Published in final edited form as:
Carcinogenesis. 2007 August ; 28(8): 1759–1764.

Hair dye use, genetic variation in N-acetyltransferase 1 (*NAT1*) and 2 (*NAT2*), and risk of non-Hodgkin lymphoma

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Abstract

Background—Several previous studies have found non-Hodgkin lymphoma (NHL) risk to be associated with hair dye use, particularly use of permanent, dark colors and use before 1980, when hair dye formulations changed.

Methods—We examined NHL risk in relation to reported hair dye use among 1,321 cases and 1,057 controls from a US population-based multicenter study. DNA was extracted from blood or buccal cells to identify genetic variation in N acetyltransferase 1 (*NAT1*) and 2 (*NAT2*), which encode enzymes that metabolize aromatic amine compounds found in hair dyes.

Results—Among women, 509 cases and 413 controls reported hair dye use (odds ratio [OR] =1.2,95% confidence interval [CI]=0.9,1.6). Risk estimates were higher for use before 1980 than for use in 1980 or later, particularly for use of permanent, intense tone (black, dark brown, dark blonde) products (<1980: OR=1.6,95%CI 0.9,2.7; ≥1980: OR=0.6,95%CI 0.4,1.1). Risk estimates were increased for women who used permanent, intense tone products before 1980 if they had the rapid/intermediate NAT2 phenotype (OR=3.3, 95%CI 1.3,8.6) or the *NAT1*10* allele (OR=2.5,95%CI 0.9,7.6), but not if they were slow *NAT2* acetylators (OR=1.5,95%CI 0.6,3.6) or had no copies of the *NAT1*10* allele (OR=1.5,95%CI 0.7,3.3). NHL risk was not increased among women who began hair dye use after 1980 or among men.

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Author contributions: Dr. Morton had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Dr. Morton also conducted the statistical analysis, and Drs. Morton and Zahm drafted the manuscript. All authors contributed to the study conception and design, acquisition of data, interpretation of data, and critical revision of the manuscript for important intellectual content.

Conflict of interest: Dr. David W. Hein is a consultant for hair dye manufacturers. All other authors declare no conflict of interest.

Conclusion—Our results support previous research demonstrating elevated NHL risk among women who used dark color or intense tone permanent hair dyes before 1980. We present the first evidence suggesting that this risk may differ by genetic variation in *NAT1* and *NAT2*.

Introduction

Non-Hodgkin lymphoma (NHL) has been associated with use of hair coloring products, particularly long-term use of dark permanent dyes, in several previous epidemiologic studies (1-9). In 1988, Cantor *et al.* (1) reported a two-fold increased risk of NHL among men who used hair coloring products. In 1992, Zahm *et al.* (2) reported odds ratios (ORs) of 2.0, 4.1, and 3.0 for NHL among women who used brown/brunette, black, and red dye, respectively, compared with women who never used hair coloring products. Long-term use of black hair dyes has also been associated with two- to four-fold increased NHL mortality (3;4). A case-control study by Zhang *et al.* (5) observed a two-fold increased risk of NHL among women who used dark-color permanent dyes for more than 25 years and began use prior to 1980, the approximate time when manufacturers removed from hair dye formulations some compounds (e.g., aromatic amine dyes) that had been found to be mutagenic and carcinogenic in animal studies (10-12). Similar findings of increased NHL risk associated with hair dye use prior to 1980 were recently reported by de Sanjose *et al.* in Europe (8).

Not all studies support a role for hair dyes in the etiology of NHL (7;9). The Nurses' Health Study cohort has not shown any increased risk for NHL among hair dye users (13), nor have several case-control studies (14-16). Additionally, no study has shown consistent and strong patterns of increasing risk with increasing frequency, duration, or early age at first use. In 1993, the International Agency for Research on Cancer classified the exposures of hairdressers and barbers and various compounds in hair dyes as probably or possibly carcinogenic (Groups 2A and 2B, respectively), but concluded that there was not sufficient evidence to evaluate personal use of hair dye (Group 3) (17).

