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Pigmentary characteristics and moles in relation to melanoma risk

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Although benign and atypical moles are considered key melanoma risk factors, previous studies of their influence were small and/or institution-based. We conducted a population-based case-control study in the state of New Hampshire. Individuals of ages 20–69 with an incident diagnosis of first primary cutaneous melanoma were ascertained through the New Hampshire State Cancer Registry. Controls were identified through New Hampshire driver's license lists and frequency-matched by age and gender to cases. We interviewed 423 eligible cases and 678 eligible controls. Host characteristics, including mole counts, were evaluated using logistic regression analyses. Our results showed that pigmentary factors, including eye color (OR = 1.57 for blue eyes compared to brown), hair color (OR = 1.85 for blonde/red hair color compared to brown/black), freckles before age 15 (OR = 2.39 for freckles present compared to absent) and sun sensitivity (OR = 2.25 for peeling sunburn followed by no tan or a light tan and 2.42 for sunburn followed by tan compared to tanning immediately), were related to melanoma risk; these associations held after adjustment for sun-related factors and for moles. In analyses confined to skin examination participants, the covariate-adjusted effects of benign and atypical moles were moderately strong. Compared to 0–4 benign moles, risk increased steadily for 5–14 moles (OR = 1.71), 15–24 moles (OR = 3.55) and $\geq$ 25 moles (OR = 4.33). Risk also increased with the number of atypical moles; compared to none, the ORs for having 1, 2–3, or $\geq$ 4 atypical moles were 2.08, 1.84 and 3.80, respectively. Although risk was highest for those with multiple benign and atypical moles, the interaction was not of statistical significance. Our findings, arising from the first population- and incidence-based study to evaluate atypical moles in relation to melanoma risk, confirm the importance of host susceptibility, represented by pigmentary factors and the tendency to develop benign or atypical moles, in the etiology of this disease.

Key words: melanoma; moles; atypical moles; pigmentary characteristics; sun sensitivity

Numerous epidemiologic studies of melanoma risk have shown a modest to moderate influence of pigmentary factors.[1–4] Associations with sunburn or sun exposure are also widely reported,[3,5–7] although findings are inconsistent.[8] Over the last 2 decades, in response to clinical reports of multiple benign and atypical moles occurring in nonfamilial melanoma patients,[9] epidemiologic studies have sought to quantify the relationship between these lesions and melanoma risk. Research results have demonstrated strong associations between melanoma risk and benign mole counts, whether self-counted[1] or assessed by examiners.[3,10–20] Although several studies have also shown strong associations between atypical moles, the putative melanoma precursors and melanoma risk, these efforts were wholly or in part institution-based and/or involved small numbers of cases.[11–16,19–22] We designed a large population-based case-control study to examine risk factors for melanoma, with a particular focus on atypical moles.

Material and methods

This study was approved by the Committee for the Protection of Human Subjects at Dartmouth College. All participants gave verbal consent (for interview) and/or signed consent (for the skin examination and medical record release).

Case subjects of ages 20 through 69 with a diagnosis of melanoma occurring between January 1995 and December 1998 were ascertained through the New Hampshire (NH) State Cancer Registry. Those potentially eligible for study enrollment were NH residents with a working telephone number who were able to participate in an English-speaking interview. We sent a letter to the physician of record requesting permission to contact the patient. If an objection was not received within a month, a letter introducing the study was mailed to the case subject, followed within 2 weeks by a telephone call from the interviewer. Using this approach, we enrolled 444 of 579 (77%) potentially eligible cases; 15 (3%) were excluded at their physician's request, 26 (4%) could not be reached, 30 (5%) had died and 64 (11%) declined to participate. Twenty-one enrolled cases were deemed ineligible; of these, 7 had a previous diagnosis of melanoma, 4 had an unknown primary site, 2 had tumors of acral lentiginous histology and for 8 persons the diagnosis of melanoma was not definitive. Thus, 423 cases of first primary cutaneous melanoma were available for analysis.

