A1*2/*2. The allele frequencies for the SULT1A1*1 common allele were 64% and 67% for Caucasian and African-American subjects, respectively. These allele frequencies are consistent with those in the published literature *(3,4)*. We considered the distribution of SULT1A1 genotype by several demographic and pathologic characteristics of the study subjects; the associations between SULT1A1 genotype and patient characteristics were consistent with what would be expected by chance, as detailed in Table 1.

The Kaplan–Meier function for survival by SULT1A1 genotype is shown in Fig. 1, A, for tamoxifen-treated patients and in Fig. 1, B, for patients not receiving tamoxifen. The proportion surviving at 5 years among tamoxifen-treated patients was 0.88 (95% confidence interval [CI] = 0.81 to 0.93) for those with the SULT1A1*1/*1 or SULT1A1*1/*2 genotype and 0.64 (95% CI = 0.48 to 0.88) in the SULT1A1*2/*2 group. The log-rank test for equality of survivor functions in the tamoxifen-treated group showed that the reference group, SULT1A1*1/*1 and SULT1A1*1/*2, had better survival (26 deaths observed compared with 31 expected) than the SULT1A1*2/*2 group (eight deaths observed compared with three expected) ($P = .004$).

As shown in Table 2, Cox proportional hazards models revealed that, among the tamoxifen-treated subjects, women with the SULT1A1*2/*2 genotype were at an approximately threefold higher risk of death than those with SULT1A1*1 genotypes (hazard ratio [HR] = 2.9; 95% CI = 1.1 to 7.6). The difference in HRs persisted when the analysis was adjusted for age, race, stage of disease at diagnosis, and hormone receptor status. There was no evidence of departure from the proportional hazards assumption. Additional adjustment for year of diagnosis or tumor grade had essentially no effect on the HR associated with SULT1A1 genotype.

There were no detectable differences in survival according to SULT1A1 genotype among patients who were not treated with tamoxifen. The proportion of patients not treated with tamoxifen who were alive at 5 years was 0.63 (95% CI = 0.54 to 0.70) for SULT1A1*1/*1 and SULT1A1*1/*2 combined and 0.74 (95% CI = 0.48 to 0.88) for SULT1A1*2/*2. The SULT1A1*1/*1 and SULT1A1*1/*2 groups had 60 deaths observed, with 58 expected, and the SULT1A1*2/*2 group had seven deaths ob-

