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# A Prospective Study of Telomere Length and the Risk of Skin Cancer

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## Abstract

Telomere length plays a critical role in tumorigenesis. Using quantitative real-time PCR, we prospectively measured relative telomere length in a nested case-control study within the Nurses' Health Study (NHS): 218 melanoma cases, 285 squamous cell carcinoma (SCC) cases, 300 basal cell carcinoma (BCC) cases, and 870 controls. We observed that shorter telomeres were associated with a decreased number of moles (p=0.002) and a decreased risk of melanoma. Women in the second and first quartiles, those with the shortest telomere length, had an OR for melanoma of 0.54 (95% CI, 0.29-1.01) and 0.59 (95% CI, 0.31-1.13), respectively, compared with those in the fourth quartile (P, trend = 0.09). There was no clear trend between telomere length and SCC risk. In contrast, we found that shorter telomere length was associated with an increased risk of BCC. Compared with those in the fourth quartile, women in the first quartile had an OR of 1.85 (95%CI, 0.94-3.62) (P, trend =0.09). The opposing associations observed should be interpreted with caution, and further research is needed to confirm these possible associations.

### Keywords

telomere length; melanoma; skin cancer

## Introduction

Telomeres are long hexameric (TTAGGG)n repeats at the ends of the p and q arms of linear eukaryotic chromosomes. Studies in mice and yeast have demonstrated the critical role of telomeres in maintaining the structural integrity of chromosomes by preventing fusion of chromosomal ends, nucleolytic decay, end-to-end fusion, and atypical recombination (Lundblad and Szostak, 1989). However, at each cell division, DNA polymerases fail to completely replicate telomeres, resulting in cumulative erosion of the telomeres (Chan and

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Blackburn, 2004; Chang and Harley, 1995). Telomeric repeats shorten by 30-200 bp after each cycle of mitotic division and have been likened to a "molecular clock" reflecting the number of divisions a cell has undergone (Harley, 1997) and ultimately determines the replication potential of a cell. When telomeres shorten to a critical length, the cells undergo senescence, apoptosis or become genomically unstable. Shorter telomere length has previously been reported to be associated with an increased risk of various cancers (Avigad *et al.*, 2007; Broberg *et al.*, 2005; McGrath *et al.*, 2007; Risques *et al.*, 2007; Shao *et al.*, 2007; Shen *et al.*, 2007; Widmann *et al.*, 2007; Wu *et al.*, 2003).

Ultraviolet light B (UVB)-induced pyrimidine dimers occur at high frequency in telomere regions, indicating that long repeats of telomere sequences are targets for UVB (Jin and Ikushima, 2004). Although the precise mechanism is not known, it has been reported that UV-damaged telomeric DNA is less well-repaired than the transcriptionally active region (Kruk *et al.*, 1995). Unrepaired UVB lesions cause daughter-strand gaps during DNA replication (Wang and Smith, 1986), resulting in substantial loss of terminal telomere repeats or telomere shortening. In addition, UVA can indirectly cause oxidative stress via reactive oxygen species generated after the absorption of light energy by cellular chromophores (Brash, 1997; Hall and Johnson, 1996; Kielbassa *et al.*, 1997). Oxidative stress may lead to telomere shortening (Houben *et al.*, 2008). Truncated telomeres lead to genomic instability and are one of the defining characteristics of most carcinomas (Maser and DePinho, 2002).

The three major types of cells in the epidermis, melanocytes, basal keratinocytes, and squamous keratinocytes, give rise to melanoma, basal cell carcinoma (BCC), and squamous cell carcinoma (SCC), respectively. Melanocytes reside on the basal layer of the epidermis exhibiting less apoptotic activity and less differentiation than keratinocytes (Gilchrest *et al.*, 1999). A brief spurt of melanocyte proliferation gives rise to melanocytic nevi, benign tumors commonly referred to as moles, which are strongly associated with an increased risk of melanoma. Shorter telomere length may limit melanocytic nevus proliferation before entry into senescence and has been shown to be associated with decreased mole counts and size (Bataille *et al.*, 2007). On the other hand, squamous keratinocytes on the surface of the epidermis undergo continual cycles of proliferation and apoptosis throughout a lifetime. Basal cells, which give rise to BCC, are less susceptible to apoptosis than squamous keratinocytes (Gilchrest *et al.*, 1999) and have a lower tendency to senesce than melanocytes. Because of the distinct proliferative features of these skin cells, telomere length may play a different role in the etiology of these three types of skin cancers.

