



## Instrumental Measurements of Skin Color and Skin Ultraviolet Light Sensitivity and Risk of Cutaneous Malignant Melanoma: A Case-Control Study in an Italian Population

Alina V. Brenner<sup>1</sup>, Jay H. Lubin<sup>2</sup>, Donato Calista<sup>3</sup>, and Maria Teresa Landi<sup>4</sup>

<sup>1</sup>Radiation Epidemiology Branch, Division of Cancer Epidemiology and Genetics, National Cancer Institute, Bethesda, MD.

<sup>2</sup>Biostatistics Branch, Division of Cancer Epidemiology and Genetics, National Cancer Institute, Bethesda, MD.

<sup>3</sup>Department of Dermatology, Bufalini Hospital, Cesena, Italy.

<sup>4</sup>Genetic Epidemiology Branch, Division of Cancer Epidemiology and Genetics, National Cancer Institute, Bethesda, MD.

Received for publication December 3, 2001; accepted for publication April 17, 2002.

The authors evaluated objective measurements of constitutive skin color and ultraviolet light sensitivity in relation to risk of cutaneous malignant melanoma (CMM). Incident CMM cases ( $n = 183$ ) were diagnosed between December 1994 and January 1999 at the Maurizio Bufalini Hospital in Cesena, Italy. Controls ( $n = 179$ ) were mostly spouses/partners of cases and were frequency-matched by age and sex. In addition to interviews, constitutive skin color and skin ultraviolet light sensitivity were assessed by colorimetry and minimal erythema dose (MED), respectively. Odds ratios were estimated using unconditional logistic regression. The odds of CMM increased by a factor of 1.20 (95 percent confidence interval: 1.12, 1.30) for each unit of skin brightness and by a factor of 1.24 (95 percent confidence interval: 1.07, 1.43) per 10 mJ/cm<sup>2</sup> of MED. These associations were largely independent of phenotypic or sun-related characteristics and were modified by sun exposure. Increased risk of CMM was observed only among subjects with the highest levels of sun exposure. Epidemiologic studies of CMM may benefit from the inclusion of colorimetric and MED measurements along with traditional risk factors to obtain more accurate, quantitative, and objective information. *Am J Epidemiol* 2002;156:353–62.

case-control studies; colorimetry; epidemiologic methods; melanoma; risk factors; skin pigmentation; spectrophotometry, ultraviolet; ultraviolet rays

Abbreviations: CIELAB, Commission International d'Eclairage 1976 L\*a\*b\* [colorimetric system]; CMM, cutaneous malignant melanoma; MED, minimal erythema dose.

The incidence of and mortality from cutaneous malignant melanoma (CMM) has rapidly increased worldwide in recent decades. Although the main increase has been observed in populations in which fair skin predominates (1–3), there is evidence that the trend is similar in Mediterranean populations (4–6). Epidemiologic studies suggest that Mediterranean populations share similar risk factors for CMM with fair-skinned populations, but the distribution and relative weight of these factors may vary (7–12).

Well-established CMM risk factors include fair skin, red or blond hair, blue or green eyes, propensity to burn, inability to tan, and, to a lesser extent, both intermittent and cumulative sun exposure (13–17). Substantial evidence on the association of these risk factors with CMM comes from

case-control studies that used retrospective assessment of sun exposure and self-reported information on individual sensitivity to ultraviolet radiation (16). This approach might result in imprecise assessment of exposures and potentially differential reporting between cases and controls. Indeed, sun sensitivity, as assessed by ability to tan but not by hair color, was found to be subject to recall bias in one study (18). However, another study found that reporting of tanning and burning was less biased than reporting of sunbathing in childhood and adulthood, and possibly reporting of freckling in childhood (19). Accurate measurements of pigmentary traits and skin reaction to sun exposure are necessary to obtain more precise estimates of CMM risk and to adjust for other risk factors in analyses.

Reprint requests to Dr. Maria Teresa Landi, National Cancer Institute, 6120 Executive Blvd., MSC 7362, Bethesda, MD 20892-7362 (e-mail: landim@mail.nih.gov).

One instrument used for the objective measurement of constitutive skin color is the portable tristimulus reflectance colorimeter (20–23). Under standardized conditions, when distortion of instrument readings is minimal, this method shows high reproducibility (21) and correlation with measurements based on direct ascertainment of pigmentary chromophores (20, 24). The instrumental method of evaluating cutaneous sensitivity to ultraviolet radiation is based on measurement of the minimal erythema dose (MED)—the minimal dose of ultraviolet radiation able to provoke perceptible erythema of the skin 20–24 hours after exposure (25, 26). Although not a perfect measure itself (27, 28), MED provides a direct and quantitative measure of the skin's sensitivity to ultraviolet light. Constitutive skin color and skin sensitivity to ultraviolet radiation have been extensively studied in healthy volunteers or groups of patients with different skin diseases (25, 27, 29–32) and in a few well-defined epidemiologic studies (33–38). However, earlier studies did not use colorimeters or specifically analyze hue and chroma values.

We report here the results of a case-control study of melanoma conducted in Cesena, Italy. The main focus of this paper is on the association between melanoma and instrumental measurements of skin color and ultraviolet light sensitivity. In addition, we investigated the relation between instrumental and questionnaire-based data and the modifying effect of sun exposure on CMM risk associated with instrumental measurements.

## MATERIALS AND METHODS

### Study population

We recruited newly diagnosed CMM cases of any stage between December 1994 and January 1999 at the Dermatology Unit of Maurizio Bufalini Hospital in Cesena, Italy. This dermatology unit serves as a referral center for the regions of southern Emilia-Romagna and northern Marche and covers a population of nearly one million people. The patients referred to the clinic represented approximately 85 percent of all CMM patients in the entire area (39). The hospital's ethical committee approved the study design, and all participants signed an informed consent form. Cases were enrolled after surgical removal of the lesion and pathologic confirmation but prior to chemotherapy or radiation therapy. Controls were spouses or partners of the cancer cases ( $n = 134$ ), outpatients referred to the hospital for treatment of small accidental traumas ( $n = 14$ ), or healthy volunteers selected from Bufalini Hospital personnel ( $n = 31$ ). Controls were frequency-matched to cases (1:1 ratio) by decade of age (range, 17–77 years) and sex. All cases and controls were from the above-described catchment area. Among eligible subjects, approximately 95 percent of cases and 83 percent of controls agreed to participate in the study. After exclusion of five subjects (three cases and two controls) with a family history of melanoma, 183 cases (87 males and 96 females) and 179 controls (89 males and 90 females) were retained in the analysis.

