IJC
International Journal of Cancer

Oral contraceptive use and cutaneous melanoma risk: a French prospective cohort study

I. Cervenka[1,2], Y. Mahamat-Saleh[1,2], I. Savoye[1,2], L. Dartois[1,2], M.C. Boutron-Ruault[1,2], A. Fournier[1,2] and M. Kvaskoff[1,2]

[1]CESP, Fac. de médecine, Univ. Paris-Sud, Fac. de médecine - UVSQ, INSERM, Université Paris-Saclay, 94805, Villejuif, France
[2]Gustave Roussy, F-94805, Villejuif, France

Cutaneous melanoma has been suspected to be influenced by female hormones. Several studies reported a positive association between oral contraceptive (OC) use and melanoma risk. However, findings were conflicting and data from large prospective studies are lacking. E3N is a prospective cohort of 98,995 French women aged 40–65 years at inclusion in 1990. Exposure to lifetime OC use was assessed in 1992 and through biennial questionnaire updates. To assess the association between OC use and melanoma risk, we used Cox models adjusted for age, pigmentary traits, residential ultraviolet (UV) exposure in county of birth and at inclusion and family history of skin cancer. Over 1992–2008, 539 melanoma cases were ascertained among 79,365 women. In age-adjusted models, we found a modest positive association between ever use of OCs and melanoma risk (hazard ratio (HR) = 1.18, 95% confidence intervals (CIs) = 0.98–1.42), which was reduced after adjustment (HR = 1.14, 95% CI = 0.95–1.38). The association was stronger in long-term users (duration ≥10 years: HR = 1.33, 95% CI = 1.00–1.75) and in women who used high-estrogen OCs (HR = 1.27, 95% CI = 1.04–1.56). Among users, there was an inverse association with age at first use (p_{trend} < 0.01), but no evidence of an association with age at last use or time since last use. OC use was positively associated with tanning bed use (OR = 1.14, CI = 1.01–1.29), sunburns (ptrend = 0.5) and sunscreen use (OR = 1.13, CI = 1.00–1.28) since age 25. Overall, our findings do not support a strong association between OC use and melanoma risk and suggest intentional UV exposure in OC users, which supports a potential confusion by UV exposure in this relationship.

Key words: cohort studies, cutaneous melanoma, epidemiology, oral contraceptives

Abbreviations: OC: oral contraceptive; EE: ethinyl estradiol; BMI: body mass index; HR: hazard ratio; CI: confidence interval; E3N: *Etude Epidémiologique auprès de femmes de l'Education Nationale*; CNIL: *Commission Nationale Informatique et Libertés*; UV: ultraviolet; MHT: menopausal hormone therapy; SD: standard deviation

Conflict of interest: None declared.

M.K. conceived and designed the study, obtained funding and collected the data for the E3N-SunExp study. I.C. performed the statistical analysis and drafted the manuscript. All authors contributed to the interpretation of data discussed in the manuscript, critically revised the manuscript and approved its final version. M.K. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Grant sponsor: Institut National de la Santé et de la Recherche Médicale; **Grant sponsor:** Institut National Du Cancer; **Grant sponsor:** Ligue Contre le Cancer; **Grant sponsor:** Mutuelle Générale de l'Education Nationale

DOI: 10.1002/ijc.31644

History: Received 24 Jan 2018; Accepted 5 Jun 2018; Online 13 Sep 2018

Correspondence to: Dr. Marina Kvaskoff, Inserm U1018, "Health across Generations" Team, Gustave Roussy, Espace Maurice Tubiana, 114 rue Edouard Vaillant, F-94805 Villejuif Cedex, France, Tel.: +33-1-4211-5864; Fax: +33-1-4211-4000, E-mail: marina.kvaskoff@gustaveroussy.fr

Introduction

Evidence suggests that cutaneous melanoma, the most lethal form of skin cancer, may be influenced by sex hormones. The hypothesis was based on several observations: the known influence of sex hormones on pigmentation and melanocytic tumors;[1] the occurrence of melasma and changes in nevus size and pigmentation during pregnancy;[2,3] the suggested progression of melanoma and worse prognosis among pregnant women;[4–7] the reported associations between reproductive factors[8] or hormone use[9–12] and melanoma risk; a higher incidence of melanoma among women compared with men in Europe;[13] and the observed "female advantage" with regards to melanoma survival, women being consistently reported to have better survival rates,[14] lower risks of metastasis[15] and lower mortality rates for melanoma than men,[16,17] regardless of body site, histological type or tumor stage.[14,18]

Oral hormones are the leading contraception method in industrialized countries, and they represent a significant source of exogenous hormone use.[19] In France, women started to use estrogens and progestogens for contraception in the 1960s, before it was even marketed for this indication. In 1970, when oral contraception started to be legally supervised in France, its use was already rising in the population, with 600,000 users. In 1972, it reached 1,200,000 users, and sales continued to rise.[20,21] Numerous types of pills have been marketed since then: doses of ethinyl estradiol (EE) progressively diminished, alternative regimens of administration appeared, with bi- or tri-phasic preparations including progestogens to

What's new?

Evidence suggests that cutaneous melanoma is influenced by female sex hormones. However, whether oral contraceptives, a major exogenous source of female hormones, are associated with melanoma remains unclear. In the present prospective cohort study, which included data on more than 79,000 French women, melanoma risk was associated with long-term oral contraceptive use and the use of high-dose estrogen oral contraceptives. Oral contraceptive users, however, were more likely to have used tanning beds and were more likely to report higher numbers of sunburns than non-oral conceptive users, suggesting that intentional ultraviolet exposure influenced the observed increase in melanoma risk.

mimic natural cycles, and several generations of less androgenic associated progestogens followed.

Oral contraceptive (OC) use has been inversely associated with ovarian, endometrial and colorectal cancer incidence.[22] However, it has also been positively associated with the risks of breast, cervical and liver cancers.[22] As a result, oral estrogen-progestogen contraceptives have been classified as carcinogenic to humans (Group 1) by the International Agency of Research on Cancer in 2007.[19]

The relation between OC use and cutaneous melanoma risk has been investigated in 7 cohort and 21 case–control studies to date, as analyzed in 3 meta-analyses.[23–25] These meta-analyses did not show consistent evidence for an association between OC use and melanoma risk, considering either current use, duration of use, time since last use or age at first use; however, there was a high level of heterogeneity in the included studies.[23]

The reported associations between reproductive factors and melanoma risk in women have been proposed to be related to different sun exposure behaviors between sexes rather than sex hormones.[26,27] However, few studies investigating hormonal exposures took sun exposure into account, and none of the previous investigations tested a potentially different effect of OC use on melanoma risk according to sun exposure.