Control subjects were ascertained from lists of licensed drivers obtained through the NH Department of Motor Vehicles and were selected at random to achieve a gender and age (in 5-year age groups) distribution similar to that of case subjects. Controls were also selected to achieve a control:case ratio of 1.6 to allow separate studies of atypical moles within the control group. Potentially eligible control subjects were NH residents with a working telephone and an ability to participate in an English-speaking interview. A letter introducing the study was sent to potential control participants, followed within 2 weeks by a telephone call from the interviewer. We enrolled 684 of 1,121 (61%) potentially eligible control subjects; 87 (8%) could not be reached, 13 (1%) controls had died and 337 (30%) declined to participate. Of the 684 control participants, 6 were deemed ineligible due to a prior diagnosis of melanoma. Thus, 678 controls were available for analysis.

The 40-min telephone interview queried participants for demographic factors, pigmentary characteristics, episodes of sunburn, sunbathing habits and hours of recreational and occupational sun exposure. At the conclusion of the interview, participants were invited to undergo a physician-conducted skin examination during which the number and site of benign and atypical moles were recorded on a standardized form. A mole was considered benign if it was raised (palpable), symmetrical in shape, well-defined in border and uniform in coloration.[21] We counted benign moles that were of at least 3 mm in diameter.[21] We defined atypical moles according to criteria developed for our previous studies[23] and required at least 3 of the following: diameter of 5 mm or more, flat (macular) component, erythema, irregular border, ill-defined border and variegated color. Through the skin examination proce-

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TABLE I – ODDS RATIO AND 95% CONFIDENCE INTERVALS FOR THE ASSOCIATION BETWEEN PIGMENTARY CHARACTERISTICS AND MELANOMA RISK

Risk factor	Cases	Controls	OR (95% CI)[1]	OR (95% CI)[2]
	n (%; *n* = 423)	*n* (%; *n* = 678)		
Gender				
Female	200 (47.3)	330 (48.7)		
Male	223 (52.7)	348 (51.3)		
Age				
≤ 39	94 (22.2)	158 (23.3)		
40–59	214 (50.6)	331 (48.8)		
≥ 60	115 (27.2)	189 (27.9)		
Eye color				
Brown	97 (22.9)	234 (34.5)	1.00	1.00
Gray/green/hazel	128 (30.3)	207 (30.5)	1.49 (1.08–2.07)	1.21 (0.85–1.71)
Blue	196 (46.3)	235 (34.7)	2.01 (14.8–2.72)	1.57 (1.12–2.22)
Missing	2	2		
Hair color				
Brown/black	202 (47.8)	410 (60.5)	1.00	1.00
Light brown	142 (33.6)	202 (29.8)	1.44 (1.10–1.90)	1.13 (0.83–1.53)
Blonde/red	79 (18.7)	65 (9.6)	2.48 (1.72–3.59)	1.85 (1.22–2.78)
Missing	0	1		
Freckles age < 15				
No	141 (33.3)	372 (54.9)	1.00	1.00
Yes	277 (65.5)	300 (44.2)	2.48 (1.92–3.20)	2.39 (1.82–3.15)
Missing	5	6		
Sun sensitivity				
Tan immediately	14 (3.3)	68 (10.0)	1.00	1.00
Peeling sunburn, no tan	204 (48.2)	282 (41.6)	3.53 (1.93–6.45)	2.25 (1.19–4.23)
Peeling sunburn, freckle	20 (4.7)	42 (6.2)	2.35 (1.07–5.17)	1.10 (0.48–2.51)
Burn followed by tan	185 (43.7)	286 (42.2)	3.16 (1.73–5.79)	2.42 (1.29–4.53)
Missing	0	0		

[1]OR adjusted for age and gender.–[2]OR adjusted for variables shown.

dures, we assessed benign and atypical moles for 323 (76%) of participating cases and 424 (65%) of controls. Unless otherwise noted, atypical and benign mole counts, based on the skin examination, were assessed using the cut points shown in Table II.