It is plausible that the role of hair dyes in lymphomagenesis is modified by common genetic variation in the enzymes *N*-acetyltransferase 1 (*NAT1*) and 2 (*NAT2*), which are responsible for metabolizing aromatic amines such as those found in hair dyes. *NAT1* and *NAT2* detoxify and activate these compounds via *N*-acetylation and *O*-acetylation following *N*-hydroxylation, which can lead to the formation of carcinogenic intermediates. Both genotypic and phenotypic variation in *NAT1* and *NAT2* lead to variation in acetylation capacity, and therefore variation in the formation of carcinogenic intermediates (18). Thus, it is worthwhile to investigate the potential effects of hair dye use taking into account common genetic variation in these enzymes.

The possible carcinogenicity of hair coloring products poses an important public health issue given the high prevalence of use. The percentage of women reporting ever use of hair dyes increased dramatically from <10% to >50% with the introduction of one-step home-use kits around 1950. Over the last thirty years, the percentage of women currently using hair dyes in the United States has remained constant at approximately 40% (19;20), and cumulative lifetime use approaches 75% among women, based on prevalence of exposure among controls in epidemiologic case-control studies (21). Hair dye use in men is somewhat less common than in women; based on a market survey conducted in 2003, 25% of men in the United States reported current hair dye use (20). The high prevalence of use and the previous epidemiologic findings led us to evaluate the association between hair coloring products and NHL in a large population-based case-control study conducted in four areas of the United States.

Materials and Methods

Study Population

Study methods have been described in detail elsewhere (22-24). Briefly, cases with a histologically-confirmed, first primary diagnosis of NHL (International Classification of Diseases for Oncology [ICD-O], Second Edition codes 9590-91, 9595, 9670-73, 9675-76, 9680-88, 9690-91, 9695-98, 9700, 9702-03, 9705-11, 9713-15, 9823, 9827) (25), aged 20 to 74 years, diagnosed between July 1998 and June 2000, were identified among residents of four Surveillance Epidemiology and End Results (SEER) registries (Iowa, Los Angeles County, metropolitan Detroit, and metropolitan Seattle). Known HIV-positive cases were excluded.

Controls were selected from residents of the same four SEER areas, frequency matching to the cases by age (within five year age groups), sex, race, and SEER area. For cases under age 65, controls were selected by one-step list-assisted random digit dialing (26), with 78.5% of contacts providing household rosters. For cases aged 65 and older, controls were selected from Center for Medicare and Medicaid Services files of residents eligible for Medicare. Controls with a history of NHL or known HIV infection were excluded.

Of 2,248 presumed eligible cases, 320 (14%) died prior to interview, 127 (6%) could not be located, 16 (1%) had moved out of the area, and 57 (3%) had physician refusals. Of the remaining 1,728 cases, 274 (16%) declined to be interviewed and 133 (8%) never responded or were not interviewed due to illness, impairment, or other reasons. A total of 1,321 eligible cases were interviewed, yielding a participation rate of 76% of the cases we attempted to contact and an overall case response rate of 59%.

Of the 2,409 controls identified, 28 (1%) died before contact, 311 (13%) could not be located, and 24 (1%) had moved out of the geographic area. Of the remaining 2,046 controls, 839 (41%) declined to be interviewed and 150 (6%) did not participate due to illness, impairment, or other reasons. A total of 1,057 controls were interviewed, yielding a participation rate of 52% and an overall control response rate of 44%.

In-person interviews were conducted by trained personnel, who administered a computer-assisted personal interview (CAPI), blinded to case-control status. All study participants were asked to provide a venous blood or mouthwash buccal cell sample. We obtained blood samples from 773 cases and 668 controls, and buccal cell samples from 399 cases and 314 controls; 149 (11%) cases and 75 (7%) controls provided neither.

The study was approved by human subjects review boards at all institutions. Written informed consent was obtained from all subjects.