### Data collection

*Interview and skin examination.* Trained interviewers who were blinded with respect to case/control status administered a questionnaire to all subjects. Lifetime residential history of at least 6 months' occupancy was recorded. The interviewers obtained information on sun exposure history (particularly between the hours of 11:00 a.m. and 3:00 p.m.), frequency and duration of sports and outdoor activities by age, and types of vacations taken. In addition, information on frequency and duration of occupational exposure to sunlight, use of ultraviolet sunlamps and sunscreens, and history of sunburns was recorded. For assessment of sunlight sensitivity, subjects were asked how their skin would respond to an initial 30 minutes of direct strong sunlight and how it would respond after repeated and prolonged exposure to sunlight. Information on sociodemographic characteristics, medical and family history of cancer and other diseases, smoking history, alcohol consumption, and diet was collected.

In addition, one dermatologist performed skin examinations of the entire body, except the genital area, for all subjects. Skin color was evaluated on the inner forearm and characterized on a three-category scale as dark/olive, medium, or light; eye color was characterized on a nine-category scale as black, dark brown, light brown, brown-green, green, blue-green, dark blue, light blue, or gray; and natural hair color was characterized on a six-category scale as black, dark brown, light brown, reddish-brown, blond, or red (40). Multiple lightly pigmented macular lesions commonly present on the face, upper back, and arms were defined as freckles. Diagnosis of dysplastic nevus and number of nevi were based on a review of photographs of the subject's back by an oncologist who was blinded as to case/control status (40). To be counted as a dysplastic nevus, a nevus had to be 5 mm or greater in diameter, be predominantly flat, and have at least two of the following criteria: variable pigmentation, indistinct borders, and an irregular outline (41). Nevi were defined as pigmented macules or papules greater than 2 mm in diameter that did not include freckles, lentigines, keratoses, and other pigmented lesions.

*Measurement of skin color.* Skin color was measured on the buttock with a Minolta CR-300 colorimeter (Minolta Company Ltd., Osaka, Japan). This instrument is a reflectance spectrophotometer that measures reflected light in the visible spectrum (range, 400–700 nm). It also works as a tristimulus chromameter, recording colors in a three-dimensional space known as Commission International d'Éclairage 1976  $L^*a^*b^*$  (CIELAB) color space (20, 21). Every color in the CIELAB colorimetric system can be described by a combination of three coordinates,  $L^*$ ,  $a^*$ , and  $b^*$  (42, 43), where  $L^*$  is the total quantity of light reflected or skin brightness (described as light, dark, etc.),  $a^*$  represents color ranging from red (positive values) to green (negative values), and  $b^*$  represents color ranging from blue (negative values) to yellow (positive values). The  $a^*$  and  $b^*$  coordinates can be converted into polar coordinates (21, 43) defined as hue angle ( $h^\circ = \arctan(b^*/a^*)$ ) and chroma, often referred to as saturation of color ( $C = [(a^*)^2 + (b^*)^2]^{1/2}$ ). Hue refers to the basic color

**TABLE 1. Correlations† between instrumental measurements of skin color and skin sensitivity among controls in a case-control study of cutaneous malignant melanoma, Cesena, Italy, 1994–1999**

Measurement	L* (brightness of the skin)	Hue (°)	Chroma (saturation of color)	Minimal erythema dose (mJ/cm <sup>2</sup> )
L* (brightness of the skin)	1.0	0.592‡	-0.570‡	-0.423‡
Hue (°)		1.0	-0.035	-0.247‡
Chroma (saturation of color)			1.0	0.342‡
Minimal erythema dose (mJ/cm <sup>2</sup> )				1.0

† Spearman correlation coefficient.

‡  $p \leq 0.001$ .

of an object, where 0° represents red and 90° represents yellow. The hue angle in our population ranged from 43° to 82°. Chroma describes the intensity of color, with higher chroma indicating greater intensity.

The instrument was white-calibrated and turned on at least 15 minutes before the start of each set of measurements. Measurements were taken by a single dermatologist after 15 minutes' acclimatization in an air-conditioned room at 19–20°C. Prior to measurement, the subject was placed in a horizontal position to avoid orthostatic effects' having an influence on skin color. During measurement, the measuring head was held steady, perpendicular to the skin surface. Special care was taken not to apply excessive pressure on the head of the instrument to avoid venous congestion that could artificially distort the measurements (21). Measurements of L\*, a\*, and b\* were repeated three times consecutively and averaged. The a\* and b\* values were then reexpressed as hue and chroma values.

**Measurement of MED.** The sensitivity of skin to ultraviolet radiation was assessed by measurement of MED. Skin was irradiated with simulated solar radiation from an artificial ultraviolet light source (the IL1700 radiometer, Blue-Point model; International Light, Inc., Newburyport, Massachusetts) and measured by an International Light detector (SED240/SCS280/W, head model; International Light, Inc.) equipped with a cosine response diffuser. A spectroradiometer (SpectraMED model; Flyby s.r.l., Livorno, Italy) was used to characterize the spectral features of the ultraviolet lamp radiation, which extended from 290 nm to 440 nm (covering the entire solar spectrum of ultraviolet light reaching the earth's surface), and to evaluate the erythema effective irradiance at the surface of the skin, which was 1.65 mW/cm<sup>2</sup>. Depending on the subject's constitutive skin color, a suberythema dose of ultraviolet light (on the order of 10–20 mJ/cm<sup>2</sup>) and additional incremental doses comparable with biologic exposures to sunlight (44) were administered by a single nurse on a very small area of buttock skin. Each area of irradiation was circled with a fine-point waterproof permanent black marker. For skin types very sensitive to ultraviolet radiation, the dose ranged between 15 mJ/cm<sup>2</sup> and 67 mJ/cm<sup>2</sup>, while for individuals with darker skin, the dose ranged between 21 mJ/cm<sup>2</sup> and 108 mJ/cm<sup>2</sup>. Incremental doses of ultraviolet radiation were as follows: 15/21, 27, 40, 54, 67, and 94/108 mJ/cm<sup>2</sup>. Subjects returned to the hospital the following day so that the marked skin areas could be

checked. The MED was defined as the lowest dose of radiation that produced the minimum noticeable (by the nurse's visual inspection) reddening of the skin 24 hours after exposure. This approach has been used and validated multiple times in subjects from the same area.