Moreover, although melanoma is a heterogeneous tumor,[28] with one specific pathway linked to chronic sun exposure, very few studies explored the potential associations between OC use and melanoma risk according to anatomic site or histologic type of the tumor.

In view of the limited available evidence, more research is warranted in order to clarify the associations between OC use and melanoma risk in women. Our aim was to explore these associations and to investigate potentially differential effects by sun exposure level and tumor characteristics, in a large prospective cohort of French women.

Materials and Methods

The E3N cohort

E3N (*Etude Epidémiologique auprès de femmes de l'Education Nationale*) is an ongoing prospective cohort study involving 98,995 French women born in 1925–1950 and insured by a national health scheme covering workers from the French National Education System and their families. Women were enrolled in 1990 after returning a baseline self-administered questionnaire on their lifestyle and medical histories along with an informed consent. Follow-up questionnaires were sent every 2–3 years thereafter and addressed medical events such as cancer, which were confirmed through pathology reports. The E3N cohort received ethical approval from the French National Commission for Data Protection and Privacy (*Commission Nationale Informatique et Libertés* [CNIL]).

Data collection

Assessment of OC use. Use of oral hormones for contraception was collected at baseline and detailed information on lifetime OC use was recorded in the 1992 questionnaire, which collected the start date and duration of each episode of hormone use, together with the corresponding brand names. To facilitate reporting, the 1992 questionnaire included a booklet with an extensive list and color pictures of the treatments marketed in France. This information was then updated in each follow-up questionnaire.

OCs were either combined estrogens and progestogens, progestogen-only, or of unknown hormone type. We classified combined formulations by dose of estrogen, as high ($\geq$50 μg of EE), moderate (30–40 μg of EE), low (<30 μg of EE) or unknown. OCs containing 50 μg of mestranol were considered with a moderate dose of estrogen, 50 μg of mestranol being bioequivalent to 35 μg of EE, its active metabolite.[29] We also examined types of progestogen by distinguishing derivatives of estrane, gonane, pregnane, spironolactone or unknown.

Assessment of ultraviolet (UV) exposure. To estimate childhood and baseline residential ultraviolet radiation exposure among participants, counties of birth and of residence as collected at baseline were linked to a European database containing mean daily UV radiation doses in French counties.[30]

In a sub-population, the E3N-SunExp nested case–control study, detailed information was collected on lifetime residential and recreational UV exposures.[31] Briefly, in 2008, a specific questionnaire on UV exposure was sent to all incident skin cancer cases and three controls per case (matched on age, county of birth, education and duration of follow-up in the cohort). The sub-cohort included 1,558 skin cancer cases and 3,647 matched controls; women were asked to complete lifetime diaries of residence and holiday locations, with information on time spent in the sun and sun protection for each location. Data were linked to an international database on average daily UV doses,[32,33] which was used to calculate

total, residential and recreational UV scores. The questionnaire also collected data on number of sunburns and sunscreen use at ages <15, 15–25 and > 25 years and lifetime tanning bed use.

Assessment of other factors. Pigmentary characteristics were collected at baseline and included hair color (red, blond, light brown, dark brown and black), skin complexion (very fair, fair, medium, dark and very dark), number of nevi and number of freckles (none, few, many and very many), and skin sensitivity to sun exposure (low, moderate and high). Family history of skin cancer in first-degree relatives was collected in 2000. Age at menarche, parity and use of infertility drugs were collected in 1990 and 1992. The 1992 questionnaire also inquired about menstrual cycle length during midlife. Information about lifetime use of menopausal hormone therapy (MHT) and premenopausal progestogens was requested in 1992 and then updated in subsequent questionnaires. Each questionnaire inquired about Pap smears performed since the last returned questionnaire, and we used this information as a proxy for gynecological follow-up. Weight was available in each questionnaire and height was collected in 1990, 1995, 2000 and 2002, allowing for the calculation of a body mass index (BMI, as weight in kilograms divided by height in meters squared) during follow-up. Education was collected at baseline, and income was determined from the reported occupation in 1992. Marital status was collected in 1990, 1997, 2002, 2005 and 2008. Smoking status was available at each questionnaire.

Analysis populations and follow-up

Women who did not return the 1992 questionnaire (n = 12,827), those who were lost to follow-up from the 1992 questionnaire (n = 1,850), those with aberrant OC use data (i.e., start date after menopause onset) (n = 182), users of an injectable contraceptive (n = 45), women who reported a history of cancer other than basal-cell carcinoma at baseline (n = 4,698) and those who reported primary amenorrhea (n = 28), were excluded. Our final sample for analysis consisted of 79,365 women.

Follow-up started at the date of return of the 1992 questionnaire. Participants contributed person-time until the date of melanoma diagnosis, date of diagnosis of any other cancer (except basal-cell carcinoma), date of last questionnaire returned or date of end of follow-up (July 2008), whichever occurred first.

Sensitivity analyses adjusting for sun exposure behaviors were performed within the E3N-SunExp subpopulation, considering only melanoma cases (n = 366) and their matched controls (n = 852). Within this population, we additionally analyzed the relations between OC use and sun exposure behaviors in the 4,196 women included as controls in the study.