Exposure Assessment

In-person interviews included questions on ages of first and last use of each episode of use of temporary (products that wash out in one shampoo), semipermanent (products that rinse out after a few shampoos), permanent (products that last until the hair grows out), and gradual (e.g., Grecian Formula®) hair coloring products. Also ascertained were color (e.g., black, brown, blonde, red), tone (e.g., light, dark), brand name, frequency of use, and whether two components (e.g., color and developer) had to be mixed together. Intense tone products (i.e., black, dark brown, dark blonde) have higher concentrations of paraphenylenediamine and other dye intermediates than light or intermediate tone products (J. Skare, 2004, personal communication). Some processes, such as bleaching without adding color, were included in the screening list to elicit complete histories, but were not considered to involve hair dye exposure. Exposure histories were truncated one year prior to the date of diagnosis for cases

and date of selection for the controls. The interview included other known and suspected risk factors for NHL, such as family history of cancer, pesticide exposure, and medical history.

The methods for evaluating *NAT1* and *NAT2* genotypes and *NAT2* acetylation phenotype in our study population have been described previously by Morton *et al*. (24). Briefly, DNA was extracted from blood clots or buffy coats from 10 ml blood for 773 cases and 668 controls at SeraCare BioServices (Gaithersburg, MD) using Puregene Autopure DNA extraction kits (Gentra Systems, Minneapolis, MN). DNA was extracted from buccal cells for an additional 399 cases and 314 controls by phenol chloroform extraction methods (27). Genotyping was carried out at the NCI Core Genotyping Facility (Advanced Technology Center, Gaithersburg, MD) using validated assays on the Taqman (Applied Biosystems, Foster City, CA) or MGB Eclipse (Epoch Biosciences, Bothell, WA) platforms to identify ten single nucleotide polymorphisms (SNPs) in *NAT1* and *NAT2*. Sequence data and assay conditions are provided at http://snp500cancer.nci.nih.gov (28). The genotype frequencies among white, non-Hispanic controls were in Hardy-Weinberg equilibrium for all ten SNPs (Pearson chi-square statistic >0.05). The SNP data were used to assign the most likely *NAT1* and *NAT2* alleles previously identified in human populations (29). For analyses of *NAT1*, *NAT1*10* was designated as the at-risk allele based on previous research (30). For analyses of *NAT2*, we designated *NAT2*4/*4* as the referent genotype, as has been done previously, because it corresponds to an absence of the nucleotide substitutions that define the other *NAT2* genotypes (30). *NAT2* acetylation phenotype was assigned at the University of Louisville (by DWH) based on *in vivo* and *in vitro* data on allelic variation in catalytic activity (18). For analysis of *NAT2* phenotypes, individuals homozygous for *NAT2* rapid- and slow-acetylator alleles were designated as rapid- and slow-acetylators, respectively; individuals possessing one rapid- and one slow-acetylator allele (heterozygotes) were designated as intermediate-acetylators (31).

Statistical Analysis

Statistical analyses were performed using the SAS system, version 9.1 (SAS Institute, Inc., Cary, NC). Relative risk of NHL, or NHL subtype, was estimated using odds ratios (OR) and 95% confidence intervals (CI) derived from dichotomous and polytomous unconditional logistic regression models, respectively. Risk of NHL subtype was estimated for follicular lymphoma, diffuse large B-cell lymphoma (DLBCL), T-cell lymphomas, and other lymphomas; risk estimates are presented only for the two most common subtypes, follicular lymphoma and DLBCL, due to sparse numbers for other subtypes. P-values for the differences in risk estimates between follicular lymphoma and DLBCL were derived from a Wald chi-square statistic (one degree of freedom). We adjusted risk estimates for sex, age (< 45, 45-64, 65 + years), race, and SEER area. Additional adjustment by education, smoking status, history of farming, and having a first degree relative with a history of NHL or any lymphoproliferative malignancy did not materially alter (>10%) the parameter estimates, and those factors were excluded from the final models.

Results

Among women, 509 (83%) NHL cases and 413 (81%) controls reported use of hair coloring products (OR=1.2, 95% CI 0.9,1.6) (Table I). We observed little to no variation in risk by ever use of permanent dyes (OR=1.1), semi-permanent dyes (OR=1.1), temporary hair coloring (OR=1.2), or progressive or other products (OR=1.0). Women with 100 or more lifetime applications had a significantly elevated OR of 1.4 (95% CI 1.0,2.0), but risk did not vary meaningfully by age at first use, frequency of use, or duration of use (data not shown). Among men, 113 (16%) cases and 101 (19%) controls reported use of any hair coloring product (OR=0.9, 95% CI 0.6,1.2). Detailed analyses by product type, color, frequency, duration, and calendar year of use were based on small numbers of male users and did not show any

significant patterns of excess risk (data not shown); therefore the remainder of this report will present data for women only.