### Statistical analysis

Nonparametric correlation analysis and one-way analysis of variance were performed to explore the relation between instrumental measurements of skin color and skin sensitivity and other characteristics among control subjects. The odds ratio was used as the measure of association between potential risk factors and CMM. Unconditional logistic regression models (45) were fitted to estimate odds ratios and compute 95 percent confidence intervals. Likelihood ratio tests (two-sided tests with an  $\alpha$  level of 0.05) were used to test hypotheses. All analyses were adjusted for matching factors (age at interview and sex). In addition, potentially confounding factors were taken into account where specified. We repeated all analyses excluding the hospital volunteers or including spouses/partners of the cases as the only control group, and we did not find major differences. This paper reports results obtained using the entire control group.

The associations between CMM and L\*, hue, chroma, and MED are also presented in terms of the exponential increase in the odds ratio per unit of measurement. These estimates were derived from the logistic regression model using the continuous measurement variables. Because of the larger range of values for the MED measurement, we present the exponential increase per 10 mJ/cm<sup>2</sup>.

### RESULTS

L\* was highly correlated with both hue and chroma, while hue and chroma were uncorrelated (table 1). The correlation coefficients for MED and the other instrumental measurements were significant but were smaller than coefficients for correlations between L\* and hue and L\* and chroma.

Table 2 summarizes the distribution of instrumental measurements among controls, by selected characteristics. Except for skin color, L\*, hue, chroma, and MED were generally unrelated to age, sex, and host characteristics such as eye color, hair color, freckling, presence of dysplastic nevi, and number of nevi. In contrast, the instrumental

**TABLE 2. Descriptive statistics for instrumental measurements of skin color and skin sensitivity, by selected characteristics, among controls in a case-control study of cutaneous malignant melanoma, Cesena, Italy, 1994–1999**

Characteristic	No. ( <i>n</i> = 179)†	L* (brightness of the skin)		Hue (°)		Chroma (saturation of color)		Minimal erythema dose (mJ/cm <sup>2</sup> )	
		Mean	<i>p</i> value‡	Mean	<i>p</i> value	Mean	<i>p</i> value	Mean	<i>p</i> value
Age (years) at interview									
<35	41	70.2	0.884	70.1	0.337	17.6	0.069	41.0	0.423
35–44	47	71.4		71.7		16.7		35.5	
45–54	37	70.8		69.6		17.1		36.9	
≥55	46	70.6		69.6		16.4		42.9	
Sex									
Female	89	70.9	0.737	71.5	0.033	16.8	0.417	38.5	0.536
Male	87	70.8		69.2		17.0		39.2	
Eye color§									
Low risk	75	70.6	0.743	70.7	0.398	17.2	0.101	39.6	0.234
Medium risk	83	71.1		70.2		16.8		39.8	
High risk	17	70.5		69.4		15.9		32.1	
Skin color¶									
Light	55	72.4	<0.001	71.5	0.099	16.0	<0.001	35.9	0.025
Medium	92	70.8		69.9		16.8		38.8	
Dark/olive	29	68.1		69.5		19.0		44.6	
Natural hair color									
Black	24	69.6	0.187	70.5	0.921	17.4	0.237	46.0	0.052
Dark brown	101	70.9		70.3		17.0		38.6	
Light brown	35	71.6		70.7		16.2		35.9	
Blond	11	71.4		70.6		16.5		35.4	
Reddish-brown	5	69.1		68.0		18.0		37.7	
Current freckling									
No	94	70.5	0.236	70.5	0.664	17.2	0.120	39.7	0.307
Yes	80	71.2		70.1		16.5		37.3	
Dysplastic nevi									
No	122	70.5	0.069	70.1	0.357	16.9	0.814	39.9	0.443
Yes	31	71.8		71.2		17.1		37.4	
No. of nevi									
0–19	62	70.9	0.900	71.0	0.274	16.7	0.745	36.9	0.407
20–29	28	70.8		70.2		16.6		42.1	
30–44	31	71.1		71.3		16.6		39.0	
45–69	29	70.5		68.8		17.1		42.1	
≥70	17	71.0		70.1		17.9		37.9	
Skin reaction to acute sun exposure									
Tan without burn	60	69.5	<0.001	69.2	0.096	17.6	<0.001	41.4	0.078
Light or medium burn	90	71.3		70.9		16.8		38.3	
Severe burn or blistering	26	72.3		71.1		15.6		34.9	
Skin reaction to chronic sun exposure									
Deep tan	61	69.4	<0.001	70.1	0.431	18.2	<0.001	42.4	0.053
Moderate tan	85	71.4		70.2		16.5		37.6	
Mild tan or no tan	25	72.4		71.4		15.0		35.9	
Lifetime recreational sun exposure# (hours)									
0	41	72.2	0.001	70.6	0.344	15.8	0.003	39.2	0.445
1–760	30	70.9		70.2		17.1		36.5	
761–2,399	42	71.2		71.8		16.6		38.0	
≥2,400	52	69.7		69.0		17.6		41.4	
Lifetime occupational sun exposure (hours)									
0	98	70.7	0.934	71.0	0.006	17.1	0.085	38.9	0.995
1–10,999	36	71.3		71.2		17.4		39.5	
≥11,000	39	70.5		67.6		16.0		38.8	

† The total number of subjects may vary across categories because of missing values.

‡ Kruskal-Wallis test of score differences.

§ Dark: black or dark brown. Medium: light brown, brown-green, green, or blue-green. Light: light blue, dark blue, or gray.

¶ As determined by a dermatologist on the inner forearm.