To this end, we prospectively evaluated the association between pre-diagnostically measured telomere length and risk of three types of skin cancers (melanoma, BCC, and SCC) simultaneously in a nested case-control study within the Nurses' Health Study.

## Results

### Descriptive characteristics of cases and controls

The characteristics of cases and controls in the skin cancer nested case-control study are presented in Table 1. Detailed description was published previously (Han *et al.*, 2006). In brief, at the beginning of the follow-up (at blood collection), the women were between 43 and 68 years old (mean age 58.7). The mean age at diagnosis of incident melanoma cases was 63.3 years, and that of SCC cases and BCC cases was 64.7 and 64.0 years, respectively. Risk factors for skin cancer included family history of skin cancer, sun exposure with a bathing suit, sunlamp use or tanning salon attendance, and lifetime sunburns. Melanoma cases were more likely to have a higher number of moles on the arms. Across all three types of skin cancers, cases were more likely to have light pigmentary phenotypes, including lighter skin and hair color, less tendency to tan, and increased tendency to burn. For mole counts among controls, the mean

was 5.1; the median was 1.0; and the 90% and 10% of the distribution were 15 and 0, respectively.

### Risk factors and telomere length

We observed that age at blood draw and pack-years of smoking were weakly inversely associated with telomere length among overall controls (Table 2). This is consistent with previous findings that telomeres shorten with age (Frenck *et al.*, 1998) and cigarette smoking due to oxidative stress (Valdes *et al.*, 2005). We also observed that there was a positive association between the number of moles on arms and telomere length ($r$ =0.17, P =0.002). The result was essentially the same after adjusting for age. In each quartile group of telomere length among overall controls, the mean age-adjusted number of moles was 4.4, 5.0, 5.3, and 5.8, respectively (P, trend, 0.09). In contrast, there was no association between telomere length and other inherited risk factors and sun exposure.

### Telomere length and skin cancer risk

We examined the association between relative telomere length and each type of skin cancer separately (Table 3). We categorized the participants into quartiles based on the relative telomere length distribution of the controls; the fourth quartile, the longest telomere length, served as the reference. We observed that shorter telomere length was associated with a decreased risk of melanoma. Women in the second and first quartiles, those with the shortest telomere length, had an OR for melanoma of 0.54 (95%CI, 0.29-1.01) and 0.59 (95% CI, 0.31-1.13) compared with those in the fourth quartile (P, trend = 0.09). In contrast, we observed that shorter telomere length was associated with an increased risk of BCC. Compared with women in the fourth quartile, those in the first quartile had an OR of 1.85 (95%CI, 0.94-3.62) (P, trend =0.09). No association was observed between telomere length and SCC risk. The results across the three types of skin cancers were essentially the same after controlling for the number of moles on the arms.

We did not observe a significant interaction between telomere length and the number of moles on skin cancer risk.

## Discussion

To our knowledge, this is the first prospective study addressing the potential contributions of relative telomere length to skin cancer risk. Relative telomere length measured pre-diagnostically in peripheral blood leukocytes (PBLs) was differentially associated with risk of three types of skin cancer (melanoma, BCC, and SCC) in women. These results were not statistically significant and should be interpreted with caution. Further studies are needed to confirm these possible findings.

Telomere shortening plays conflicting roles in cancer development. On one hand, the progressive loss of telomeric repeats with each cell division limits the total number of times a cell can divide. A DNA damage response is triggered once telomeres reach a critical length causing the cell to undergo senescence or apoptosis. Senescent cells express β-galactosidase, develop a characteristic flat and vacuole-rich cytoplasmic morphology, and undergo a permanent and irreversible cell cycle arrest in G1 phase, but remain metabolically active (Mooi and Peeper, 2006). This constraint on proliferation prevents the accumulation of oncogenic mutations and subsequent malignant transformation. However, if this checkpoint is bypassed, the cell continues to proliferate resulting in additional telomere erosion (Hackett and Greider, 2002; Wong and Collins, 2003). Once a threshold length of 12.8 repeats (77 base pairs) is reached, chromosome ends become fusogenic resulting in chromosome instability (Capper *et al.*, 2007), which may also potentially lead to malignant transformation.