Pigmentary factors (eye color, hair color, freckles, sun sensitivity) were self-reported at the time of the interview. In our analyses, eye color was considered in 3 levels, blue, gray/green/hazel and brown. Hair color was defined as the participant's natural hair color at age 20 and assessed as blonde/red (red, blonde, or strawberry blonde), light brown and brown/black (brown, dark brown, or black). Subjects were also asked whether they had freckles before the age of 15 (yes/no). Sun sensitivity was assessed by asking subjects how their skin would react if exposed to 1 hr of strong summer sunlight; answer options included sunburn with peeling followed by no tan or a light tan; sunburn with peeling followed by freckles; sunburn followed by tan; and immediate tanning.

We evaluated sun-related variables, including sunburn occurrences and hours of sun exposure, as potential confounders and effect modifiers. We asked the subject to report the number of peeling sunburns and the number of blistering sunburns occurring during 10-year age intervals starting at age 10. For each type of sunburn, we summed the number of occurrences over the age intervals. The 2 types of sunburn were then analyzed separately as the number of occurrences before age 20, age ≥ 20 and over the lifetime (up to the reference date).

Hours of sun exposure (capped at 10 hr per day) were calculated for 3 separate variables: sunbathing, recreational exposure and occupational exposure. We asked study participants how many times per summer they usually sunbathed, defined as relaxing in the sun or trying to get a tan, how many hours they sunbathed on a day of sunbathing, and the first and last months of sunbathing during 10-year age intervals, starting at age 10. Recreational sun exposure was assessed by asking subjects to report the number of years, number of episodes per year and number of hours per episode they participated in 11 standard outdoor recreational activities (and an unrestricted number of other activities), starting at age 10. For each activity (and for multiple renewals of the same activity), we obtained the first and last ages of participation (defined as at least 6 episodes per year). Occupational sun exposure was assessed by asking subjects to report the number of years they worked outdoors in the summer and the number of hours per week of summer, starting at age 6 (to accommodate farm work).

The analyses considered the number of hours of sun exposure represented by the measures described above (sunbathing, recreational and occupational sun exposure), as well as a variable representing the combined hours from all 3 sun exposure measures. For each individual sun exposure measure, and for the combined exposure variable, we evaluated exposure hours occurring before age 20, age ≥ 20 and over the lifetime (up to the reference date). The reference date was 1 year prior to the date of diagnosis for cases and randomly assigned to controls based on the frequency of diagnosis dates in the case group.

Preliminary analyses included frequency distributions and descriptive statistics. We used chi-square tests to assess differences in proportions; *t*-tests and ANOVAs were used to evaluate differences in means. Odds ratios (ORs) and 95% confidence intervals (CIs) were computed from logistic regression models[24] to examine the influence of host characteristics on melanoma risk. Initially, pigmentary characteristics and moles were evaluated singly in models that included terms for age and gender; multivariate models evaluated possible confounding. Sun-related factors assessed as potential confounders were those that were significantly associated with melanoma risk and were not mutually correlated. Two variables filled these requirements: the number of peeling sunburns before age 20 and the total hours of recreational sun exposure. Due to the strong association between sun sensitivity and variables representing sunburn, the number of peeling sunburns before age 20 was omitted from multivariate models evaluating the covariate-adjusted effects of sun sensitivity. Similarly, we omitted sun sensitivity from multivariate models assessing the effects of eye color, hair color and freckles adjusted for the 2 sun-related variables.

To test the hypothesis that atypical moles are markers of etiologic heterogeneity in melanoma, we evaluated the age and gender-adjusted effects of pigmentary characteristics and sun-related factors in 2 separate subgroups of cases and controls: those with