# Hours of lifetime recreational sun exposure between 11:00 a.m. and 3:00 p.m.

**TABLE 3. Odds ratios for relations between cutaneous malignant melanoma and instrumental measurements of skin color and skin sensitivity in a case-control study, Cesena, Italy, 1994–1999**

Measurement	Cases (n = 183)†	Controls (n = 179)†	Odds ratio‡	95% confidence interval	Odds ratio	95% confidence interval
L* (brightness of the skin)						
≤70.0	30	54	1.00§		1.20¶	1.12, 1.30
>70.0–≤72.3	37	52	1.30	0.70, 2.41		
>72.3–≤74.0	52	40	2.19	1.18, 4.08		
>74	64	30	4.35	2.29, 8.27		
Hue (°)						
≤68	36	53	1.00§		1.03¶	0.99, 1.07
>68–≤71.3	51	37	2.10	1.13, 3.87		
>71.3–≤75	52	45	1.63	0.90, 2.95		
>75	44	41	1.59	0.86, 2.94		
Chroma (saturation of color)						
≤14.7	51	38	1.86	1.01, 3.24	1.16¶	1.06, 1.26
>14.7–≤16.2	52	40	1.81	0.99, 3.31		
>16.2–≤17.8	42	46	1.25	0.68, 2.30		
>17.8	38	52	1.00§			
Minimal erythema dose (mJ/cm <sup>2</sup> )						
≤26	45	36	2.11	1.14, 3.91	1.24#	1.07, 1.43
>26–≤33	47	42	1.90	1.04, 3.46		
>33–≤40	51	39	2.22	1.22, 4.03		
>40	38	59	1.00§			

† Numbers may not add up to column totals because of missing data.

‡ Adjusted for age at interview and sex.

§ Referent group.

¶ Odds ratio per unit of change in instrumental measurement (increasing for L\* and hue and decreasing for chroma and minimal erythema dose) based on a logistic regression model using continuous data.

# Odds ratio per 10 mJ/cm<sup>2</sup> of decreasing change in minimal erythema dose based on a logistic regression model using continuous data.

measurements differed markedly by skin color. As expected, higher values of L\* and hue and lower values of chroma and MED were associated with light skin color. Measurements did not vary substantially by season of evaluation (data not shown).

Subjects were queried about their skin's reaction to acute and chronic sun exposure. Higher measured values for L\* and hue and lower measured values for chroma and MED were associated with a self-reported propensity to severely burn or blister and an inability to tan, although for hue and MED, *p* values did not reach the traditional level of statistical significance. Finally, we estimated lifetime sun exposure from recreational and occupational activities (40). Among controls, the associations of instrumental measurements and estimated sun exposure were less consistent. L\* and chroma were related to recreational sun exposure, but hue and MED were not. Only hue showed a significant relation with occupational sun exposure.

Table 3 shows odds ratios for CMM associated with instrumental measurements (both categorical and continuous) of skin color and skin sensitivity after adjustment for matching factors (age at interview and sex). Odds ratios for

CMM increased significantly with increasing levels of L\* and decreasing levels of chroma and MED, although the latter trend was not monotonic. Odds ratios for hue angle were elevated but not significant. Prior analysis demonstrated that in this population there was no association between CMM risk and lifetime hours of sun exposure between 11:00 a.m. and 3:00 p.m. from either recreational activity or occupational activity (40). In addition, inclusion of sun exposure covariates in the model did not change risk estimates for any of the instrumental measurements (data not shown).

Table 4 summarizes the effect of adjustment of instrumental measurements of skin color and skin sensitivity on CMM risk. The association between L\* and CMM persisted after adjustment for the other instrumental measurements, while the associations for hue and chroma disappeared after adjustment for L\*. The odds ratio pattern with MED was unchanged after adjustment for hue and chroma, but it was attenuated after adjustment for L\*.

Table 5 shows CMM risk estimates for L\* and MED after adjustment for phenotypic characteristics (eye color, natural hair color, skin color, current freckling, and dysplastic nevi)

**TABLE 4. Effect of mutual adjustment for instrumental measurements of skin color and skin sensitivity on risk of cutaneous malignant melanoma in a case-control study, Cesena, Italy, 1994–1999**

Adjustment factors	L* (brightness of the skin)		Hue (°)		Chroma (saturation of color)		Minimal erythema dose (mJ/cm <sup>2</sup> )	
	OR†,‡	95% CI†	OR	95% CI	OR	95% CI	OR	95% CI
Age and sex	1.20	1.12, 1.30	1.03	0.99, 1.07	1.16	1.06, 1.26	1.24	1.07, 1.43
Age, sex, and hue	1.25	1.14, 1.37			1.16	1.06, 1.26	1.22	1.05, 1.41
Age, sex, and chroma	1.20	1.10, 1.32	1.03	0.99, 1.07			1.16	1.00, 1.36
Age, sex, and minimal erythema dose	1.20	1.10, 1.30	1.03	0.99, 1.07	1.12	1.02, 1.23		
Age, sex, and L*			0.96	0.92, 1.01	1.00	0.89, 1.12	1.06	0.90, 1.25

† OR, odds ratio; CI, confidence interval.

‡ Odds ratio per unit of change in instrumental measurement (per 10 mJ/cm<sup>2</sup> for minimal erythema dose) based on a logistic regression model using continuous data.

and sun-related characteristics (skin reaction to acute or chronic sun exposure) known to be associated with melanoma risk in this population (40). Generally, the adjustment did not change the odds ratio pattern. The risk estimates associated with skin color and L\* or MED, while slightly attenuated, persisted when both variables were included in the same model. Adjustment for the independent predictors (eye color, skin color, dysplastic nevi, and skin reaction to chronic sun exposure) in addition to age resulted in the most pronounced attenuation of CMM risk estimates for L\* and MED. There were no significant differences in the odds ratio patterns for L\*, hue, or chroma by sex. The increase in CMM risk per 10 mJ/cm<sup>2</sup> of MED was 1.12 (95 percent confidence interval: 0.93, 1.34) in females and 1.43 (95 percent confidence interval: 1.12, 1.83) in males after adjustment for age at interview, but this difference could have been due to chance (*p* for homogeneity = 0.10).

Table 6 summarizes the effects of L\* and MED within categories of lifetime sun exposure. Among subjects in the lowest tertile of sun exposure, there was little evidence of an increased CMM risk with either L\* or MED. Among subjects in the highest two tertiles of total sun exposure, risk of CMM increased with L\* and decreased with MED, although the result of the homogeneity test for the former was not statistically significant.