Table II presents data by calendar year women began use of any hair coloring product and use of permanent dark color or intense tone products. Compared with women who never used hair coloring products, risk estimates were higher among women who began using hair dyes before 1980 than among women who began use in 1980 or later, particularly for permanent, intense tone (i.e., black, dark brown, and dark blonde) products (before 1980: OR=1.6, 95% CI 0.9,2.7; 1980 or later: OR=0.6, 95% CI 0.4,1.1). Risk of NHL was four-fold for women who used permanent, intense color tone products for 15 or more years prior to 1980 (17 cases, 4 controls, OR=3.9, 95% CI 1.2,12.5), but no consistent dose-response patterns were observed with frequency, duration, or total lifetime applications (Table III). Analyses by NHL subtype revealed consistently higher risk estimates for follicular lymphoma than for DLBCL (Table IV).

Tables V and VI present data according to NAT2 phenotype and *NAT1* genotype, respectively, and characteristics of permanent hair coloring use before 1980. Increased NHL risk estimates associated with hair dye use before 1980 were observed among NAT2 rapid/intermediate acetylators, but not among NAT2 slow acetylators (Table V). These findings were again most pronounced for use of permanent, intense tone products (NAT2 rapid/intermediate acetylators: OR=3.3, 95% CI 1.3,8.6; NAT2 slow acetylators: OR=1.5, 95% CI 0.6,3.6). Women with one or two copies of the *NAT1*10* allele also had larger increases in NHL risk associated with use of permanent, dark color or permanent, intense tone products prior to 1980 than women with no copies of the *NAT1*10* allele (Table VI). However, risks did not increase consistently with number of applications per year, years of use, or total lifetime applications. Analyses by NHL subtype again revealed that the increased risk estimates for permanent hair coloring use before 1980 among women with the rapid/intermediate NAT2 phenotype, but not among slow NAT2 acetylators, and among women with one or two copies of the *NAT1*10* allele, but not among women with no copies of the *NAT1*10* allele, were more pronounced for follicular lymphoma than for DLBCL, albeit with wide confidence intervals due to small numbers (data not shown).

The NAT2 rapid/intermediate phenotype or the *NAT1*10* allele alone did not increase risk of NHL among women who never used any hair coloring products (Supplementary Table I). Risk was slightly elevated for women who began use of permanent hair dyes before 1980 among NAT2 slow acetylators and women without any copies of the *NAT1*10* allele, and risk increased further among NAT2 rapid/intermediate acetylators or who had at least one copy of the *NAT1*10* allele.

Discussion

The etiology of NHL remains largely unknown apart from some rare genetic syndromes, acquired immunodeficiency syndrome (AIDS) associated with human immunodeficiency virus infection, and a few other infections or medical conditions marked by immune dysregulation. Given the high prevalence of hair coloring products use, the presence of compounds in hair dyes that are carcinogenic and mutagenic in bioassays, and associations observed in previous epidemiologic studies, we evaluated the role of hair coloring products in this large case-control study by ascertaining detailed use data from interviews and jointly evaluating the exposure and potential genetic susceptibility.

On the whole, there was little evidence of increased risk of NHL among most users of hair dyes, but women who used dark color or intense tone (i.e., black, dark brown, dark blonde) permanent dyes prior to1980 had significantly increased risk. However, risk did not increase consistently with frequency, duration, or total lifetime applications. These findings are

consistent with several earlier studies (1;2;4-6;8), including the studies by Zhang *et al.* (5) and de Sanjose *et al.* (8) that also reported risk among long-term users of dark permanent hair coloring products who began use prior to 1980, but not among women who began use in recent decades after formulation changes were made [(10); J. Skare, 2004, personal communication]. We also observed that the risk estimates associated with use of dark color or intense tone permanent dyes prior to 1980 appeared to be higher for follicular lymphoma than for DLBCL, which is also consistent with the findings from Zhang *et al.* (5) and de Sanjose *et al.* (8), but should be confirmed in future research. Previous studies that have not shown associations between hair dye use and NHL (13-16) include the Nurses Health Study (13), which may be limited by ascertainment of exposure earlier in life than in case-control studies of NHL and by number of cases (21), and three case-control studies (14-16), two of which had no information on duration, age, or (for one study only) shade. These detailed characteristics of use were important determinants of risk in the current study and others (32).