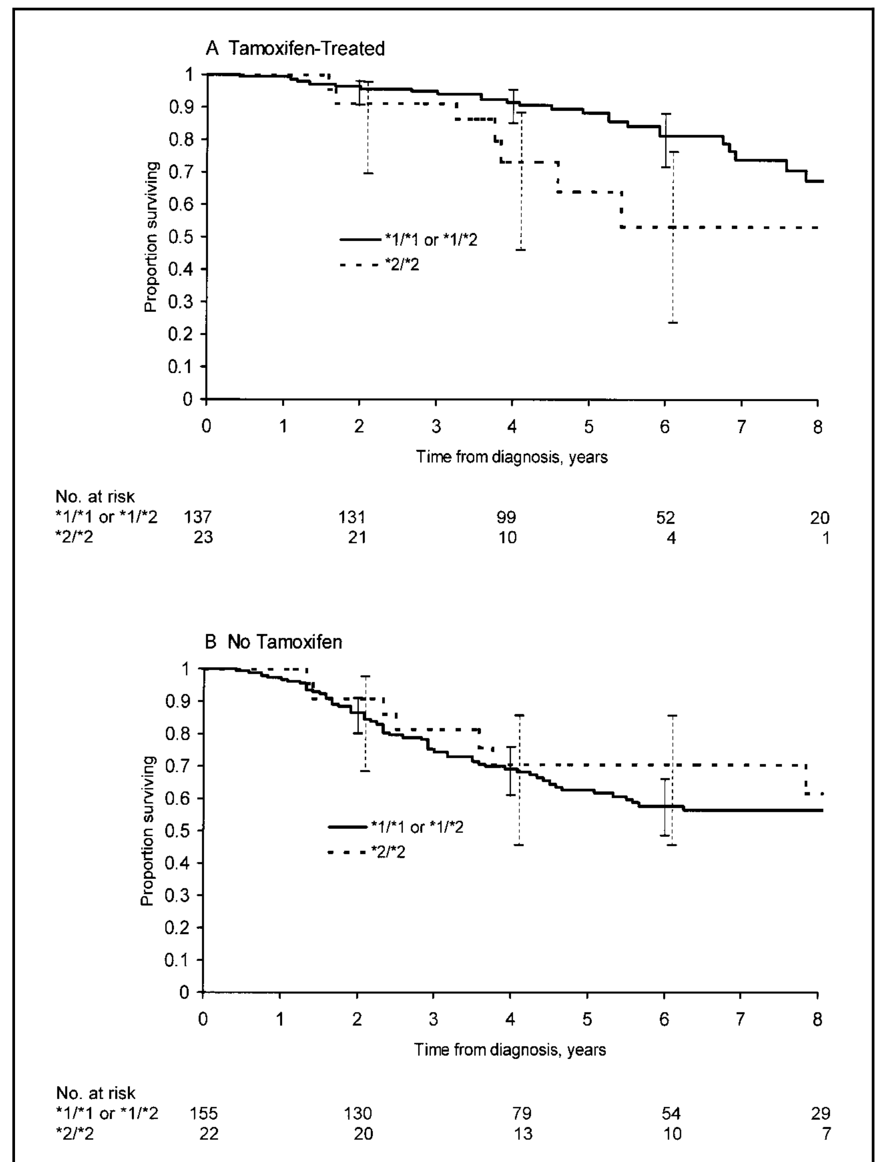

Fig. 1. Overall survival of breast cancer patients by SULT1A1 genotype. **A**) Patients receiving tamoxifen therapy. **B**) Patients not receiving tamoxifen therapy. The genotypes homozygous SULT1A1*1/*1, heterozygous SULT1A1*1/*2, and homozygous SULT1A1*2/*2 are abbreviated as *1/*1, *1/*2, and *2/*2 on the figure. The **solid lines** represent the patients with genotypes homozygous SULT1A1*1/*1 and heterozygous SULT1A1*1/*2 combined. The 95% confidence intervals (CIs) are shown at 2, 4, and 6 years after the time of diagnosis. The CIs drawn as **solid lines** go with the **solid line/curves** and those drawn as **dotted lines** go with the **broken line/curves.**

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Table 2. Survival of breast cancer patients stratified by sulfotransferase 1A1 (SULT1A1) genotype and by tamoxifen treatment status

SULT1A1 genotype	No. of patients	Person-years	Deaths	Crude HR (95% CI)†	Adjusted HR (95% CI)‡
Tamoxifen-treated					
*1/*1	73	388.4	12	1.0 (Referent)	1.0 (Referent)
*1/*2	64	354.8	14		
*2/*2	23	97.7	8	3.1 (1.4 to 7.1)	2.9 (1.1 to 7.6)
No tamoxifen					
*1/*1	72	391.0	25	1 (Referent)	1 (Referent)
*1/*2	83	369.0	35		
*2/*2	22	126.6	7	0.8 (0.4 to 1.7)	0.7 (0.3 to 1.5)

†Hazard ratios (HRs) and 95% confidence intervals (CIs) were calculated from a Cox proportional hazards model.

‡Adjusted for age, stage at diagnosis, race, and hormone-receptor status.

served, with nine expected ($P = .45$). Cox proportional hazards analysis revealed no association between survival and SULT1A1 genotype in patients who did not receive tamoxifen (HR = 0.7, 95% CI = 0.3 to 1.5).

In a model that included both patient groups (those who did and those who did not receive tamoxifen treatment), there was statistical evidence of interaction ($P = .02$) between SULT1A1 genotype and tamoxifen treatment. When the group of women who did not receive tamoxifen was restricted to patients with ER+ tumors (n = 84), there was no survival difference according to SULT1A1 genotype in the ER+ patients not receiving tamoxifen ($P = .73$).

DISCUSSION

In this study, we found that women treated with tamoxifen who were homozygous for the low-activity SULT1A1*2 allele had poorer survival than women who were homozygous or heterozygous for the high-activity SULT1A1*1 allele. This association persisted even when adjustments were made for age, race, clinical stage of tumor at diagnosis, and presence or absence of progesterone receptor. The same effect by genotype was not noted among women who did not receive hormonal therapy, indicating that variability in prognosis is through the impact of SULT1A1 on the metabolism of tamoxifen. To our knowledge, our study is the first to examine the relationship of SULT1A1 genotype and therapeutic outcome in women receiving tamoxifen for breast cancer.

SULT1A1 is the primary sulfotransferase isoform responsible for the sulfation of 4-OH TAM *(2)*. Sulfation of compounds such as estradiol is generally considered to result in their inactivation, in that sulfated estrogens are poor ligands for the estrogen receptor. Therefore, we had hypothesized that lower SULT1A1 activity conferred by the SULT1A1*2 allele would decrease clearance of 4-OH TAM and increase the efficacy of tamoxifen treatment. However, when we examined SULT1A1 genotype in relation to survival of breast cancer in patients receiving tamoxifen therapy, we found a strong association between survival and the SULT1A1*1 common allele, contrary to the expected outcome if higher activity did in fact result in rapid removal of the drug from the target tissue.

Although these results were the opposite of what we had expected based on our original hypothesis, there are some biologically plausible explanations for the observed association. These explanatory mechanisms include the possibility that sulfation of 4-OH TAM may modify the pharmacokinetics of tamoxifen therapy or beneficially alter the receptor-binding properties of 4-OH TAM. Additionally, tamoxifen treatment could lead to enhanced sulfation of other deleterious substrates.