Statistical analyses

Statistical analyses were performed using the SAS statistical software package (version 9.4). We estimated hazard ratios (HRs) and 95% confidence intervals (CIs) using Cox proportional hazards regression models with age as the time scale. Associations between OC use and melanoma risk were first assessed in age-adjusted models, stratified on year of birth (in 5-year categories), then in models additionally adjusted for pigmentary traits (hair and skin color, sensitivity to sun exposure, number of nevi and freckles), residential UV exposure in county of birth and at inclusion and family history of skin cancer. In subsequent separate models, we performed additional adjustment for height (continuous), BMI (<18.5, 18.5–22.4, 22.5–24, 25–29 and $\geq$30 kg/m^2), parity (nulliparous, <3 and $\geq$3 children), menopausal status (pre- or postmenopausal), ever use of other hormonal treatments (MHT, premenopausal progestogens and infertility drugs), menstrual parameters (age at menarche, menstrual cycle length), gynecologic follow-up (Pap smear test reported in all returned questionnaires: yes or no) income (quartiles), education (<12 years, 12–15 years, $\geq$15 years), marital status (in a relationship, single, separated, widowed), and smoking (ever, never). Ever use of OC, age at last use, time since last use and duration of use were considered as time-dependent variables, as well as menopausal status and MHT use. However, we also checked the stability of the results using baseline characteristics only. Tests for linear trend were performed using an ordinal score for each quartile. Each OC type was considered in non-mutually exclusive categories, as most women took several OCs. Types of OCs were analyzed in models adjusted for the use of any other formulation; we excluded the only participant using an OC with a spirolactone derivative.

To further assess the association between duration of OC use and melanoma risk, we performed restricted cubic regressions based on the Cox proportional hazards regression. We used the RCS SAS Macro with duration of OC use at end of follow-up as a continuous variable, using 4 knots corresponding to quartile groups among users and a null duration as reference.[34] The model was adjusted for year of birth, age at inclusion, pigmentary traits, residential UV exposure and family history of skin cancer.

We analyzed melanoma risk according to anatomic site and histologic subtype using competing-risk models with the cause-specific hazards approach.[35,36] Cases with missing information on anatomic site (n = 45) or histologic subtype (n = 29) were excluded from these analyses. Tests for homogeneity using Chi-square tests were performed to compare estimates over strata.

We tested for effect modification by socioeconomic factors (income, occupation, education and marital status) use of other hormonal treatments, pigmentary traits, and height, using interaction tests.

Analyses in E3N-SunExp were performed through logistic regression modeling: associations between OC use and

melanoma risk were investigated using conditional logistic regression, considering the matched triplets of cases and controls, adjusting for recreational sun exposure; unconditional logistic regression adjusted for age and year of birth was performed when exploring associations between sun exposure and OC use in controls. We investigated effect modification by sun exposure of the relation between OC use and melanoma risk and by pigmentary traits of the relation between sun exposure and OC use in controls.

Women with missing data for OC exposure factors were excluded separately in each factor; figures are provided as footnotes in Table 2. For adjustment variables, missing values were imputed to the median or to the modal category if occurring in <5% of observations;[37] otherwise a missing category was created.

RESULTS

Over 966,604 woman-years of follow-up, 539 melanoma cases were ascertained among the 79,365 included women (median follow-up: 13.4 years).

Overall, compared with never-users, OC users were younger, had higher education levels and were more likely to belong to the highest quarter of the cohort in terms of income (Table 1). They were more likely to be in a relationship, to be parous and to have used other hormonal treatments (premenopausal progestogens or MHT). OC users were also more likely to report higher height and to have ever smoked. They were slightly more likely to report higher numbers of nevi and freckles and more likely to report frequent gynecologic follow-up.

The mean cumulative duration of OC use was 7.3 years (Table 2). The mean age at start of use was 28.7 years. At inclusion, most OC users were past users (78%), consistently with a mean age at last use of 38.2 years. Most users reported the use of several types of OCs in their lifetime (63%). A majority of users reported the use of a combined OC with high doses of estrogen (≥50 μg of EE; 58%) or with a gonane derivative (60%). The distribution of OC types ever used according to birth cohort shows that combined OCs with high doses of estrogens were the most used types of OCs and that its use increased across generations.

Compared with never use, ever use of OC was associated with a modest increase in melanoma risk in the age-adjusted model (HR = 1.18, CI = 0.98–1.42) and the relation was slightly reduced after adjustment (HR = 1.14, CI = 0.95–1.38) (Table 3). However, women who used OCs for 10 years or more over their lifetime had a higher risk of melanoma compared with never users and the association was stable after adjustment (HR = 1.33, CI = 1.00–1.75; p_{trend} = 0.06). A cubic spline regression confirmed a higher melanoma risk with increased duration of OC use, particularly after 7 years of use (Figure 1). Among OC users, we found a positive association with duration of use (HR = 1.02, CI = 1.00–1.04 for each additional year of use). There was an inverse association between age at first OC use and melanoma risk among users (p_{trend} < 0.01), consistent with a negative correlation between age at first use and duration of use (r = −0.23). However, age at last use or time since last use among users were not associated with melanoma risk.

Use of formulations with high doses of estrogen (≥50 μg of EE) was positively associated with melanoma risk, even after adjustment (HR = 1.27, CI = 1.04–1.56). However, there was no evidence of a dose–response relationship across categories of increasing doses of estrogen in formulations (low, medium, high), and no other type of formulation was associated with melanoma risk.

Results were unchanged when models were additionally adjusted for parity, menopausal status, age at menarche, menstrual cycle length, use of other hormonal treatments, gynecologic follow-up, height, BMI, education, income, marital status, or smoking (data not shown).

When HRs were computed for each generation stratum (born <1930, 1930–1934, 1935–1939, 1940–1945 and >1945), we found a stronger association between OC use and melanoma risk among women born between 1930 and 1935 (HR = 1.68, CI = 1.00–2.81 in models adjusted for pigmentary traits, residential UV exposure and family history of skin cancer), although no heterogeneity was detected across generations ($p_{homogeneity}$ = 0.59).

We found no effect modification of the association between OC use and melanoma risk by pigmentary traits, use of other hormonal treatments, height, and socio-economic factors. However, the association between OC use and melanoma risk was stronger in women working in the National Education system but who were not teachers (HR = 1.69, CI = 1.05–2.71 in adjusted model) than in teachers (HR = 1.06, CI = 0.85–1.32) ($p_{interaction}$ = 0.07 in non-adjusted models, and $p_{interaction}$ = 0.08 in adjusted models).

In competing-risk analyses considering histologic type (superficial spreading, lentigo maligna, nodular, acrolentiginous and other) and melanoma site (head and neck, trunk, upper or lower limbs), associations were positive for all types and sites, except the trunk (Table 4) and no heterogeneity was detected across subtypes ($p_{homogeneity}$ = 0.54) or sites ($p_{homogeneity}$ = 0.23).