TABLE II – ODDS RATIOS AND 95% CONFIDENCE INTERVALS FOR THE RELATIONSHIP OF BENIGN AND ATYPICAL MOLES TO MELANOMA RISK

Number of moles	Cases (n = 323)	Controls (n = 424)	OR (95% CI)[1]	OR (95% CI)[2]	OR (95% CI)[3]
Benign moles					
0–4	106 (30.5%)	242 (69.5%)	1.00	1.00	1.00
5–14	119 (46.1%)	139 (53.9%)	2.00 (1.42–2.81)	1.71 (1.20–2.43)	1.71 (1.16–2.50)
15–24	58 (65.9%)	30 (34.1%)	4.56 (2.75–7.58)	3.38 (1.99–5.73)	3.55 (2.00–6.29)
≥ 25	40 (75.5%)	13 (24.5%)	7.16 (3.63–14.11)	3.63 (1.74–7.56)	4.33 (1.97–9.53)
Atypical moles					
None	198 (35.2%)	365 (64.8%)	1.00	1.00	1.00
Any	125 (67.9%)	59 (32.1%)	4.01 (2.78–5.77)	2.70 (1.82–4.01)	2.46 (1.62–3.75)
1	33 (57.9%)	24 (42.1%)	2.61 (1.49–4.57)	2.00 (1.12–3.56)	2.08 (1.13–3.83)
2–3	31 (63.3%)	18 (36.7%)	3.11 (1.68–5.77)	2.21 (1.16–4.20)	1.84 (0.93–3.63)
≥ 4	61 (78.2%)	17 (21.8%)	7.02 (3.95–12.47)	4.61 (2.51–8.46)	3.80 (1.98–7.31)

[1]Adjusted for age and gender.–[2]Adjusted for age, gender and the other lesion type.–[3]Adjusted for age, gender, the other lesion type, eye color, hair color, freckles before age 15, total hours of recreational sun exposure, number of peeling sunburns before age 20 and family history of melanoma.

atypical moles and those without atypical moles. Potential interactions between atypical moles and other factors were formally assessed using likelihood ratio tests, in which the model containing age, gender and the main effects of the 2 variables of interest was compared to a similar model additionally containing their interaction term.

Results

A larger proportion of cases compared to controls reported having light hair color (blonde/red; light brown), whereas controls were more likely to have dark hair (brown/black). Blue eyes and freckles before age 15 were also more commonly reported by cases than by controls. Compared to controls, cases appeared to be more sun-sensitive and were less likely to tan immediately in response to strong summer sunlight.

The pigmentary factors, assessed in the entire study population, were moderately associated with melanoma risk when evaluated singly in models containing terms for age and gender, and these effects were attenuated after mutual adjustment (Table I). After adjustment for the other pigmentary factors, the effect of gray/green/hazel eye color relative to brown eye color was compatible with chance. Blue eyes relative to brown were associated with a modest and statistically significant increase in risk. Relative to brown/black hair, the adjusted effects of light brown hair were compatible with chance, but blonde/red hair was modestly and significantly associated with risk. The effect of having freckles before age 15 compared to their absence was minimally reduced and remained moderately associated with risk after controlling for the other pigmentary factors. Although the influence of sun sensitivity was attenuated, moderate and statistically significant effects were observed for the tendency to develop a peeling sunburn followed by no tan or a light tan, or to burn before tanning, relative to immediate tanning.

We found no evidence (data not shown) that the effects of the pigmentary factors were confounded by sun-related variables, family history of melanoma, or by mole counts when additionally evaluated among the skin examination participants. In particular, we were concerned that freckles might be confounded by mole counts; however, the age- and gender-adjusted OR for freckles was essentially unchanged after additional adjustment for the number of benign and atypical moles (OR = 2.48 *vs.* 2.45, respectively).

In our data, 305 (94.3%) of cases and 364 (85.9%) of controls had clinical evidence of at least one benign mole. The mean (median) number of benign moles in those having at least one benign mole was 13.1 (9) in cases and 7.0 (4.5) in controls. A test of linear trend showed a significant relationship between the number of moles and melanoma risk ($p < 0.0001$). Based on the categories shown, we observed a moderately strong influence of benign mole counts on melanoma risk; however, for all levels of counts, the effects were substantially attenuated after adjustment for the number of atypical moles (Table II). Except for the highest category of benign mole counts, the ORs changed minimally after further adjustment for hair color, eye color, the 2 sun-related variables and family history of melanoma (Table II).