Sulfation may affect the bioavailability of 4-OH TAM. Sulfation of most molecules facilitates their elimination; however, sulfation of steroids has been demonstrated to reduce the clearance of the steroid from the circulation *(8,9)*. For example, the C-19 steroid dehydroepiandrosterone (DHEA) is present in the circulation primarily as DHEA-sulfate (DHEAS). DHEAS binds tightly to albumin and is selectively reabsorbed in the kidney *(10);* both of these factors result in very slow clearance of DHEAS from the circulation. Delayed clearance of steroid sulfates can provide a reservoir of precursor molecules for the synthesis of active estrogens in target tissues such as the breast. Breast cells, including breast tumors, express the deconjugating enzyme steroid sulfatase, which regulates the transport of estrone- and DHEA-sulfate from the plasma into the tumor cells as unconjugated steroids *(11)*. It is possible that hepatic sulfation of 4-OH TAM, followed by reabsorption of the metabolite by the kidney, would slow the rate of clearance of 4-OH TAM. The levels of sulfated 4-OH TAM formed would be genotype-dependent, with individuals possessing the higher activity SULT1A1 alleles producing higher levels of circulating metabolite. Deconjugation of 4-OH TAM sulfate by steroid sulfatase expressed in breast tumors would then serve to deliver the active metabolite to the target site. Additionally, SULT1A1 is also expressed in breast tumors *(12)*. The expression of SULT1A1, in combination with steroid sulfatase, could lead to cycling of 4-OH TAM between the sulfated and nonsulfated forms within the tumor cell. This cycling could delay the excretion of 4-OH TAM from the cell and prolong the effective exposure of the drug at the target tissue. Again, this effect would be genotype-dependent.

It is also possible that there is an undefined beneficial activity of the sulfated form of 4-OH TAM. The interaction of sulfated 4-OH TAM with the estrogen receptor has not been specifically tested. Metabolism of tamoxifen to 4-OH TAM serves to increase the affinity of the metabolite for the estrogen receptor; subsequent metabolism by SULT1A1 may enhance this effect. Although it is probable that, analogous to sulfation of estradiol, sulfation of 4-OH TAM is an inactivation reaction in terms of receptor binding, the affinity of sulfated 4-OH TAM for the estrogen receptor has not been investigated and should be defined experimentally.

Another possible explanation for the association between high-activity SULT1A1 genotypes and survival in tamoxifen-treated patients involves global induction of the SULT1A1 enzyme. A recent study *(7)* has shown that exposure of the human breast cancer cell line ZR75–1 to 4-OH TAM results in a 10-fold increase in the amount of SULT1A transcript. Of the more than 8000 transcripts analyzed, only SULT1A was affected by this treatment. Although the authors could not determine whether the transcript was SULT1A1 or SULT1A2, expression of SULT1A2 as a protein has not been demonstrated in any human tissue thus far, although it has been detected as RNA in the liver *(4,13)*. It is therefore likely that the induced transcript is in fact SULT1A1. Increased expression of transcript results in elevated levels of SULT1A1 protein and elevated enzymatic activity *(4),* facilitating the detoxification of many potentially harmful endogenous and environmental substances. Increased amounts of SULT1A1 protein in breast tumors could also facilitate

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elimination of estrogenic substrates at that site, and expression of the high-activity SULT1A1 alleles could enhance this effect.

Thus, although the direction of our results was unexpected, we believe that our findings can be explained by a biologic mechanism. It is unlikely that the association we observed is the result of bias related to study design. In this cohort design, the association between the exposure of interest, SULT1A1 genotype, and survival could be influenced by selection bias only if follow-up and ascertainment of outcomes were differential according to genotype. Follow-up was almost complete, genotype was not strongly related to disease characteristics, as shown in Table 1, and it is unlikely that genotype would be related to any other characteristic that might cause a patient's death to be incorrectly classified by a hospital registry. Nevertheless, the size of the study population examined in this report is modest, so we cannot entirely rule out the possibility that the observed association is due to chance. The possibility also exists that the clinical effect observed is not due to the polymorphism in SULT1A1 but to a second, undefined polymorphism that is in linkage disequilibrium with SULT1A1. Studies on the mechanistic basis of the relationship between SULT1A1 genotype and overall survival of women treated with tamoxifen for breast cancer are currently underway in our laboratory. Although this study needs to be replicated, our results suggest that there may be decreased efficacy of tamoxifen among approximately 13% of the population, based on SULT1A1 genotype. Elucidation of the role of SULT1A1 in tamoxifen treatment may begin to explain therapeutic failures and could lead to modifications in treatment strategies that would result in more effective treatment of breast cancer.

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Note

Manuscript received March 13, 2002; revised August 15, 2002; accepted August 28, 2002.

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