In the E3N-SunExp population, the association between OC use and melanoma risk was positive although not statistically significant. Adjustment for residential and recreational UV exposure did not substantially modify the results (data not tabulated).

When investigating the potential associations between UV exposure behaviors and OC use among controls, we found that even after adjustment for age and stratification on year of birth, OC users were more likely to report tanning bed use compared with non-users (OR = 1.14, CI = 1.01–1.29). They were also more likely to report higher numbers of sunburns since age 25 (p_{trend} = 0.05). Finally, OC use was positively associated with the sunscreen use at ages 15–25 (p_{trend} = 0.04)

Table 1. Characteristics of study participants[1] according to ever use of OCs at the end of follow-up, E3N cohort (n = 79,365)

	OC use	
	Never (n = 31,249), n (%)	Ever (n = 48,116), n (%)
Year of birth		
‹1930	5,055 (16.2)	920 (1.9)
1930–1934	6,974 (22.3)	3,078 (6.4)
1935–1939	7,663 (24.5)	7,198 (15.0)
1940–1945	6,234 (20.0)	12,815 (26.6)
›1945	5,323 (17.0)	24,105 (50.1)
Education level		
‹12 years	5,400 (17.3)	4,498 (9.4)
12–14 years	16,904 (54.1)	24,745 (51.4)
≥15 years	8,945 (28.6)	18,873 (39.2)
Income (Fr/year)[2]		
Quartile 1	4,093 (13.1)	5,390 (11.2)
Quartile 2	8,473 (27.1)	13,729 (28.5)
Quartile 3	6,773 (21.7)	9,213 (19.2)
Quartile 4	7,809 (25.0)	14,970 (31.1)
Missing	4,101 (13.12)	4,814 (10.00)
Marital status		
In a relationship	20,469 (65.5)	34,799 (72.3)
Single	2,926 (9.4)	2,470 (5.1)
Separated	2,539 (8.1)	6,769 (14.1)
Widowed	5,315 (17.0)	4,078 (8.5)
Age at menarche		
‹13 years	13,805 (44.2)	21,907 (45.5)
13–14 years	14,234 (45.6)	22,069 (45.9)
≥15 years	3,210 (10.3)	4,140 (8.6)
Length of menstruation cycles		
≤24 days	1,434 (4.6)	2,062 (4.3)
25–31 days	18,715 (59. 9)	31,241 (64.9)
≥32 days	1,048 (3.4)	1,969 (4.1)
Irregular	2,191 (7.0)	3,360 (7.0)
Unknown	7,861 (25.2)	9,484 (19.7)
Parity		
Nulliparous	5,023 (16.1)	4,162 (8.7)
‹3 children	16,886 (54.0)	30,151 (62.7)
≥3 children	9,340 (29.9)	13,803 (28.7)
Menopausal status[3]		
Premenopausal	454 (1.5)	1,985 (4.1)
Postmenopausal	30,795 (98.5)	46,131 (95.9)
Ever use of premenopausal progestogens[3]		
No	19,217 (61.5)	20,831 (43.3)
Yes	12,032 (38.5)	27,285 (56.7)
Ever use of menopausal hormone therapy[3,4]		
No	11,101 (36.1)	10,692 (23.2)
Yes	19,694 (63.9)	35,439 (76.8)
Ever use of fertility drugs		
No	30,158 (96.5)	45,907 (95.4)
Yes	1,091 (3.5)	2,209 (4.6)
Height (cm)[2]		
Quartile 1	8,413 (26.9)	10,470 (21.8)
Quartile 2	7,035 (22.5)	10,447 (21.7)
Quartile 3	7,014 (22.5)	11,400 (23.7)
Quartile 4	8,787 (28.1)	15,799 (32.8)
Body mass index (kg/m^2)		
‹18.5	1,140 (3.7)	1,477 (3.1)
18.5–22.4	10,513 (33.6)	19,516 (40.6)
22.5–24	8,608 (27.6)	13,373 (27.8)
25–29	8,376 (26.8)	10,846 (22.5)
≥30	2,612 (8.4)	2,904 (6.0)
Smoking at inclusion		
Ever	11,539 (36.9)	25,061 (52.1)
Never	19,703 (63.1)	23,037 (47.9)
Skin color		
Very fair	325 (1.0)	589 (1.2)
Fair	17,976 (57.5)	28,238 (58.7)
Medium	12,413 (39.7)	18,592 (38.6)
Dark	519 (1.7)	661 (1.4)
Very dark	16 (0.1)	36 (0.1)
Hair color		
Red	507 (1.6)	815 (1.7)
Blond	3,019 (9.7)	4,954 (10.3)
Light brown	18,966 (60.7)	28,806 (59.9)
Dark brown	7,190 (23.0)	11,264 (23.4)
Black	1,567 (5.0)	2,277 (4.7)
Number of nevi		
Very many	2,686 (8.6)	5,835 (12.1)
Many	12,649 (40.5)	21,743 (45.2)
A few	12,070 (38.6)	16,653 (34.6)
None	38,44 (12.3)	3,885 (8.1)
Number of freckles		
Very many	1,436 (4.6)	2,657 (5.5)
Many	8,471 (27.1)	14,455 (30.0)
A few	7,456 (23.9)	11,691 (24.3)
None	13,886 (44.4)	19,313 (40.1)

(Continues)

Table 1. Continued

	OC use	
	Never (*n* = 31,249), *n* (%)	**Ever (*n* = 48,116), *n* (%)**
Skin sensitivity to sun exposure		
High	8,477 (27.1)	13,929 (28.9)
Moderate	15,072 (48.2)	23,862 (49.6)
Low	7,700 (24.6)	10,325 (21.5)
Family history of skin cancer[5]		
No	30,962 (99.1)	47,572 (98.9)
Yes	287 (0.9)	544 (1.1)
Residential UV dose at inclusion[2]		
Quartile 1	6,720 (21.5)	10,412 (21.6)
Quartile 2	8,249 (26.4)	13,626 (28.3)
Quartile 3	8,034 (25.7)	12,444 (25.9)
Quartile 4	8,246 (26.4)	11,634 (24.2)
Residential UV dose at birth[2]		
Quartile 1	6,939 (22.2)	10,311 (21.4)
Quartile 2	7,334 (23.5)	11,828 (24.6)
Quartile 3	6,884 (22.0)	10,537 (21.9)
Quartile 4	7,531 (24.1)	11,790 (24.5)
Missing	2,561 (8.2)	3,650 (7.6)
Pap smear reported in all returned questionnaires[3]		
No	20,685 (66.2)	23,447 (48.7)
Yes	10,564 (33.8)	24,669 (51.3)