Our data indicated that 125 (38.7%) of cases and 59 (13.9%) of controls had clinical evidence of at least one atypical mole. The mean (median) number of atypical moles in those with at least 1 was 7.6 (3) in cases and 4.2 (2) in controls. A test of linear trend indicated an association of marginal statistical significance between the number of atypical moles and melanoma risk ($p = 0.07$). The age- and gender-adjusted OR for the presence of any atypical moles relative to none was strongly associated with melanoma risk; the effect was substantially reduced after adjustment for benign mole counts, but only minimally attenuated after further adjustment for the pigmentary factors, sun-related factors and family history of melanoma (Table II). Similarly, based on the categories shown, the age- and gender-adjusted ORs showed a moderately strong association between the number of atypical moles and melanoma risk, but the ORs were substantially diminished after adjustment for the number of benign moles. After further adjustment for hair color, eye color, freckles before age 15, the sun-related factors and family history of melanoma, the ORs corresponding to the higher counts of atypical moles were somewhat diminished; the moderate effect of 2–3 atypical moles was compatible with chance, while strong and significant effects were observed for ≥ 4 atypical moles.

We also evaluated the effects of having at least one atypical mole relative to none in a model that classified a study participant as positive for an atypical mole if any of the following 3 criteria were met: pathologic evidence of a previously excised atypical mole, an atypical mole found in histologic contiguity with the primary melanoma and/or clinical evidence of at least one atypical mole based on the study-related skin examination. This approach allowed us to extend our assessment to 792 subjects, including 362 (86%) cases and 430 (63%) controls for whom we had either skin examination findings or pathology records. Using these sources to classify atypical mole status, and after adjusting for age, gender and the number of benign moles, the OR pertaining to melanoma risk was 2.34 (95% CI = 1.67–3.28) for those with any atypical moles compared to none.

The distribution of benign and atypical moles in cases and controls according to age and gender is shown in Table III. Overall, based on the categories shown, benign mole counts were significantly associated with age ($p = 0.0001$, chi-square). In every age group, a larger proportion of cases than controls had high counts (≥ 15) of benign moles. In both the case and control groups, the proportion of persons affected by high mole counts was substantially greater in the youngest age group than in the oldest, but the age-related decrease was not entirely consistent. In the overall group, mean benign mole counts, assessed in subjects with at least