## DISCUSSION

Among instrumental measurements of skin color and ultraviolet radiation sensitivity, brightness of the skin as determined by L\* value and, to a lesser extent, MED were the strongest predictors of CMM risk in this study. Furthermore, these effects largely persisted after adjustment for phenotypic or sun-related factors. L\* and MED were associ-

**TABLE 5. Effect of adjustment for phenotypic and sun-related characteristics on risk of cutaneous malignant melanoma associated with instrumental measurements of skin color and skin sensitivity in a case-control study, Cesena, Italy, 1994–1999**

Adjustment factors	L* (brightness of the skin)		Minimal erythema dose (mJ/cm <sup>2</sup> )	
	OR†,‡	95% CI†	OR§	95% CI
Age and sex	1.20	1.12, 1.30	1.23	1.07, 1.43
Age and eye color	1.20	1.11, 1.29	1.23	1.06, 1.42
Age and natural hair color	1.19	1.10, 1.29	1.19	1.03, 1.38
Age and skin color	1.14	1.05, 1.23	1.16	1.00, 1.35
Age and current freckles	1.20	1.11, 1.30	1.23	1.06, 1.43
Age and dysplastic nevi	1.18	1.09, 1.27	1.19	1.02, 1.38
Age and skin reaction to acute sun exposure	1.18	1.10, 1.28	1.20	1.04, 1.39
Age and skin reaction to chronic sun exposure	1.17	1.08, 1.27	1.22	1.04, 1.42
Age, eye color, skin color, dysplastic nevi, and skin reaction to chronic sun exposure¶	1.12	1.02, 1.22	1.14	0.97, 1.34

† OR, odds ratio; CI, confidence interval.

‡ Odds ratio per unit of change in L\* based on a logistic regression model using continuous data.

§ Odds ratio per 10 mJ/cm<sup>2</sup> based on a logistic regression model using continuous data.

¶ Factors found to be independent predictors by Landi et al. (Br J Cancer 2001;85:1304–10).

**TABLE 6. Modifying effect of lifetime sun exposure on the relations between skin brightness and minimal erythema dose and risk of cutaneous malignant melanoma in a case-control study, Cesena, Italy, 1994–1999**

Measurement	Lifetime sun exposure† (hours)						<i>p</i> for interaction
	0–1,100		1,101–6,000		≥6,001		
	OR‡,§	95% CI‡	OR	95% CI	OR	95% CI	
<b>L* (brightness of the skin)</b>							
≤70.0	1.00¶		1.00¶		1.00¶		
>70.0–≤72.3	0.38	0.10, 1.51	2.05	0.69, 6.07	2.02	0.64, 6.40	
>72.3–≤74.0	0.73	0.22, 2.46	4.26	1.36, 13.30	3.37	0.99, 11.42	
>74	1.45	0.45, 4.64	6.58	1.71, 25.37	6.69	1.94, 23.02	
OR per unit of L*	1.08	0.95, 1.22	1.33	1.13, 1.57	1.24	1.08, 1.44	0.13
<b>Minimal erythema dose (mJ/cm<sup>2</sup>)</b>							
≤26	0.39	0.12, 1.25	6.31	1.92, 20.72	6.39	1.69, 24.17	
>26–≤33	0.64	0.19, 2.17	3.29	1.03, 10.5	3.83	1.14, 12.89	
>33–≤40	0.48	0.15, 1.57	2.86	0.95, 8.63	8.34	2.27, 30.65	
>40	1.00¶		1.00¶		1.00¶		
OR per 10 mJ/cm <sup>2</sup>	0.87	0.65, 1.17	1.49	1.13, 1.96	1.44	1.06, 1.95	0.02

† Lifetime sun exposure was computed as the sum of hours of sun exposure due to recreational exposure and occupational exposure.

‡ OR, odds ratio; CI, confidence interval.

§ Adjusted for age at interview and sex.

¶ Referent group.

ated with increased risk of CMM primarily among subjects with the highest levels of sun exposure and were not associated with CMM risk among subjects with the lowest level of sun exposure.

Traditionally, epidemiologic studies have relied either on self-assessment of skin color or its assessment by an observer against referent color standards. In both cases, the resulting color is a subjective perception that often fails to distinguish between the pigmented substances in the skin and the actual sensation of color. In an effort to improve on these subjective measures, some researchers have attempted to measure melanin content objectively by studying skin color reflectance after irradiation with wavelengths similar to those of ultraviolet light (35, 37, 38). The present study used a portable tristimulus reflectance spectrophotometer that assesses skin color and its main components, brightness, hue, and chroma, in a manner analogous to the human eye (20, 21, 23, 43). While such measurements are objective and more accurate, they still do not provide direct information about the primary chromophore responsible for color (23).

L\* measures the brightness component of color, and it varies on an achromatic gray scale between a value of 100 for a white surface and 0 for a black surface. Previous studies have shown that L\* is negatively correlated with the amount of epidermal melanin, showing an approximately linear relation at low melanin levels (20, 46). Colorimetric measurements appear to be affected by the amount of hemoglobin in the superficial vascular plexus (20, 46) and skin surface struc-

ture (21, 23). In addition, a high serum  $\beta$ -carotene level may affect skin color (47) and consequently colorimetric estimate. Although our study included subjects of Italian origin, L\* measurements taken on the buttock met criteria for low pigmentation (L\* > 60) (46). As might be expected on the basis of the relation between L\* and amount of melanin, higher L\* levels were associated with lighter skin, increased sensitivity to burning, and reduced tanning ability, as well as lower actual sun exposure from recreational activities. This latter finding suggests active sun avoidance by subjects with lighter skin. Similarly to other investigators (48), we did not see variation in L\* values by eye color or hair color. CMM risk associated with L\* was consistently increased in all of the analyses and persisted after adjustment for a variety of potential confounders. Interestingly, when skin color and L\* were included in the same model, CMM risk estimates for both factors, while attenuated slightly, were not materially changed. This might imply that skin color and L\* carry additional information not accounted for by the other factor.

The chroma component of color or saturation, which is visually perceived as “weak” or “strong,” does not have a clear physiologic determinant. We observed a strong correlation between L\* and chroma, while b\* and chroma were correlated by construction. Allowing that b\* value is a good indicator of tanning (24, 49) and that our findings show similar variation in L\* and chroma by phenotypic and sun-related characteristics among controls, it appears that L\* and chroma are both related to skin pigmentation. In univariate

analyses, chroma was associated with increased risk of CMM. However, after we included  $L^*$  and chroma in a single model, the effect of chroma disappeared while the effect of  $L^*$  remained unchanged. This pattern suggests that in our data, either  $L^*$  is the main risk factor for melanoma or both factors are related to CMM risk but chroma is not an independent predictor of risk.