[1]All parameters are given at baseline, except when mentioned in the footnotes
[2]Cut-off points for quartiles were 111 kF, 116 kF, and 158 kF for income; 158 cm, 161 cm, and 165 cm for height; and 2.4kJ/m^2, 2.5kJ/m^2, and 2.7kJ/m^2 for residential UV dose in places of birth and of residence at inclusion.
[3]At the end of follow-up
[4]Reported in 2000
[5]Among postmenopausal women

and $\geq$ 25 years (p_{trend} = 0.01), and inversely associated with residential hours of UV exposure (p_{trend} < 0.01), but these associations were no longer statistically significant after adjusting for age and year of birth. We found no association with hours of sun exposure or UV scores, and no interaction between sun exposure behaviors and pigmentary traits in relation to OC use.

DISCUSSION

In this large prospective cohort, we found a positive association between OC use and melanoma risk, which was restricted to long-term users and users of OCs containing high doses of estrogen. We found no effect modification of sun exposure on the relation between OC use and melanoma risk. However, OC users were more likely to report use of tanning beds and

Table 2. Description of OC use in ever users of OCs as assessed at the end of follow-up, E3N cohort (*n* = 48,116)

	OC, *n* (%) *n* = 48,116
Use at inclusion	
Current	4,173 (8.7)
Past	37,597 (78.1)
Status unknown	6,243 (13.0)
Type of OC used[1]	
Progestogen only	2,295 (4.8)
Combined OC, by estrogen dose	
High	27,843 (57.9)
Moderate	18,842 (39.2)
Low	1,889 (3.9)
Unknown estrogen dose	8,795 (18.3)
Combined OC, by type of progestogen	
Estrane	12,910 (26.8)
Gonane	28,726 (59.7)
Pregnane	568 (1.2)
Spirolactone derivate	1 (0.0)
Unknown progestogen	10,848 (22.6)
Duration of OC use (years), mean (SD)[2]	7.3 (6.0)
Age at start of OC (years), mean (SD)[3]	28.7 (6.5)
Age at last OC use (years), mean (SD)[4,5]	38.2 (8.1)

[1]11,669 missing values among ever users.
[2]17,162 missing values among ever users.
[3]15,889 missing values among ever users.
[4]Among past users.
[5]18,123 missing values among ever users

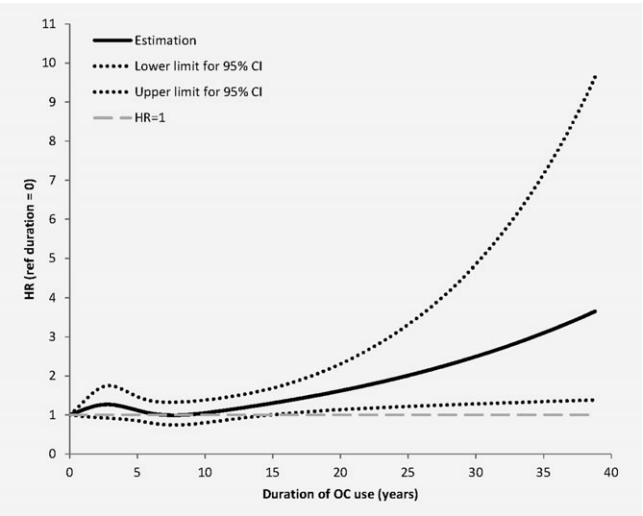

Figure 1. Spline regression of the association between duration of OC use at the end of follow-up and melanoma risk (obtained by spline regression with four knots [0, 2.5, 6 and 11 years] and never OC use as the reference), E3N cohort (*n* = 79,365).

Table 3. Hazard ratios (HRs) and 95% confidence intervals (CIs) for the associations between oral contraceptive use and melanoma risk, E3N cohort (n = 79,365)

	Cases n = 539	Person-years	HR (95% CI)[1]	HR (95% CI)[2]
OC use				
Never	206	218,147	Ref	Ref
Ever	333	353,887	1.18 (0.98–1.42)	1.14 (0.95–1.38)
Past	322	391,972	1.20 (0.98–1.46)	1.16 (0.94–1.41)
Current	7	17,973	0.77 (0.31–1.91)	0.74 (0.30–1.84)
Status unknown	4	12,258	0.69 (0.17–2.83)	0.67 (0.16–2.77)
Duration of OC use				
Never use	206	218,147	Ref	Ref
<10 years	138	185,917	1.14 (0.90–1.44)	1.10 (0.87–1.38)
≥10 years	77	85,659	**1.38 (1.05–1.82)***	**1.33 (1.00–1.75)***
p-trend			**0.03***	0.06
Type of OC ever used[3]				
Never use	206	218,147	Ref	Ref
Progestogen only	11	28,415	0.71 (0.39–1.29)	0.70 (0.38–1.28)
Combinations, by estrogen dose				
High	206	342,075	**1.31 (1.07–1.61)***	**1.27 (1.04–1.56)***
Moderate	122	232,510	0.99 (0.79–1.25)	0.96 (0.76–1.20)
Low	7	23,534	0.63 (0.29–1.33)	0.61 (0.29–1.31)
Unknown estrogen dose	68	107,803	1.16 (0.89–1.51)	1.15 (0.88–1.50)
Combinations, by type of progestogen				
Estrane	84	158,825	0.95 (0.74–1.22)	0.91 (0.71–1.16)
Gonane	198	353,480	1.18 (0.96–1.45)	1.14 (0.93–1.41)
Pregnane	5	6,974	1.46 (0.60–3.55)	1.38 (0.57–3.34)
Unknown progestogen type	81	133,322	1.13 (0.88–1.45)	1.13 (0.88–1.45)
Duration of OC use among users				
Continuous			**1.02 (1.00–1.04)***	**1.02 (1.00–1.04)***
≤2 years	50	50,009	Ref	Ref
2–5 years	47	63,980	0.92 (0.61–1.39)	0.91 (0.60–1.38)
5–10 years	41	72,122	0.95 (0.64–1.42)	0.93 (0.63–1.38)
≥10 years	77	85,659	1.13 (0.77–1.66)	1.12 (0.77–1.65)
p-trend			0.47	0.47
Age at first use among users[4]				
Continuous			0.97 (0.94–1.00)	0.97 (0.94–1.00)
Quartile 1	47	116,380	Ref	Ref
Quartile 2	52	86,728	0.73 (0.49–1.09)	0.75 (0.50–1.11)
Quartile 3	59	98,533	**0.66 (0.45–0.99)***	0.68 (0.46–1.02)
Quartile 4	61	95,014	**0.54 (0.35–0.84)***	**0.56 (0.36–0.86)***
p-trend			**<0.01***	**<0.01***
Age at last use among users[4]				
Continuous			0.99 (0.97–1.01)	0.99 (0.97–1.01)
Quartile 1	50	69,960	Ref	Ref
Quartile 2	53	63,934	1.00 (0.67–1.47)	0.96 (0.65–1.42)
Quartile 3	65	83,757	0.97 (0.66–1.41)	0.95 (0.65–1.39)
Quartile 4	55	70,007	0.76 (0.50–1.16)	0.75 (0.49–1.15)
p-trend			0.16	0.16