TABLE III – DISTRIBUTION AND MEAN COUNTS OF BENIGN AND ATYPICAL MOLES BY AGE AND GENDER IN CASES AND CONTROLS

Benign moles	Cases				Controls			
	0–4	5–14	15–24	≥ 25	Mean (SE)	0–4	5–14	15–24	≥ 25	Mean (SE)
Age										
≤ 39	12 (20.7%)	19 (32.8%)	13 (22.4%)	14 (24.1%)	17.2 (2.1)	25 (39.1%)	30 (46.9%)	5 (7.8%)	4 (6.3%)	8.5 (1.0)
40–49	16 (21.6%)	29 (39.2%)	18 (24.3%)	11 (14.9%)	15.1 (1.8)	42 (56.8%)	24 (32.4%)	5 (6.8%)	3 (4.1%)	7.7 (1.3)
50–59	31 (31.6%)	38 (38.8%)	19 (19.4%)	10 (10.2%)	13.2 (1.3)	79 (56.0%)	43 (30.5%)	15 (10.6%)	4 (2.8%)	7.4 (0.7)
≥ 60	47 (50.5%)	33 (35.5%)	8 (8.6%)	5 (5.4%)	8.6 (1.2)	96 (66.2%)	42 (29.0%)	5 (3.4%)	2 (1.4%)	5.5 (0.5)
Gender										
Male	55 (32.0%)	62 (36.0%)	36 (20.9%)	19 (11.0%)	13.1 (1.0)	141 (55.5%)	90 (35.4%)	16 (6.3%)	7 (2.8%)	7.1 (0.5)
Female	51 (33.8%)	57 (37.7%)	22 (14.6%)	21 (13.9%)	13.1 (1.2)	101 (59.4%)	49 (28.8%)	14 (8.2%)	6 (3.5%)	6.8 (0.6)
Atypical moles	None	1	2–3	≥ 4	Mean (SE)	None	1	2–3	≥ 4	Mean (SE)
Age										
≤ 39	25 (43.1%)	6 (10.3%)	10 (17.2%)	17 (29.3%)	7.5 (1.6)	47 (73.4%)	8 (12.5%)	4 (6.3%)	5 (7.8%)	4.1 (1.3)
40–49	39 (52.7%)	5 (6.8%)	10 (13.5%)	20 (27.0%)	10.3 (3.2)	64 (86.5%)	5 (6.8%)	3 (4.1%)	2 (2.7%)	5.5 (3.3)
50–59	63 (64.3%)	13 (13.3%)	7 (7.1%)	15 (15.3%)	6.3 (1.7)	118 (83.7%)	7 (5.0%)	11 (7.8%)	5 (3.5%)	3.7 (1.1)
≥ 60	71 (76.3%)	9 (9.7%)	4 (4.3%)	9 (9.7%)	5.7 (1.3)	136 (93.8%)	4 (2.8%)	0 (0.0%)	5 (3.4%)	4.3 (1.4)
Gender										
Male	103 (59.9%)	19 (11.0%)	13 (7.6%)	37 (21.5%)	8.4 (1.6)	216 (85.0%)	15 (5.9%)	9 (3.5%)	14 (5.5%)	4.7 (1.1)
Female	95 (62.9%)	14 (9.3%)	18 (11.9%)	24 (15.9%)	6.6 (1.6)	149 (87.6%)	9 (5.3%)	9 (5.3%)	3 (1.8%)	3.5 (1.1)

Distribution in percents calculated in rows. Mean counts are calculated in those with at least one mole of the type specified.

one benign mole, were significantly different across the age groups ($p < 0.0001$, ANOVA). In each age group, the mean counts of benign moles were higher for cases than controls. The mean counts declined consistently over age in both cases and controls, with a more striking decrease in cases. The overall distribution of benign moles was similar for men and women ($p = 0.58$, chi-square), and there were no obvious gender differences when the data were grouped by case and control status. Overall, the mean number of benign moles was also similar for men (9.7) and women (10.0; $p = 0.76$, *t*-test). Mean counts were higher in cases than in controls, but no gender differences were apparent within the case or control groups.

Overall, based on the categories shown, atypical mole counts were significantly associated with age ($p = 0.001$, chi-square). In cases and controls, the proportion affected by any atypical moles was greater in the youngest age group than in the oldest; the age-related decrease was consistent in cases, but not in controls. All categories of atypical mole counts also showed a higher affected proportion in the youngest *vs*. the oldest age group, but the age-related decrease was not uniform in either the case or control group. In cases and controls, as in the overall group, the mean number of atypical moles, assessed in those with at least one, was greatest for those of ages 40–49, but an overall test failed to support significant differences ($p = 0.28$, ANOVA). The proportion affected by any atypical moles was similar for men and women in both the case and control groups. Although a higher proportion of men than women had ≥ 4 atypical moles, the reverse was true for 2–3 atypical moles, and an overall test, based on the categories shown, did not support an association between gender and atypical mole counts ($p = 0.15$, chi-square). The tendency of cases to have higher mean counts of atypical moles was evident for both men and women. Overall, the mean number of atypical moles was slightly higher for men (7.1) than for women (5.8), a difference that was not significant ($p = 0.43$, *t*-test).

We explored the joint effects of age (< 40, 40–59, ≥ 60) and atypical moles (0, 1–2, ≥ 3) on melanoma risk. These analyses showed no consistent patterns, although confidence intervals were wide (data not shown). In addition, a test of interaction between atypical moles (present, absent) and age (< 50, ≥ 50) was not statistically significant ($p = 0.85$). Our data did not support an interaction between gender and the presence of at least one atypical mole or the number of atypical moles (0, 1–2, ≥ 3) in relation to melanoma ($p = 0.80$ and 0.87 for the respective interactions).