The opposing associations observed between relative telomere length and the various skin cancers may stem from the proliferative characteristics of the corresponding cell types from which the cancers arise and how they deal with telomere shortening. Presumably cutaneous melanocytes are evolutionarily preserved because they produce melanin, which is responsible for constitutional skin pigmentation and protective tanning response to UV. They are characterized by low levels of proliferation and a limited capacity to undergo apoptosis, perhaps due to a high content of anti-apoptotic proteins, such as BCL2 and Slug (Mooi and Peeper, 2006). Frequently, activating mutations of the BRAF oncogene occur in melanoctyes, resulting in a transient increase in proliferation and the formation of melanocytic nevi (Gray-Schopfer *et al.*, 2006). Rather than undergo apoptosis, these melanocytes are more likely to senesce in response to oncogenic stress, which allows them to remain functional while preventing the propagation of the oncogenic mutation (Mooi and Peeper, 2006). It has been hypothesized that mutated cells with longer telomere lengths experience a delay in senescence as a result of the greater replication potential. Prolonged nevus development leads to the increased formation of nevi, which are strongly associated with an increased risk of melanoma (Bastian, 2003). Consistent with this hypothesis, we observed a positive association between relative telomere length and risk of melanoma.

In contrast to melanocytes, squamous keratinocytes are well differentiated cells that form the uppermost layer of skin. These cells have a lower apoptotic threshold making the apoptotic pathway the predominant protective mechanism, especially when cells are challenged by genotoxic stress such as UV-induced DNA damage. Sunburn cells are squamous keratinocytes undergoing apoptosis. Programmed cell death is also part of the normal terminal differentiation process that squamous keratinocytes undergo to form the water-resistant, outermost protective layer of the epidermis (Nemes and Steinert, 1999). This process is presumably independent of telomere length. As a result, it is not surprising that we did not observe an association between relative telomere length and SCC risk.

Basal cells, which give rise to BCC, are less susceptible to apoptosis than squamous keratinocytes (Gilchrest *et al.*, 1999) and have a lower tendency to senescence than melanocytes. The proliferative demand of basal cells coupled with UV-induced damage of telomeric DNA may increase the likelihood of unstable telomeres and trigger chromosomal rearrangements. Under these circumstances, cells with comparatively shorter telomere lengths may potentially reach genomic instability within a smaller number of cell divisions and therefore be at greater risk for malignant transformation. Such a mechanism may explain the association we observed between shorter relative telomere lengths and an increased risk of BCC in our study population.

While replicative senescence has been hypothesized to halt the initial growth of melanocytic nevi (Bastian, 2003), experimental evidence suggests otherwise. It is believed that activating mutations in BRAF, which are found in 82% of nevi (Pollock *et al.*, 2003), initiate the development of nevi with the onset of rapid proliferation. The proliferation is transient as it is generally followed by an increase in expression of p16, resulting in growth arrest and a senescent phenotype in a process known as oncogene-induced scenescence. Telomere attrition does not appear to be the main contributor to the initial growth arrest as telomere length in nevi did not differ significantly from surrounding tissue and nevi generally lack expression of p53 and p21, markers of telomere-induced senescence. These cells can remain arrested for decades (Gray-Schopfer *et al.*, 2006; Michaloglou *et al.*, 2005; Mooi and Peeper, 2006).

It has also been noted, that atypical nevi and early melanomas express p53 and p21 and that p16 is deleted or silenced in a majority of primary melanomas (Gray-Schopfer *et al.*, 2006). While telomere attrition may not play a role in the initial formation of melanocytic nevi, these observations suggest replicative senescence may act as a second line of defense in melanocytes.

If melanocytes eventually escape oncogene-induced senescence and reenter the cell cycle, cells with short telomeres will reach the critical length earlier causing cells to undergo replicative senescence or apoptosis, which may explain the inverse correlation between number of nevi and age in individuals over 30 years of age (Bataille *et al.*, 2007). Longer telomeres in melanocytes, which already have two oncogenic mutations (activating BRAF and silenced/deleted p16), may result in delayed senescence or apoptosis providing these cells with a greater opportunity to acquire additional mutations, increasing the probability of malignant transformation. The positive association we observed between relative telomere length and melanoma risk would be consistent with such a mechanism.

In this study, we used self-reported information on nevus counts. The validity of self-reported nevus counts was evaluated in previous studies. For example, the majority of studies on nevus counts have shown substantial agreement between nevus self-counts and dermatologist-counts (Buettner and Garbe, 2000; Little *et al.*, 1995; Melia *et al.*, 2000). The Spearman correlation coefficient was 0.91 between nevus counts by patients and physicians, suggesting quite a good correlation between the two measurements (Mikkilineni and Weinstock, 2000). Some case-control studies involving self-counts of nevi have demonstrated a substantial correlation of those counts with melanoma risk (Lawson *et al.*, 1994; Weinstock *et al.*, 1989). We did not have information on the total number of body moles and used the number of moles on the arms as a proxy for the total body mole counts. We previously examined moles on all four limbs versus one limb in relation to melanoma risk and found that the risks calculated from both calculations were comparable (Bain *et al.*, 1988).