(Continues)

Table 3. Continued

	Cases *n* = 539	Person-years	HR (95% CI)[1]	HR (95% CI)[2]
Time since last use among users[4]				
Continuous			1.01 (0.99–1.03)	1.01 (0.99–1.03)
Quartile 1	126	195,118	Ref	Ref
Quartile 2	66	83,079	1.00 (0.73–1.37)	1.00 (0.73–1.37)
Quartile 3	22	17,754	1.02 (0.62–1.66)	1.03 (0.63–1.69)
Quartile 4	9	3,166	1.34 (0.66–2.73)	1.39 (0.68–2.82)
p-trend			0.58	0.56

*Significant at P value $\leq$ 0.05.
[1]Model adjusted for age and stratified according to year of birth.
[2]Model additionally adjusted for residential UV exposure at birth and at inclusion, pigmentary traits and family history of skin cancer.
[3]Analyses additionally adjusted for unknown type of OC.
[4]Cutoff points for quartiles were 24, 27 and 32 years for age at first use; 32, 38 and 45 years for age at last use; and 19, 27 and 32 years for time since last use.

Table 4. Hazard ratios (HRs) and 95% confidence intervals (CIs) for the associations between oral contraceptive use and melanoma risk according to histological type and body site of melanoma, E3N cohort (*n* = 79,365)

	HR (95% CI)[1]	HR (95% CI)[2]
OC use		
Never	Ref	Ref
Ever		
Body site		
Trunk (*n* = 86)	0.77 (0.49–1.21)	0.74 (0.47–1.17)
Upper limbs (*n* = 106)	1.30 (0.85–1.97)	1.25 (0.82–1.91)
Lower limbs (*n* = 243)	1.29 (0.98–1.71)	1.26 (0.95–1.66)
Head and neck (*n* = 59)	1.31 (0.74–2.34)	1.28 (0.72–2.29)
Histological type		
Superficial spreading (*n* = 334)	1.10 (0.87–1.39)	1.06 (0.84–1.34)
Nodular (*n* = 13)	1.12 (0.34–3.70)	1.14 (0.34–3.80)
Lentigo maligna (*n* = 51)	1.49 (0.82–2.77)	1.45 (0.78–2.69)
Acro lentiginous (*n* = 19)	2.39 (0.86–6.74)	2.42 (0.85–6.90)
Other (*n* = 93)	1.34 (0.87–2.11)	1.29 (0.82–2.03)

[1]Model adjusted for age and stratified according to year of birth.
[2]Model additionally adjusted for residential UV exposure at birth and at inclusion, pigmentary traits and family history of skin cancer.

of sunscreen in adulthood, although other sun exposure behaviors were not associated with OC use.

Overall, these results contrast with the findings from three meta-analyses that suggest no influence of OC use on melanoma risk in women overall.[23–25] Duration of OC use was associated with a higher risk of melanoma among users in our study, especially after 7 years of use. In most previous studies considering duration of use, no relation was found, except in a prospective cohort in which a positive association was observed in women who used OCs for at least 10 years, but for current use only.[38]

In E3N, information on OC exposure was very detailed, and we had the opportunity to study different types of hormones used. Most women took multiple OC types in their lifetime, making it difficult to analyze each type of treatment taken exclusively. However, when analyzing each type of OC while adjusting simultaneously for other types, formulations with high doses of estrogen were positively associated with melanoma risk, but not with other formulations. This is consistent with the positive association found in women using OCs with high cumulative daily doses in a previous case–control study.[11] However, our results do not support a dose–response relationship since we did not observe any trend across increasing doses. This pattern of association may have at least two explanations. First, OCs with high doses of estrogen were the first ones on the market, and in the cohort a majority of OC users reported to have ever used this type of OCs, suggesting a higher statistical power in the analyses compared with other OCs. Second, as older generations are more likely to have used OCs with high estrogen doses, it could be hypothesized that the association with high doses of estrogen is due to a cohort effect. However, in our study population, type of OC was missing for a large fraction of participants among older generations, and so this hypothesis cannot be verified. The 'high dose of estrogens' category was more frequent in women from recent birth cohorts, i.e. who were born >1945.

The E3N-SunExp nested case–control study enabled us to consider detailed data on both residential and recreational sun exposure. While we found no association between OC use and lifetime hours of sun exposure or UV score, we observed that OC use was positively associated with sunburns after 25 years and tanning bed use, even after adjusting for age and year of birth. However, adjusting for sunburns and tanning beds had no substantial effect on the relation between OC use and melanoma risk in our study. Nevertheless, these observations underline the importance of considering the potential confounding effect of sun exposure behaviors when exploring the

relation between OC use and melanoma risk, and the need to further explore the sun exposure profile of OC users in future research.