In our data, the mean (median) number of benign moles was 19.3 (15.0) in cases with at least one atypical mole and 8.0 (5.0) in cases who had no atypical moles. The mean (median) number of benign moles was 12.2 (11.0) in controls affected by atypical moles and 5.0 (3.0) in controls unaffected by atypical moles. Table IV shows the joint effects of the 2 mole types; risk of melanoma is substantially elevated for those with ≥ 3 atypical moles and 5–14 benign moles, and strikingly elevated for those with ≥ 15 benign moles and any atypical moles. Confidence intervals were wide, and the interaction between the number of benign and atypical moles was not statistically significant ($p = 0.21$).

We hypothesized that atypical moles might be markers of etiologic heterogeneity in melanoma. Thus, we used stratified analyses to explore possible differences in melanoma risk factors according to the presence or absence of atypical moles. These analyses, adjusted for age and gender, suggested somewhat lesser effects of pigmentary factors in the cases and controls affected by atypical moles than in those unaffected by atypical moles. For example, the OR for blue eye color compared to brown was 1.65 (95% CI = 0.74–3.67) in the subgroup of cases and controls affected by atypical moles and 2.39 (95% CI = 1.53–3.73) in the subgroup unaffected by atypical moles. In contrast, the sun-related variables appeared to have somewhat greater influence in the subgroup affected by atypical moles than in those unaffected by atypical moles. For example, the OR for the highest tercile of

TABLE IV – ODDS RATIOS AND 95% CONFIDENCE INTERVALS FOR THE JOINT EFFECTS OF BENIGN AND ATYPICAL MOLES ON MELANOMA RISK

	Number of benign moles, OR (95% CI)		
	< 5	5–14	≥ 15
Atypical moles			
None	1.00	1.84 (1.25–2.70)	3.09 (17.5–5.46)
1–2	3.86 (1.52–9.80)	2.40 (1.17–4.93)	11.88 (4.67–30.22)
≥ 3	1.99 (0.61–6.50)	7.25 (3.31–15.85)	12.58 (5.76–27.42)

OR adjusted for age and gender. $p = 0.21$ for the interaction between benign and atypical mole counts in relation to melanoma risk.

total recreational sun exposure relative to the lowest was 2.46 (95% CI = 1.05–5.75) in the subgroup with atypical moles and 1.51 (95% CI = 0.97–2.36) in those without atypical moles. None of the potential interactions, however, was statistically significant when formally tested using likelihood ratio tests.

Discussion

Substantial evidence indicates that atypical moles are the nonobligate pathologic precursors of melanoma.[25–27] Although the role of atypical moles in melanoma risk has been evaluated in epidemiologic studies,[11–16,19–22] few were conducted in the United States,[11,16,20] and only 2 previous studies[16,22] enrolled more than 125 melanoma cases. One previous population-based study, conducted in Sweden,[13] was limited to 121 melanoma cases, including prevalent diagnoses. A study conducted in England enrolled population-based cases and institution-based controls.[22]

Our intention was to accomplish a population-based study in the United States; however, participation in the skin examination component of our study was disappointing, particularly among the control subjects, introducing the possibility of bias. We were unable to assess directly whether self-selection for the skin examination distorted our counts of benign or atypical moles. However, with regard to the pigmentary risk factors, we found reasonably consistent results for skin examination participants *vs.* nonparticipants; the most discrepant findings were for blue eyes (compared to brown), with respective ORs of 2.11 and 1.56. Also, the age- and gender-adjusted effects of having any atypical moles were similar whether classified based on the skin examination or the combined source variable.

Our findings with regard to the percent of case and control participants affected by atypical moles are consistent with those noted in most previous reports. Our study and 2 previous studies conducted in the northeastern United States found that 39% of cases had either clinical[20] or histologic evidence[28] of atypical moles. Similar findings were noted in 2 studies conducted in Australia (34–37%),[12,21] a study in France (34%)[15] and a multicenter study in Germany (37%),[14] although a lower prevalence was noted in England (27%).[22] A higher prevalence (51–56%) was observed in a study conducted in Sweden,[13] one in Italy[19] and 2 studies conducted wholly[11] or in part[16] at a pigmented lesion clinic in San Francisco.