Our study showed significantly increased risks associated with use of dark color or intense tone permanent hair dyes before 1980 among women if they had the *NAT2* rapid/intermediate phenotype or if they had one or two copies of the *NAT1*10* allele, but not if they were slow NAT2 acetylators or had no copies of the *NAT1*10* allele. In an earlier report from this study (24), we observed increased risk for NHL among individuals with the *NAT1*10/10* genotype compared with individuals with other *NAT1* genotypes, and a dose-dependent increase in risk among NAT2 intermediate- and rapid-acetylators in comparison with slow acetylators. These patterns were less evident among women than men, but not significantly so, accounting for the null association between *NAT1* and *NAT2* genetic variation and NHL risk presented here. The first study to examine malignant lymphoma in relation to acetylator status, using dapsone to determine acetylator phenotype, failed to find an association, possibly due to chance in this small study (33). Four previous epidemiologic studies, which had smaller numbers of subjects and genotyped fewer functional or potentially functional SNPs compared with our study, also observed no association between *NAT1* or *NAT2* genotypes and NHL risk (34-36), although one study suggested an association between the NAT2 slow-acetylation phenotype and low-grade lymphoma (37). There are no previous studies of *NAT1* and *NAT2* in relation to hair dye use and NHL risk. One study of bladder cancer has shown increased risk for hair dyes among NAT2 slow acetylators and those lacking the *NAT1*10* allele (38-40), whereas another study showed no association between hair dyes and risk of bladder cancer, even after stratifying by *NAT1* and *NAT2* (41). The distribution and levels of NAT expression is tissue specific, however, with NAT1 present in most tissues and NAT2 expressed predominantly in the liver and gastrointestinal tracts (42). Hair dyes are most likely to first encounter NAT1 in the skin, where it may affect metabolism of aromatic amines (42;43). The phenotype of *NAT1*10* is unclear, although the *NAT1*10* has been shown in some studies to be the rapid allele (42-44). NAT1 and NAT2 both detoxify and activate various aromatic amines. Increased risk of NHL associated with hair coloring product use among NAT2 rapid/intermediate acetylators is consistent with activation of aryldiamines via *N*-acetylation (31;45).

The exact compounds in hair dyes that could increase risk of lymphoma are not known because publicly available information on hair dye formulations and contents and changes over time is limited. Some hair dye compounds that were typically used in dark, permanent dyes and have been found to be carcinogenic or mutagenic in animal studies, such as 2,4-diaminoanisole (also called 4-methoxy-m-phenylenediamine, or 4MMPD), 2,4-diaminotoluene, and 4-amino-2-nitrophenol, were removed by manufacturers in the late 1970s and early 1980s, although others, such as 2-nitro-*p*-phenylenediamine, were still in use in the late 1990s (10;19). Current hair dye formulations have also been shown to contain 4-aminobiphenyl (4-ABP), which is known to be a bladder carcinogen but has not been linked to lymphoma risk (46). In the late 1970s and early 1980s, manufacturers also removed some carcinogenic azo dyes, such as Direct Black 38 and Direct Blue 6 (10;19;47), which are metabolized to benzidine, a known bladder

carcinogen that has been associated with risk of lymphoma in some studies (48;49). Importantly, *N*-acetylation activates benzidine from benzidine-based dye exposure *in vivo* in humans to form highly reactive DNA-binding compounds, which can gain access to the hematopoietic compartment (50). Other undocumented changes in hair dye formulations and contents are also likely to have occurred. Based on the limited availability of information on hair dye contents, our observations of increased NHL risk among women who used dark color or intense tone permanent hair dyes before 1980 are consistent with known changes in hair dye formulations.