It could be hypothesized that OC users might use the medical care system or perform skin self-examinations more frequently, and thus be more likely to be diagnosed or to have their pigmented lesions removed. In our analysis, we used Pap smears as a proxy for gynecologic follow-up, but we did not observe any effect modification with this parameter. Also, using oral hormones for contraception was more frequent among educated and wealthy women a few decades ago, which could suggest a bias related to socioeconomic factors. Such a bias would be less likely nowadays since the pill is now widely used regardless of socioeconomic status in the general population.[39] Nevertheless, we found no impact of socioeconomic factors on the relation between OC use and melanoma risk. OC use seemed to be more strongly associated with melanoma risk among women working in the National Education system but who were not teachers, which underlines the importance of repeating this analysis in study populations that are more representative of the general population.

Our study has several strengths, particularly its sample size – it is currently the largest prospective study on the association between OC use and melanoma risk to date; the ability to study different melanoma sites and types; the availability of detailed data on OC use over lifetime, enabling us to study different types of hormonal preparations; and the availability of sun exposure data that enabled us to explore the potential confounding or modification effects of sun exposure behaviors on the association between OC use and melanoma risk. We were also able to explore the sun exposure profile of OC users, which has never been investigated to date to our knowledge. However, several limitations need to be considered in the interpretation of our findings. The major limitation is the relatively high age of E3N participants at cohort inclusion (40–65 years): the long delay between women's OC exposure and our ability to observe incident melanoma cases implies that we were only able to study long-term effects of OCs, as opposed to current use of OCs and their potential shorter-term effects on melanoma risk. This is an important limitation considering that sex hormones were recently suggested to affect the risk of early-onset melanoma.[40,43] Another, more moderate limitation, is the overall higher socioeconomic status of the cohort, with a high proportion of teachers. However, this is also an advantage in terms of reliability of self-reported features in this highly-motivated population.[41] Finally, the potential effects of UV exposure might not have been fully addressed in our models, since UV was estimated through proxy measures instead of objectively validated questionnaires, as recently developed.[42] Nevertheless, an asset of our study was to consider lifetime sun exposure.

Conclusion

We found a positive association between OC use and melanoma risk, which was restricted to long-term users and users of OCs containing high doses of estrogens. Our results also suggest a specific profile of OC users regarding their attitude towards tanning. Therefore, we cannot exclude that the observed relation between OC use and melanoma risk, which has been debated in the literature for almost 40 years, could relate to particular sun exposure behaviors in users. Further research is thus critical to increase our understanding of the risk profile of women diagnosed with melanoma, and to question the hypothesis of a hormonal influence on melanoma risk. This research should use prospective cohort data spanning younger ages at melanoma diagnosis, and include social and behavioral description of OC users.

Acknowledgments

We are grateful to the study participants for their continued participation and to practitioners for providing pathology reports. We also thank all members of the E3N study group, particularly Rafika Chaït, Lyan Hoang, Maxime Valdenaire, Marie Fangon, Camille Laplanche, Pascale Gerbouin-Rérolle and Sabine Moreira-Faria for their technical assistance. The E3N cohort from the French National Institute of Health and Medical Research (INSERM) was supported by the Mutuelle Générale de l'Education Nationale; the Gustave Roussy Institute; and the French League against Cancer (LNCC). The E3N-SunExp study was supported by a grant from the French National Cancer Institute (INCA #2010-1-PL SHS-11-INSERM 11-1). IC was supported by a PhD scholarship from the French Ministry of Research.

References

1. Mitkov M, Joseph R, Copland J. Steroid hormone influence on melanomagenesis. *Mol Cell Endocrinol* 2015;417:94–102.
2. Zampino MR, Corazza M, Costantino D, et al. Are melanocytic nevi influenced by pregnancy? A dermoscopic evaluation. *Dermatol Surg Off Publ Am Soc Dermatol Surg Al* 2006;32:1497–504.
3. Richtig G, Byrom L, Kupsa R, et al. Pregnancy as driver for melanoma. *Br J Dermatol* 2016;177:854–7.
4. Kyrgidis A, Lallas A, Moscarella E, et al. Does pregnancy influence melanoma prognosis? A meta-analysis. *Melanoma Res* 2017;27:289–99.
5. Johansson ALV, Andersson TM-L, Plym A, et al. Mortality in women with pregnancy-associated malignant melanoma. *J Am Acad Dermatol* 2014;71:1093–1.
6. Byrom L, Olsen C, Knight L, et al. Increased mortality for pregnancy-associated melanoma: systematic review and meta-analysis. *J Eur Acad Dermatol Venereol JEADV* 2015;29:1457–66.
7. Wielowieyska-Szybińska DK, Spałkowska M, Wojas-Pelc A. Melanoma in pregnancy: a case report and review of the literature. *Adv Dermatol Allergol Dermatol Alergol* 2015;32:483–7.
8. Kvaskoff M, Bijon A, Mesrine S, et al. Cutaneous melanoma and endogenous hormonal factors: a large French prospective study. *Am J Epidemiol* 2011;173:1192–202.
9. Botteri E, Støer NC, Sakshaug S, et al. Menopausal hormone therapy and risk of melanoma: do estrogens and progestins have a different role? *Int J Cancer* 2017;141:1763–70.
10. Bhupathiraju SN, Grodstein F, Stampfer MJ, et al. Exogenous hormone use: oral contraceptives, postmenopausal hormone therapy, and Health outcomes in the Nurses' Health Study. *Am J Public Health* 2016;106:1631–7.
11. Koomen ER, Joosse A, Herings RMC, et al. Estrogens, oral contraceptives and hormonal replacement therapy increase the incidence of