We used real-time PCR and DNA derived from PBLs to determine relative telomere length. The observed correlation with age and cigarette smoking confirms that real-time PCR provides a biologically meaningful measure of telomere length. Measuring telomeres by real-time PCR is currently the most economical and versatile high-throughput method. It generates a T/S ratio that is proportional to a cell's average telomere length. Although the values are not an actual kilobase pair length of telomeres as do Southern Blots, the relative T/S ratio has been confirmed to be highly consistent with the Southern Blot assay, which measures telomere length as terminal restriction fragments (Cawthon, 2002). In fact, the coefficients of variation between replicates are often below 5% whereas densitometry readings from Southern Blots range between 10-15%. Additionally, the Real Time PCR method does not pick up subtelomeric DNA like Southern blots.

Even though relative telomere lengths were not directly measured in melanocytes and keratinocytes, PBLs represent a good proxy for telomere length in other tissues of the same individual, including skin (Friedrich *et al.*, 2000). The correlation between the PBL telomere length and mole counts observed in our study and the previous study (Bataille *et al.*, 2007) further suggest the biological relevance of the relative telomere length measure in the PBL.

In summary, our findings suggest the involvement of telomere length in the development of skin cancer. Distinct genetic constitutions across different types of cells and tissues evoke different DNA damage responses against telomere shortening and malignant transformation. Nevertheless, we cannot exclude the possibility that these findings may be due to chance. Further research is needed to confirm these possible associations.

## Materials and Methods

### Study population

The NHS was established in 1976, when 121,700 female registered nurses between the ages of 30 and 55, residing in 11 larger U.S. states, completed a self-administered questionnaire on their medical histories and baseline health-related exposures. Updated information has been

obtained by questionnaires every 2 years. Between 1989 and 1990, blood samples were collected from 32,826 of the cohort members. The distribution of risk factors for skin cancer in the subcohort of those who donated blood samples was very similar to that in the overall cohort (Han *et al.*, 2006). Eligible cases in this study were women with incident skin cancer from the subcohort who had given a blood specimen, including SCC and BCC cases with a diagnosis any time after blood collection up to June 1, 1998 and melanoma cases up to June 1, 2000 who had no previously diagnosed skin cancer. A common control series was randomly selected from participants who gave a blood sample and were free of diagnosed skin cancer up to and including the questionnaire cycle in which the case was diagnosed. One or two controls were matched to each case by year of birth (±1 year). All cases and controls were self-described as being of European ancestry. The nested case-control study consisted of 218 melanoma cases, 285 SCC cases, 300 BCC cases, and 870 matched controls. The study protocol was approved by the Committee on Use of Human Subjects of the Brigham and Women's Hospital, Boston, MA. The clinical investigation was conducted according to Declaration of Helsinki principles, and written informed consent was obtained from all patients.

## Exposure data

Information regarding skin cancer risk factors was obtained from the prospective biennial questionnaires and the retrospective supplementary questionnaire. Questions on natural hair color, childhood and adolescent tanning tendency, and childhood sunburn reaction were asked in the 1982 prospective questionnaire; the number of moles on arms (larger than 3 mm diameter) in the 1986 prospective questionnaire. In the skin cancer nested case-control study, natural skin color and other sun exposure-related information was collected by the retrospective supplementary questionnaire in 2002. Estimation of past sunlight exposure for each subject was described previously (Han *et al.*, 2006).

## Laboratory assays

Genomic DNA was extracted from buffy coat fractions using the QIAmp (Qiagen, Chatsworth, CA) 96-spin blood protocol. We conducted a pico-green quantitation using a Molecular Devices 96-well spectrophotometer and confirmed results using a Nanodrop SD-1000 spectrophotometer. Subsequent standardization by drying down the genomic DNA and resuspending ensured accurate and uniform DNA concentrations.