The strengths of this study include its population-based design, relatively large number of cases, detailed exposure data, calendar time of the exposure histories, and incorporation of genetic susceptibility. Our study was based on over 1300 NHL cases (over 600 women), was able to evaluate the effect of the product changes over time, and collected detailed hair coloring product histories based on methods developed by Johns Hopkins University and Clairol, Inc. (51). Our study was the first NHL investigation to consider hair dye tone, an important determinant of risk, instead of color alone, and acetylation status. Our study was also the first to compute the actual frequency, duration, and cumulative use of hair coloring products prior to 1980, rather than just stratifying by first use prior to 1980 versus 1980 or after, as has been done previously. If the calendar period of use does, in fact, affect NHL risk associated with hair coloring product use, this method of exposure assessment should substantially reduce exposure misclassification. Despite our more detailed assessment, we still did not observe consistent patterns in risk with frequency, duration, or total lifetime applications. Potential confounders were dealt with by exclusion (HIV-positive subjects), adjusted for in the analysis (age, sex, study area), or found not to meaningfully affect risk estimates (education, smoking status, history of farming, and having a first degree relative with a history of NHL or any lymphoproliferative malignancy).

Although our finding that the increased NHL risk was limited to women who used dark color or intense tone permanent hair dyes before 1980 is consistent with the other studies that also considered time period of use (5;8), we cannot rule out the possibility that we observed no increased risk with hair dye use after 1980 because insufficient time has passed for the induction/latency period. Additional potential limitations of the current study include the low response rates, possible misclassification or bias in exposure data, possible confounding, disease heterogeneity within NHL, and chance. The loss of eligible subjects due to refusal to participate is typical of current population-based studies (52), and we know of no data suggesting differential participation by hair dye use. In our study, demographic characteristics (age, education, sex) for individuals who provided blood compared with those who provided buccal cells and compared with those who provided neither blood nor buccal cell samples were equivalent within each study site (53). Errors in recall of hair coloring product use are likely to be small for ever versus never use, color, and tone, but may be greater for reporting age started and stopped, and frequency of use. Although our results by NHL subtype were consistent with previous literature, confirmation of differences in the effects of hair dye use by NHL subtype is warranted. Finally, chance may have played a role in our findings, particularly given the large number of comparisons made, although our findings showed internal consistency.

In summary, NHL risk was elevated among women who used dark color or intense tone permanent hair dyes before 1980, particularly for those women with the rapid/intermediate NAT2 phenotype and those women with one or two copies of the *NAT1*10* allele. There was no evidence of increased risk of NHL among women who began hair dye use after 1980, nor among men.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgements

The authors gratefully acknowledge the contributions of Ronald K. Ross, MD, University of Southern California, who participated in the early stages of this project before his untimely death. We also thank Michael Stagner of Information Management Services, Incorporated, for providing programming support; Geoffrey Tobias, National Cancer Institute, for study administration; Westat, Incorporated, for serving as the coordinating center; SeraCare BioServices for specimen handling; the NCI Core Genotyping Facility; and the SEER study coordinators, interviewers, and support staff for their diligent work.

Financial support: This research was supported in part by the National Cancer Institute, National Institutes of Health Intramural Research Program and National Cancer Institute contracts N01-PC-67010, N01-PC-67008, N01-PC-67009, N01-PC-65064, and N02-PC-71105. Studies at the University of Louisville were supported by National Cancer Institute grant CA-34627.

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Table I

Number of non-Hodgkin lymphoma cases and controls and odds ratios according to hair coloring product use and sex.[a]

Hair coloring product use	Women			Men		
	Cases	Controls	OR (95% CI)[b]	Cases	Controls	OR (95% CI)[b]
None	101	98	1.0 (referent)	596	444	1.0 (referent)
Any	509	413	1.2 (0.9,1.6)	113	101	0.9 (0.6,1.2)
Permanent	281	226	1.1 (0.8,1.6)	49	39	1.1 (0.7,1.8)
Semi-permanent	168	135	1.1 (0.8,1.6)	17	20	0.7 (0.4,1.5)
Temporary	119	101	1.2 (0.8,1.8)	28	17	1.2 (0.7,2.3)
Other	198	164	1.0 (0.7,1.5)	34	30	0.8 (0.5,1.3)

[a] Two cases and one control had unknown use and were excluded from the analysis.