The hue component of color, another chromaticity coordinate, was positively correlated with  $L^*$ . However, unlike  $L^*$  or chroma, it did not vary significantly by skin color or skin reaction to sun exposure and was not associated with CMM risk. This implies that hue angle is not a direct indicator of skin pigmentation or other skin characteristics associated with CMM risk.

MED is a well-accepted and often preferred parameter for evaluation of a subject's sensitivity to phototherapy or solar radiation (26, 28, 50–53). However, it has several limitations (27). Since MED determination is made by humans, it may be subjective, and interobserver variability could take place. In addition, MED values are limited to the specific categories of ultraviolet doses used, and uncontrolled unknown factors might influence the degree of individual vascular dilation. Moreover, it requires that subjects make two clinic visits, one for irradiation and the second for evaluation of erythema after 24 hours. This may affect participation rates and the complexity of field activities. Although we cannot exclude the effect of uncontrolled factors, it is unlikely that MED measurements in this study were subject to systematic error, since a single trained nurse who was blinded with respect to the subject's status performed the readings.

Our study showed a negative association between MED and  $L^*$ , which is consistent with most other studies (25, 54–58). It is possible that melanin, because of its absorption spectrum, influences both primary defense against ultraviolet radiation and skin color (57). Indeed, MED significantly varied by category of constitutive skin color and, to a lesser extent, by reaction to acute and chronic sun exposure in our study. This may reflect the subjectivity of self-reported skin reaction to sunlight, which may not be a reliable measure of ultraviolet light sensitivity. Other studies using alternative methods to measure skin reaction to ultraviolet radiation—including slope of the erythral dose-response curve (29, 58, 59), ratio between facultative and constitutional skin color (58–60), colorimetric quantification of erythral response after irradiation (60), and measurement of minimal immediate pigment darkening dose and minimal delayed tanning dose (58)—have reported variable associations with skin type. In our study, MED was associated with increased risk of CMM; this finding is in agreement with one study that found higher light sensitivity among melanoma patients (61) but not with other studies (55, 62). However, none of the previous studies conducted risk estimation or adjustment for potential confounders. In this study, after adjustment for hue and chroma but not for  $L^*$ , risk estimates associated with MED remained elevated, providing supporting evidence that risk depends mainly on melanin pigmentation, with other characteristics of the skin, such as epidermal thickness, possibly playing a role.

The role of sun exposure in the etiology of CMM is unresolved (14, 17). It was not associated with increased risk of

CMM in our study (40), even after we controlled for the presence of dysplastic nevi and pigmentation characteristics. Since most control subjects were spouses/partners of the cases, these groups had similar levels of adult sun exposure. In addition, controls lived in the same geographic areas as cases, most for their entire lives. Thus, differences in recreational sun exposure between cases and controls were not large. However, sun exposure could still be an important modifier of the effect of potential sensitivity to the sun on melanoma risk. We did not find any interaction between subjective measurements of pigmentation characteristics and sun exposure in the association with melanoma risk, but we observed an increased risk with  $L^*$  and MED only among subjects with a high number of cumulative hours of sun exposure. Similar patterns were observed when recreational and occupational sun exposures were analyzed separately; however, the statistical power of these tests was lower. It is unlikely that our findings could be attributable to greater misclassification of  $L^*$  among subjects with low total sun exposure, since results were similar for  $L^*$  and MED and measurements were made on the buttock, an area that is not affected by sun exposure. In addition, these findings are in agreement with those of other case-control studies (63–68) that measured sun exposure and sun sensitivity in a number of different ways. This suggests that skin sensitivity in the presence of sun exposure is an important risk factor for CMM and that reduction of sun exposure could be especially beneficial for ultraviolet-sensitive persons. Finally, this finding further supports the use of instrumental measurements in epidemiologic studies. Use of these instruments can help researchers to identify those elements of the pigmentary trait that relate to CMM risk. In addition, by adjusting for the risk due to pigmentation phenotype, investigators can more fully evaluate associations with other potential risk factors for melanoma, such as sun exposure, sunscreen use, and genetic determinants.

In our study, skin examination was performed by a single dermatologist. Participation rates were high for both cases and controls. Detailed information on a large number of risk factors was collected and analyzed. A possible weakness of the study was the heterogeneous nature of the control series, which included spouses/partners of cases, hospital controls, and healthy volunteers. However, limiting the analyses to spouses/partners did not materially influence our findings. While the measurements of skin color and sun sensitivity were within the range of those reported in studies conducted among populations of other European origins, generalization of these findings to fair-skinned populations should be cautious. Finally, the limited statistical power of these findings, especially for certain subgroup analyses, should be noted.

In summary, brightness of constitutive skin color and MED in the presence of sun exposure emerged as important risk factors for CMM in a case-control study conducted in an Italian population. This association was largely independent of other phenotypic and sun-related characteristics. Epidemiologic studies of melanoma and other skin diseases may benefit from the inclusion of instrumental measurements along with traditionally assessed risk factors to obtain more accurate, quantitative, and objective information.

## ACKNOWLEDGMENTS

This study was supported by a grant (CA 65558-01A2) to Dr. M. T. Landi from the National Institutes of Health.

The authors are indebted to Dr. G. Landi for overseeing the study procedures and to Angela Aguzzoni for MED assessment. The authors are grateful to Paola Minghetti for organizational and technical support.