We employed a modified version of the Real-Time PCR telomere assay for use in a high-throughput 384-well format with an Applied Biosystems 7900HT PCR System to determine the relative average telomere length. Briefly, 5 ng of buffy-coat-derived genomic DNA was dried down in a 384-well plate and resuspended in 10μL of either the Telomere or 36B4 PCR reaction mixture for 2 hours at 4C. The telomere reaction mixture consisted of 1x Qiagen Quantitect Sybr Green Master Mix, 2.5mM of DTT, 270nM of Tel-1b primer (GGTTTTTGAGGGTGAGGGTGAGGGTGAGGGTGAGGGT), and 900nM of Tel-2b primer (TCCCGACTATCCCTATCCCTATCCCTATCCCTATCCCTA). The reaction proceeded for 1 cycle at 95C for 5:00, followed by 40 cycles at 95C for 0:15, and 54C for 2:00. The 36B4 reaction consisted of 1x Qiagen Quantitect Sybr Green Master Mix, 300nM of 36B4U primer (CAGCAAGTGGGAAGGTGTAATCC), and 500nM of 36B4D primer (CCCATTCTATCATCAACGGGTACAA). The 36B4 reaction proceeded for 1 cycle at 95C for 5:00, followed by 40 cycles at 95C for 0:15, and 58C for 1:10. All samples for both the Telomere and single-copy gene (36B4) reactions were performed in triplicate on different plates, and the threshold value for both reactions was set to 0.5. In addition to the samples, each 384-well plate contained a 6-point standard curve from 1.25ng to 30ng using pooled buffy coat-derived genomic DNA. The purpose of the standard curve was to assess and compensate for inter-plate variations in PCR efficiency. The slope of the standard curve for both the telomere and 36B4 reactions were -3.55 ± 0.15, and acceptable linear correlation coefficient

($R^2$) value for both reactions were 0.98 and 0.99, respectively. The T/S ratio (-dCt) for each sample was calculated by subtracting the average 36B4 Ct value from the average Telomere Ct value. The Relative T/S ratio (-ddCt) was determined by subtracting the T/S ratio value of the 5ng standard curve point from the T/S ratio of each unknown sample (Cawthon, 2002).

For the overall controls, the mean T/S ratio was 1.20 (median, 1.13) with standard deviation of 0.41. There were 143 blinded quality control samples interspersed throughout the dataset in order to assess inter-plate and intra-plate variability of threshold cycle (Ct) values. These quality controls were duplicated samples in 22 sets. The inter-assay coefficients of variation (CV) of the telomere and single-gene assay were 4.62% and 3.21%, respectively. The intra-assay CV of the telomere and single-gene assay were 3.02% and 2.07%, respectively.

### Statistical methods

Spearman correlation coefficients were calculated to assess the correlations between age, pack-years of smoking, other factors related to skin cancer, and telomere length among controls only. Spearman's partial correlation coefficients, adjusted for age, were used to assess the correlations.

We examined the association between telomere length and each type of skin cancer separately. We categorized telomere length into quartiles according to the distribution in each cancer-specific control population. We used conditional logistic regression to calculate odds ratios (OR) and 95% confidence intervals (95% CI). Tests for trend were conducted by assigning the median values for quartiles of telomere length among controls to both cases and controls as continuous variables.

To test statistical interactions between telomere length and the number of moles on skin cancer risk, we modeled both predictors as quartile ordinal variables to test the statistical significance of a single multiplicative interaction term. The *P* values are two-sided; *P* values <0.05 were considered statistically significant. We used the SAS Version 9.1 software (SAS Institute, Cary, NC).

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## Abbreviations

BCC, basal cell carcinoma; SCC, squamous cell carcinoma; CI, confidence interval; OR, odds ratio; UV, ultraviolet.

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**Table1**

Characteristics of skin cancer cases and controls in the nested case-control study

| Characteristic | Controls (n=870) | Melanoma cases (n=218) | SCC cases (n=285) | BCC cases (n=300) |
|---|---|---|---|---|
| Age at diagnosis, mean (SD) | 64.5 (7.0) | 63.3 (7.3) | 64.7 (7.0) | 64.0 (7.1) |
| Age at blood draw, mean (SD) | 58.8 (6.8) | 57.4 (7.0) | 59.3 (6.7) | 58.3 (6.6) |
| Family history of skin cancer (%) | 25.1 | 36.5 | 35.7 | 42.7 |
| Highest quartile of sun exposure with a bathing suit (%) | 33.4 | 53.3 | 46.1 | 42.6 |
| Sunlamp use or tanning salon attendance (%) | 10.0 | 19.2 | 14.3 | 14.7 |
| Number of lifetime severe sunburns, mean | 5.4 | 9.6 | 7.8 | 8.2 |
| Number of moles on arms, mean (SD) | 5.1 (10.3) | 11.3 (21.9) | 4.8 (12.0) | 7.7 (17.3) |
| Childhood tendency to tan (%) | 48.0 | 38.8 | 42.7 | 45.0 |
| Childhood tendency to burn (%) | 33.0 | 49.8 | 51.0 | 52.0 |
| Natural red or blonde hair color (%) | 12.5 | 23.3 | 20.3 | 20.7 |
| Natural fair skin color (%) | 39.9 | 57.1 | 54.6 | 53.0 |
| Pack year of smoking*, mean (SD) | 22.5 (19.0) | 20.9 (18.7) | 22.7 (18.0) | 22.9 (18.9) |