With regard to control subjects, we found that 14% were affected by atypical moles, consistent with the prevalence of 11–18% observed in 4 European studies,[13,14,19,29] including 2 population-based studies,[13,29] and in 3 U.S. studies,[11,16,20] including 1 with greater than 95% control participation.[11] A lower prevalence (6%) was found in England,[22] while a higher prevalence (21–22%) has been reported by a study in France[15] and another in Australia.[12] The counts of benign moles in our study are lower than those reported by most other groups.[12,13,16,19,20,22] We required a size of ≥ 3 mm and palpability, criteria imposed to reduce the likelihood of misclassifying freckles or solar lentigines as benign moles. While our practice may have underestimated the counts of benign moles, particularly nonpalpable junctional moles, it affected the case and control groups, so would not have biased our effect estimates.

To our knowledge, this study provides the first population-based report of the distribution of benign and atypical moles in melanoma cases and controls according to age and gender. In general, we found that age was associated with a decreasing proportion of individuals affected by high counts of benign or atypical moles. Age-related decreases have been reported previously for moles[12,21] as well as benign moles[16] and, specifically, atypical moles.[16,21] In our data, the mean number of benign moles decreased with increasing age, whereas the mean number of atypical moles appeared to be elevated in those of ages 40–49, particularly in cases.

We found little evidence of gender differences in the distribution of benign or atypical moles. The mean counts of benign moles were also similar for men and women, and counts of atypical moles were only slightly higher in men than in women. Previous studies also noted similar proportions of men and women affected by atypical moles[13,21] and similar gender-specific counts of moles,[12] benign moles[30] and atypical moles.[21] Consistent with other investigations, we found no evidence of an interaction between atypical moles and either age[16] or gender[16,22] in relation to melanoma risk.

In our analyses, as in previous reports,[11–14,16,19,22] the influence of benign and atypical mole counts diminished when adjusted for the other lesion type, although the effects of each remained moderately strong. We also found strong joint effects for the 2 lesion types, as has been reported previously.[22] Numerous studies, including 2 previous population-based studies conducted in Sweden,[30,31] have shown that the number of benign moles is strongly associated with the presence of clinically atypical moles. While it is possible that benign moles are in the causal pathway for the development of atypical moles and melanoma, the strong joint effects are consistent with the notion of a melanoma risk syndrome characterized by atypical moles and multiple benign moles.[32]

We were interested in evaluating the possibility that melanoma arising in the context of atypical moles might represent a variant form of this disease, a hypothesis suggested by the results of our previous studies.[23,28] Using stratified analyses, we attempted to identify disparate risk factors for melanoma arising in individuals affected by atypical moles as opposed to those unaffected by atypical moles. The stratum comparing cases and controls affected by atypical moles was of particular interest, as the analyses might elucidate factors associated with the malignant transformation of atypical moles to melanoma. The results of our stratified analyses implied that pigmentary characteristics were more important in the subgroup without atypical moles, and that sun-related variables were more influential in the subgroup with atypical moles, an observation consistent with speculation that sun exposure influences the transformation of atypical moles to melanoma. Our sample size was limited for these explorations, however, and the interactions were not statistically significant.

In summary, we conducted a population-based study of pigmentary factors and moles in relation to melanoma risk. Our findings indicated that these factors, which represent host char-

acteristics, are moderately to strongly related to melanoma risk. We found particularly strong effects for the combination of atypical moles and high counts of benign moles. Collectively, our results indicate that susceptibility, represented by a spectrum of markers including hair and eye color, sun sensitivity, tendency to freckle and the tendency to develop multiple benign or atypical moles, plays a key role in melanoma risk. Although the results of our stratified analyses were compatible with the notion that atypical moles distinguish a divergent melanoma pathway, the findings were inconclusive, and larger studies are needed to pursue this possibility.

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