## REFERENCES

- Jemal A, Devesa SS, Fears TR, et al. Cancer surveillance series: changing patterns of cutaneous malignant melanoma mortality rates among whites in the United States. *J Natl Cancer Inst* 2000;92:811-18.
- Dennis LK. Analysis of the melanoma epidemic, both apparent and real: data from the 1973 through 1994 Surveillance, Epidemiology, and End Results Program registry. *Arch Dermatol* 1999;135:275-80.
- Serraino D, Fratino L, Gianni W, et al. Epidemiological aspects of cutaneous malignant melanoma (review). *Oncol Rep* 1998;5:905-9.
- Muir CS, Waterhouse J, Mack T, et al. Cancer incidence in five continents. Vol 5. (IARC Scientific Publication no. 88). Lyon, France: International Agency for Research on Cancer, 1987.
- Parkin DM, Muir CS, Whelan SL, et al. Cancer incidence in five continents. Vol 6. (IARC Scientific Publication no. 120). Lyon, France: International Agency for Research on Cancer, 1992.
- Parkin DM, Whelan SL, Ferlay J, et al. Cancer incidence in five continents. Vol 7. (IARC Scientific Publication no. 143). Lyon, France: International Agency for Research on Cancer, 1997.
- Cristofolini M, Franceschi S, Tassin L, et al. Risk factors for cutaneous malignant melanoma in a northern Italian population. *Int J Cancer* 1987;39:150-4.
- Zanetti R, Rosso S, Colonna S, et al. Case-control study on malignant skin melanoma in the Turin province. (In Italian). *G Ital Dermatol Venereol* 1988;123:461-8.
- Carli P, Biggeri A, Giannotti B. Malignant melanoma in Italy: risks associated with common and clinically atypical melanocytic nevi. *J Am Acad Dermatol* 1995;32:734-9.
- Rodenas JM, Delgado-Rodriguez M, Herranz MT, et al. Sun exposure, pigmentary traits, and risk of cutaneous malignant melanoma: a case-control study in a Mediterranean population. *Cancer Causes Control* 1996;7:275-83.
- Espinosa Arranz J, Sanchez Hernandez JJ, Bravo Fernandez P, et al. Cutaneous malignant melanoma and sun exposure in Spain. *Melanoma Res* 1999;9:199-205.
- Naldi L, Lorenzo IG, Parazzini F, et al. Pigmentary traits, modalities of sun reaction, history of sunburns, and melanocytic nevi as risk factors for cutaneous malignant melanoma in the Italian population: results of a collaborative case-control study. *Cancer* 2000;88:2703-10.
- Elwood JM, Jopson J. Melanoma and sun exposure: an overview of published studies. *Int J Cancer* 1997;73:198-203.
- Katsambas A, Nicolaidou E. Cutaneous malignant melanoma and sun exposure: recent developments in epidemiology. *Arch Dermatol* 1996;132:444-50.
- Franceschi S, La Vecchia C, Lucchini F, et al. The epidemiology of cutaneous malignant melanoma: aetiology and European data. *Eur J Cancer Prev* 1991;1:9-22.
- Evans RD, Kopf AW, Lew RA, et al. Risk factors for the development of malignant melanoma. I. Review of case-control studies. *J Dermatol Surg Oncol* 1988;14:393-408.
- Armstrong BK. Epidemiology of malignant melanoma: intermittent or total accumulated exposure to the sun? *J Dermatol Surg Oncol* 1988;14:835-49.
- Weinstock MA, Colditz GA, Willett WC, et al. Recall (report) bias and reliability in the retrospective assessment of melanoma risk. *Am J Epidemiol* 1991;133:240-5.
- Cockburn M, Hamilton A, Mack T. Recall bias in self-reported melanoma risk factors. *Am J Epidemiol* 2001;153:1021-6.
- Takiwaki H. Measurement of skin color: practical application and theoretical considerations. *J Med Invest* 1998;44:121-6.
- Fullerton A, Fischer T, Lahti A, et al. Guidelines for measurement of skin colour and erythema. A report from the Standardization Group of the European Society of Contact Dermatitis. *Contact Dermatitis* 1996;35:1-10.
- Andreassi L, Flori L. Practical applications of cutaneous colorimetry. *Clin Dermatol* 1995;13:369-73.
- Kollias N. The physical basis of skin color and its evaluation. *Clin Dermatol* 1995;13:361-7.
- Takiwaki H, Overgaard L, Serup J. Comparison of narrow-band reflectance spectrophotometric and tristimulus colorimetric measurements of skin color. Twenty-three anatomical sites evaluated by the Dermaspectrometer and the Chroma Meter CR-200. *Skin Pharmacol* 1994;7:217-25.
- Damian DL, Halliday GM, Barnetson RS. Prediction of minimal erythema dose with a reflectance melanin meter. *Br J Dermatol* 1997;136:714-18.
- Rampen FH, Fleuren BA, de Boo TM, et al. Unreliability of self-reported burning tendency and tanning ability. *Arch Dermatol* 1988;124:885-8.
- Weinstock MA. Assessment of sun sensitivity by questionnaire: validity of items and formulation of a prediction rule. *J Clin Epidemiol* 1992;45:547-52.
- Lock-Andersen J, Wulf HC. Threshold level for measurement of UV sensitivity: reproducibility of phototest. *Photodermatol Photoimmunol Photomed* 1996;12:154-61.
- Westerhof W, Estevez-Uscanga O, Meens J, et al. The relation between constitutional skin color and photosensitivity estimated from UV-induced erythema and pigmentation dose-response curves. *J Invest Dermatol* 1990;94:812-16.
- Amblard P, Beani J, Gautron R, et al. Statistical study of individual variations in sunburn sensitivity in 303 volunteers without photodermatosis. *Arch Dermatol Res* 1982;274:195-206.
- Rubegni P, Cevenini G, Flori ML, et al. Relationship between skin color and sun exposure history: a statistical classification approach. *Photochem Photobiol* 1997;65:347-51.
- Aubin F, Manteaux A, Zultak M, et al. Cutaneous response to ultraviolet radiation-induced erythema in patients with type A dysplastic nevi. *Photodermatol Photoimmunol Photomed* 1991;8:7-11.
- Azizi E, Wax Y, Lusky A, et al. The recovery from ultraviolet radiation-induced erythema and melanoma risk factors: a case-control study. *J Am Acad Dermatol* 1990;23:30-6.
- Lock-Andersen J, Drzewiecki KT, Wulf HC. The measurement of constitutive and facultative skin pigmentation and estimation of sun exposure in Caucasians with basal cell carcinoma and cutaneous malignant melanoma. *Br J Dermatol* 1998;139:610-17.
- Green A, Martin NG. Measurement and perception of skin colour in a skin cancer survey. *Br J Dermatol* 1990;123:77-84.
- Hertzman C, Walter SD, From L, et al. Observer perception of skin color in a study of malignant melanoma. *Am J Epidemiol* 1987;126:901-11.
- Holman CD, Armstrong BK. Pigmentary traits, ethnic origin, benign nevi, and family history as risk factors for cutaneous malignant melanoma. *J Natl Cancer Inst* 1984;72:257-66.