[b] Odds ratio (95% CI), adjusted for age, race, and SEER area (unconditional logistic regression).

Table II

Number of non-Hodgkin lymphoma cases and controls and odds ratios according to hair coloring product type, color, tone, and calendar year of first use (women only).

	< 1980			≥ 1980		
Hair coloring product use	**Cases**	**Controls**	**OR (95% CI)[a]**	**Cases**	**Controls**	**OR (95% CI)[a]**
Any	317	260	1.2 (0.8,1.7)	192	148	1.1 (0.8,1.6)
Permanent dye (any)	169	123	1.3 (0.9,1.9)	111	102	0.9 (0.6,1.4)
Permanent, dark colors (black, brown, red)	102	76	1.3 (0.9,2.0)	87	75	1.2 (0.7,1.7)
Permanent, intense tones (black, dark brown, dark blonde)	61	36	1.6 (0.9,2.7)	33	42	0.6 (0.4,1.1)

[a] Odds ratio (95% CI), adjusted for age, race, and SEER area, relative to 101 cases and 98 controls who never used hair dye products.

Table III

Number of non-Hodgkin lymphoma cases and controls and odds ratios according to permanent hair coloring use before 1980, by color, tone, frequency, duration, and total lifetime applications (women only).

	Permanent dye (any)			Permanent, dark colors (black, brown, red)			Permanent, intense tones (black, dark brown, dark blonde)		
Hair coloring product use < 1980	**Cases**	**Controls**	**OR (95% CI)[a]**	**Cases**	**Controls**	**OR (95% CI)[a]**	**Cases**	**Controls**	**OR (95% CI)[a]**
Frequency (number of applications/year)									
1-4	34	28	1.1 (0.6,2.0)	20	19	1.1 (0.6,2.3)	20	9	2.2 (0.9,5.3)
5-7	60	39	1.5 (0.9,2.5)	43	26	1.7 (0.9,3.0)	22	10	2.1 (0.9,4.9)
≥ 8	75	56	1.2 (0.8,2.0)	39	31	1.2 (0.7,2.1)	19	17	1.0 (0.5,2.1)
Duration (years)									
1-4	64	42	1.5 (0.9,2.4)	42	30	1.4 (0.8,2.5)	30	20	1.5 (0.8,2.9)
5-14	62	47	1.2 (0.7,2.0)	37	31	1.1 (0.6,2.0)	14	12	1.0 (0.4,2.4)
≥ 15	43	34	1.2 (0.7,2.1)	23	15	1.5 (0.7,3.2)	17	4	3.9 (1.2,12.5)
Cumulative applications									
1-24	42	56	1.3 (0.8,2.2)	39	29	1.4 (0.8,2.5)	27	17	1.6 (0.8,3.2)
25-99	36	58	1.5 (0.9,2.5)	35	22	1.5 (0.8,2.8)	18	9	1.7 (0.7,4.2)
≥ 100	45	55	1.2 (0.7,1.9)	28	25	1.1 (0.6,2.1)	16	10	1.4 (0.6,3.4)

[a] Odds ratio (95% CI), adjusted for age, race, and SEER area, relative to 101 cases and 98 controls who never used hair dye products.

Table IV

Number of follicular lymphoma and DLBCL non-Hodgkin lymphoma cases and controls and odds ratios according to hair coloring product use before 1980, by type, color, and tone (women only).

Hair coloring product use	Controls	Follicular lymphoma		DLBCL		
		Cases	OR (95% CI)[a]	Cases	OR (95% CI)[a]	P[b]
None	98	24	1.0 (referent)	34	1.0 (referent)	
Any	260	90	1.5 (0.9,2.6)	90	1.0 (0.6,1.6)	0.172
Permanent dye (any)	123	49	1.9 (1.0,3.4)	40	0.9 (0.5,1.6)	0.045
Permanent, dark colors (black, brown, red)	76	28	1.8 (0.9,3.4)	22	0.9 (0.5,1.7)	0.100
Permanent, intense tones (black, dark brown, dark blonde)	36	17	2.0 (0.9,4.4)	51	1.1 (0.5,2.4)	0.209

[a] Odds ratio (95% CI), adjusted for age, race, and SEER area.