cutaneous melanoma: a population-based case-control study. *Ann Oncol Off J Eur Soc Med Oncol* 2009;20:358–64.
12. Holly EA, Cress RD, Ahn DK. Cutaneous melanoma in women: ovulatory life, menopause, and use of exogenous estrogens. *Cancer Epidemiol Biomark Prev Publ Am Assoc Cancer Res Cosponsored Am Soc Prev Oncol* 1994;3:661–8.
13. ENHIS 2009 http://www.euro.who.int/__data/assets/pdf_file/0009/97029/4.2.-Incidence-of-melanoma-EDITED_layouted.pdf.
14. Lasithiotakis K, Leiter U, Meier F, et al. Age and gender are significant independent predictors of survival in primary cutaneous melanoma. *Cancer* 2008;112:1795–804.
15. Joosse A, de Vries E, Eckel R, et al. Gender differences in melanoma survival: female patients have a decreased risk of metastasis. *J Invest Dermatol* 2011;131:719–26.
16. Binder-Foucard F, Bossard N, Delafosse P, et al. Cancer incidence and mortality in France over the 1980-2012 period: solid tumors. *Rev Epidemiol Sante Publique* 2014;62:95–108.
17. ENCR factsheet 2015 http://www.encr.eu/images/docs/factsheets/ENCR_Factsheet_Malignant_Melanoma_2015.pdf.
18. de Vries E, Nijsten TEC, Visser O, et al. Superior survival of females among 10,538 Dutch melanoma patients is independent of Breslow thickness, histologic type and tumor site. *Ann Oncol Off J Eur Soc Med Oncol* 2008;19:583–9.
19. IARC Monographs- Monographs available in PDF format [Internet]. [cited 2017 Dec 5]; Available from: http://monographs.iarc.fr/ENG/Monographs/vol91/index.php
20. Leridon H, Sardon J-P, Collomb P, et al. La contraception en France en 1978. Une enquête INED-INSEE. *Population* 1979;34:1349–90.
21. Chauveau S. Les espoirs déçus de la loi Neuwirth. *Clio Femmes Genre Hist* 2003;18:223–39.
22. Gierisch JM, Coeytaux RR, Urrutia RP, et al. Oral contraceptive use and risk of breast, cervical, colorectal, and endometrial cancers: a systematic review. *Cancer Epidemiol Biomark Prev Publ Am Assoc Cancer Res Cosponsored Am Soc Prev Oncol* 2013;22:1931–43.
23. Gandini S, Iodice S, Koomen E, et al. Hormonal and reproductive factors in relation to melanoma in women: current review and meta-analysis. *Eur J Cancer Oxf Engl 1990* 2011;47:2607–17.
24. Karagas MR, Stukel TA, Dykes J, et al. A pooled analysis of 10 case-control studies of melanoma and oral contraceptive use. *Br J Cancer* 2002;86:1085–92.
25. Gefeller O, Hassan K, Wille L. Cutaneous malignant melanoma in women and the role of oral contraceptives. *Br J Dermatol* 1998;138:122–4.
26. Kravdal O. Is the relationship between childbearing and cancer incidence due to biology or lifestyle? Examples of the importance of using data on men. *Int J Epidemiol* 1995;24:477–84.
27. Kaae J, Andersen A, Boyd HA, et al. Reproductive history and cutaneous malignant melanoma: a comparison between women and men. *Am J Epidemiol* 2007;165:1265–70.
28. Whiteman DC, Watt P, Purdie DM, et al. Melanocytic nevi, solar keratoses, and divergent pathways to cutaneous melanoma. *J Natl Cancer Inst* 2003;95:806–12.
29. Brody SA, Turkes A, Goldzieher JW. Pharmacokinetics of three bioequivalent norethindrone/mestranol-50 micrograms and three norethindrone/ethinyl estradiol-35 micrograms OC formulations: are "low-dose" pills really lower? *Contraception* 1989;40:269–84.
30. Verdebout J. A European satellite-derived UV climatology available for impact studies. *Radiat Prot Dosimetry* 2004;111:407–11.
31. Savoye I, Olsen CM, Whiteman DC, et al. Patterns of ultraviolet radiation exposure and skin cancer risk: the E3N-SunExp study. *J Epidemiol* 2018;28:27–33.
32. Kift R, Webb AR, Page J, et al. A web-based tool for UV lrradiance data: predictions for European and Southeast Asian Sites. *Photochem Photobiol* 2006;82:579–86.
33. Gschwind B, Ménard L, Albuisson M, et al. Converting a successful research project into a sustainable service: The case of the SoDa Web service. *Environ Model Softw* 2006;21:1555–61.
34. Desquilbet L, Mariotti F. Dose-response analyses using restricted cubic spline functions in public health research. *Stat Med* 2010;29:1037–57.
35. Putter H, Fiocco M, Geskus RB. Tutorial in biostatistics: competing risks and multi-state models. *Stat Med* 2007;26:2389–430.
36. Pintilie M. Analysing and interpreting competing risk data. *Stat Med* 2007;26:1360–7.
37. Garcia-Acosta S, Clavel-Chapelon F. Dealing with missing, abnormal and incoherent data in E3N cohort study. *Rev Epidemiol Sante Publique* 1999;47:515–23.
38. Feskanich D, Hunter DJ, Willett WC, et al. Oral contraceptive use and risk of melanoma in premenopausal women. *Br J Cancer* 1999;81:918–23.
39. Bajos N, Oustry P, Leridon H, et al. Les inégalités sociales d'accès à la contraception en France, Abstract, Resumen. *Population* 2004;59:479–502.
40. Liu-Smith F, Ziogas A. An age-dependent interaction between sex and geographical UV index in melanoma risk. *J Am Acad Dermatol* 2017, in press.
41. Clavel-Chapelon F. E3N Study Group. Cohort Profile: The French E3N Cohort Study. *Int J Epidemiol* 2015;44:801–9.
42. Køster B, Søndergaard J, Nielsen JB, et al. The validated sun exposure questionnaire: association of objective and subjective measures of sun exposure in a Danish population-based sample. *Br J Dermatol* 2017;176:446–56.
43. Anderson WF, Pfeiffer RM, Tucker MA, Rosenberg PS. Divergent cancer pathways for early-onset and late-onset cutaneous malignant melanoma. *Cancer* 2009;115:4176–85.

THE DIFFERENCE OF BREAKTHROUGH MOMENTS

WITH COMPLETE SOLUTIONS FOR GROUNDBREAKING DISCOVERIES FROM A TRUSTED PARTNER.

Your next breakthrough could be closer than you imagine, especially with the right resources to help you advance your research. At BD, we are dedicated to helping you get the data you need, when, where and how you need it. Our integrated solutions in instrumentation, software and reagents are optimized to work together to bring you closer to your next breakthrough moment. And you can depend on us for world-class training, service and support to help you get the most from the results your research depends on. Discover a range of optimized solutions that open endless possibilities for your future research. **Discover the new BD.**

Learn how you can advance your research >

BD