38. Langholz B, Richardson J, Rappaport E, et al. Skin characteristics and risk of superficial spreading and nodular melanoma (United States). *Cancer Causes Control* 2000;11:741–50.
39. Landi MT, Baccarelli A, Tarone RE, et al. DNA repair, dysplastic nevi, and sunlight sensitivity in the development of cutaneous malignant melanoma. *J Natl Cancer Inst* 2002;94:94–101.
40. Landi MT, Baccarelli A, Calista D, et al. Combined risk factors for melanoma in a Mediterranean population. *Br J Cancer* 2001;85:1304–10.
41. Landi MT, Calista D, Landi G, et al. Clinical characteristics of 20 Italian melanoma-prone families. *Arch Dermatol* 1999;135:1554–5.
42. Billmeyer FW, Saltzman M. Principles of color technology. New York, NY: Wiley-Interscience, 1981.
43. Weatherall IL, Coombs BD. Skin color measurements in terms of CIELAB color space values. *J Invest Dermatol* 1992;99:468–73.
44. Bataille V, Bykov VJ, Sasieni P, et al. Photoadaptation to ultraviolet (UV) radiation in vivo: photoproducts in epidermal cells following UVB therapy for psoriasis. *Br J Dermatol* 2000;143:477–83.
45. SAS Institute, Inc. SAS/STAT user's guide, version 8. Cary, NC: SAS Institute, Inc, 1999.
46. Shriver MD, Parra EJ. Comparison of narrow-band reflectance spectroscopy and tristimulus colorimetry for measurements of skin and hair color in persons of different biological ancestry. *Am J Phys Anthropol* 2000;112:17–27.
47. Nishimura J, Ishii N, Sugita Y, et al. A case of carotoderma caused by a diet of the dried seaweed called Nori. *J Dermatol* 1998;25:685–7.
48. Lock-Andersen J, Wulf HC, Knudstorp ND. Interdependence of eye and hair colour, skin type and skin pigmentation in a Caucasian population. *Acta Derm Venereol* 1998;78:214–19.
49. Seitz JC, Whitmore CG. Measurement of erythema and tanning responses in human skin using a tri-stimulus colorimeter. *Dermatologica* 1988;177:70–5.
50. Jansen CT. Self-reported skin type and reactivity to UVB, UVA and PUVA irradiation. *Photodermatology* 1989;6:234–6.
51. Abel EA. Phototherapy. *Dermatol Clin* 1995;13:841–9.
52. Honigsmann H. Phototherapy for psoriasis. *Clin Exp Dermatol* 2001;26:343–50.
53. Snellman E, Jansen Ct, Leszczynski K, et al. Ultraviolet erythema sensitivity in anamnestic (I–IV) and phototested (1–4) Caucasian skin phenotype: the need for a new classification system. *Photochem Photobiol* 1995;62:769–72.
54. Lock-Andersen J, Therkildsen P, de Fine OF, et al. Epidermal thickness, skin pigmentation and constitutive photosensitivity. *Photodermatol Photoimmunol Photomed* 1997;13:153–8.
55. Lock-Andersen J, Gniadecka M, de Fine OF, et al. UV induced erythema evaluated 24 h post-exposure by skin reflectance and laser Doppler flowmetry is identical in healthy persons and patients with cutaneous malignant melanoma and basal cell cancer. *J Photochem Photobiol B* 1997;41:30–5.
56. Lee JH, Kim TY. Relationship between constitutive skin color and ultraviolet light sensitivity in Koreans. *Photodermatol Photoimmunol Photomed* 1999;15:231–5.
57. Shono S, Imura M, Ota M, et al. The relationship of skin color, UVB-induced erythema, and melanogenesis. *J Invest Dermatol* 1985;84:265–7.
58. Leenutaphong V. Relationship between skin color and cutaneous response to ultraviolet radiation in Thai. *Photodermatol Photoimmunol Photomed* 1996;11:198–203.
59. Selgrade MK, Smith MV, Oberhelman-Bragg LJ, et al. Dose-response for UV-induced immune suppression in people of color: differences based on erythema reactivity rather than skin pigmentation. *Photochem Photobiol* 2001;74:88–95.
60. Wee LK, Chong TK, Quee DK. Assessment of skin types, skin colours and cutaneous responses to ultraviolet radiation in an Asian population. *Photodermatol Photoimmunol Photomed* 1997;13:169–72.
61. Beitner H, Ringborg U, Wennersten G, et al. Further evidence for increased light sensitivity in patients with malignant melanoma. *Br J Dermatol* 1981;104:289–94.
62. Galosi A, Plewig G, Przybilla B, et al. UV-ray sensitivity of patients with malignant melanoma. (In German). *Hautarzt* 1985;36:449–52.
63. Elwood JM, Gallagher RP, Hill GB, et al. Cutaneous melanoma in relation to intermittent and constant sun exposure—The Western Canada Melanoma Study. *Int J Cancer* 1985;35:427–33.
64. Holman CD, Armstrong BK, Heenan PJ. Relationship of cutaneous malignant melanoma to individual sunlight-exposure habits. *J Natl Cancer Inst* 1986;76:403–14.
65. Weinstock MA, Colditz GA, Willett WC, et al. Melanoma and the sun: the effect of swimsuits and a “healthy” tan on the risk of nonfamilial malignant melanoma in women. *Am J Epidemiol* 1991;134:462–70.
66. Dubin N, Moseson M, Pasternack BS. Sun exposure and malignant melanoma among susceptible individuals. *Environ Health Perspect* 1989;81:139–51.
67. White E, Kirkpatrick CS, Lee JA. Case-control study of malignant melanoma in Washington State. I. Constitutional factors and sun exposure. *Am J Epidemiol* 1994;139:857–68.
68. Nelemans PJ, Groenendal H, Kiemeneys LA, et al. Effect of intermittent exposure to sunlight on melanoma risk among indoor workers and sun-sensitive individuals. *Environ Health Perspect* 1993;101:252–5.