[b] P-value for difference between risk estimates for follicular lymphoma and DLBCL.

Table V

Number of non-Hodgkin lymphoma cases and controls and odds ratios according to *NAT2* phenotype and permanent hair coloring product use before 1980, by color, tone, frequency, duration, and total lifetime applications (women only).

Hair coloring product use	*NAT2 slow*			*NAT2 rapid/intermediate*		
	Cases	Controls	OR (95% CI)[a]	Cases	Controls	OR (95% CI)[a]
No hair dye < 1980	37	38	1.0 (referent)	32	41	1.0 (referent)
Any hair dye < 1980	139	109	1.3 (0.7,2.2)	103	73	1.9 (1.0,3.4)
Permanent dye (any)	68	48	1.3 (0.7,2.4)	58	37	2.0 (1.0,4.0)
Permanent, dark colors (black, brown, red)	45	26	1.6 (0.8,3.2)	36	25	2.2 (0.98,4.9)
Permanent, intense tones (black, dark brown, dark blonde)	23	14	1.5 (0.6,3.6)	25	11	3.3 (1.3,8.6)
Frequency (number of applications/year)						
1-4	8	1	-	7	5	2.3 (0.6,8.9)
≥ 5	15	13	0.9 (0.3,2.5)	18	6	4.2 (1.3,13.2)
Duration (years)						
1-4	14	8	1.6 (0.6,4.7)	11	7	2.4 (0.8,7.3)
≥ 5	9	6	1.3 (0.4,4.5)	14	4	5.4 (1.4,21.1)
Cumulative applications						
1-24	14	6	2.1 (0.7,6.6)	9	6	2.3 (0.7,7.9)
≥ 25	9	8	1.0 (0.3,3.1)	16	5	4.6 (1.4,15.7)

[a] Odds ratio (95% CI), adjusted for age, race, and SEER area.

- Odds ratio not calculated if observed less than 2.

Table VI

Number of non-Hodgkin lymphoma cases and controls and odds ratios according to *NAT1* genotype and permanent hair coloring use before 1980, by color, tone, frequency, duration, and total lifetime applications (women only).

	NAT1 any			*NAT1 *10/any + NAT1*10/*10*		
Hair coloring product use	**Cases**	**Controls**	**OR (95% CI)[a]**	**Cases**	**Controls**	**OR (95% CI)[a]**
No hair dye < 1980	46	43	1.0 (referent)	23	30	1.0 (referent)
Any hair dye < 1980	130	107	1.2 (0.7,2.0)	97	64	1.8 (0.9,3.6)
Permanent dye (any)	65	53	1.1 (0.6,2.0)	58	29	2.5 (1.1,5.9)
Permanent, dark colors (black, brown, red)	38	32	1.1 (0.6,2.2)	33	18	3.0 (1.1,8.1)
Permanent, intense tones (black, dark brown, dark blonde)	26	16	1.5 (0.7,3.3)	22	10	2.5 (0.9,7.6)
Frequency (number of applications/year)						
1-4	8	4	1.8 (0.5,6.6)	7	2	4.9 (0.7,34.4)
≥ 5	18	12	1.3 (0.5,3.2)	15	2	2.1 (0.6,6.8)
Duration (years)						
1-4	13	9	1.3 (0.5,3.6)	12	7	1.8 (0.5,6.4)
≥ 5	13	7	1.6 (0.5,4.6)	10	3	4.7 (0.9,25.2)
Cumulative applications						
1-24	12	8	1.4 (0.5,4.0)	11	5	2.8 (0.7,11.2)
≥ 25	14	8	1.5 (0.5,4.1)	11	5	2.3 (0.5,9.5)

[a] Odds ratio (95% CI), adjusted for age, race, and SEER area.