*Only among smokers

**Table 2**

Spearman correlation between telomere length and risk factors among controls

| | Coefficient (P value) |
|---|---|
| Age at blood draw | -0.09 (0.01) |
| Age at blood draw* | -0.09 (0.007) |
| Pack-years of smoking** | -0.05 (0.15) |
| Moles on arms | 0.17 (0.002) |
| Moles on arms** | 0.16 (0.003) |
| Natural hair color (black to red) | 0.02 (0.49) |
| Natural skin color (fair to black) | -0.02 (0.54) |
| Childhood tendency to tan | 0.02 (0.58) |
| Childhood tendency to burn | -0.03 (0.44) |
| Sun exposure while wearing a bathing suit** | -0.02 (0.62) |

* controlling for pack-years of smoking

** controlling for age at blood draw

## Table 3

Association between relative telomere length and skin cancer risk

| Melanoma | | | | |
|---|---|---|---|---|
| **Relative telomere length (T/S ratio)** [#] | **Case, n(%)** | **Control, n(%)** | **OR (95%CI)** [*] | **OR (95%CI)** [**] |
| **4th quartile (>1.40)** | 63 (30.9) | 55 (24.8) | 1.00 | 1.00 |
| **3rd quartile (1.15-1.40)** | 49 (24.0) | 54 (24.3) | 0.76 (0.43 - 1.34) | 0.79 (0.44 - 1.41) |
| **2nd quartile (0.97-1.15)** | 44 (21.6) | 58 (26.1) | 0.54 (0.29 - 1.01) | 0.57 (0.30 - 1.00) |
| **1st quartile (<0.97)** | 48 (23.5) | 55 (24.8) | 0.59 (0.31 - 1.13) | 0.62 (0.31 - 1.23) |
| | | P, trend | 0.09 | 0.15 |
| ***SCC*** | | | | |
| **Relative telomere length (T/S ratio)** | **Case, n(%)** | **Control, n(%)** | **OR (95%CI)** [*] | **OR (95%CI)** [**] |
| **4th quartile (>1.44)** | 61 (24.0) | 67 (24.5) | 1.00 | 1.00 |
| **3rd quartile (1.08-1.44)** | 78 (30.7) | 70 (25.6) | 1.24 (0.71 - 2.15) | 1.19 (0.67 - 2.12) |
| **2nd quartile (0.85-1.08)** | 59 (23.2) | 67 (24.5) | 0.90 (0.48 - 1.71) | 0.89 (0.46 - 1.72) |
| **1st quartile (<0.85)** | 56 (22.0) | 69 (25.3) | 0.73 (0.34 - 1.55) | 0.72 (0.33 - 1.55) |
| | | P, trend | 0.30 | 0.29 |
| **BCC** | | | | |
| **Relative telomere length (T/S ratio)** | **Case, n(%)** | **Control, n(%)** | **OR (95%CI)** [*] | **OR (95%CI)** [**] |
| **4th quartile (>1.52)** | 57 (20.2) | 75 (24.5) | 1.00 | 1.00 |
| **3rd quartile (1.17-1.52)** | 76 (27.0) | 79 (25.8) | 1.46 (0.85 - 2.52) | 1.49 (0.86 - 2.59) |
| **2nd quartile (0.90-1.17)** | 74 (26.2) | 76 (24.8) | 1.57 (0.86 - 2.87) | 1.61 (0.88 - 2.96) |
| **1st quartile (<0.90)** | 75 (26.6) | 76 (24.8) | 1.85 (0.94 - 3.62) | 1.87 (0.95 - 3.68) |
| | | P, trend | 0.09 | 0.09 |

[*] Conditional logistic regression

[**] Conditional logistic regression adjusted for the number of moles on arms

[#] Relative T/S ratio was described in detail in the text.