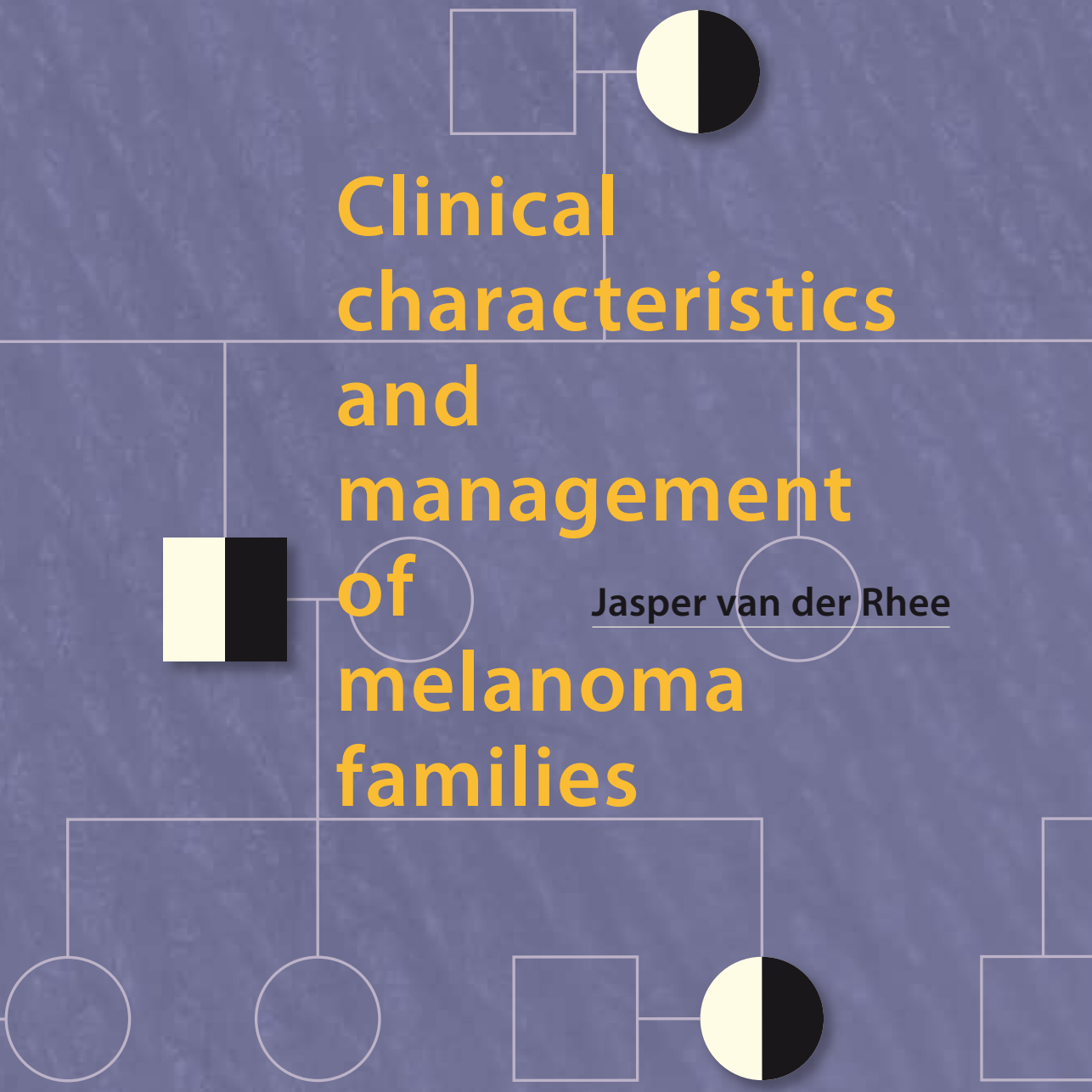

Clinical characteristics and management of melanoma families

Jasper van der Rhee

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Clinical characteristics and management of melanoma families

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Quiero hacer contigo
Lo que la primavera hace con los cerezos.

Uit "Twintig liefdesgedichten en een wanhoopslied"
Pablo Neruda (1904 – 1973)

Voor Carla, Lieke en Jonathan

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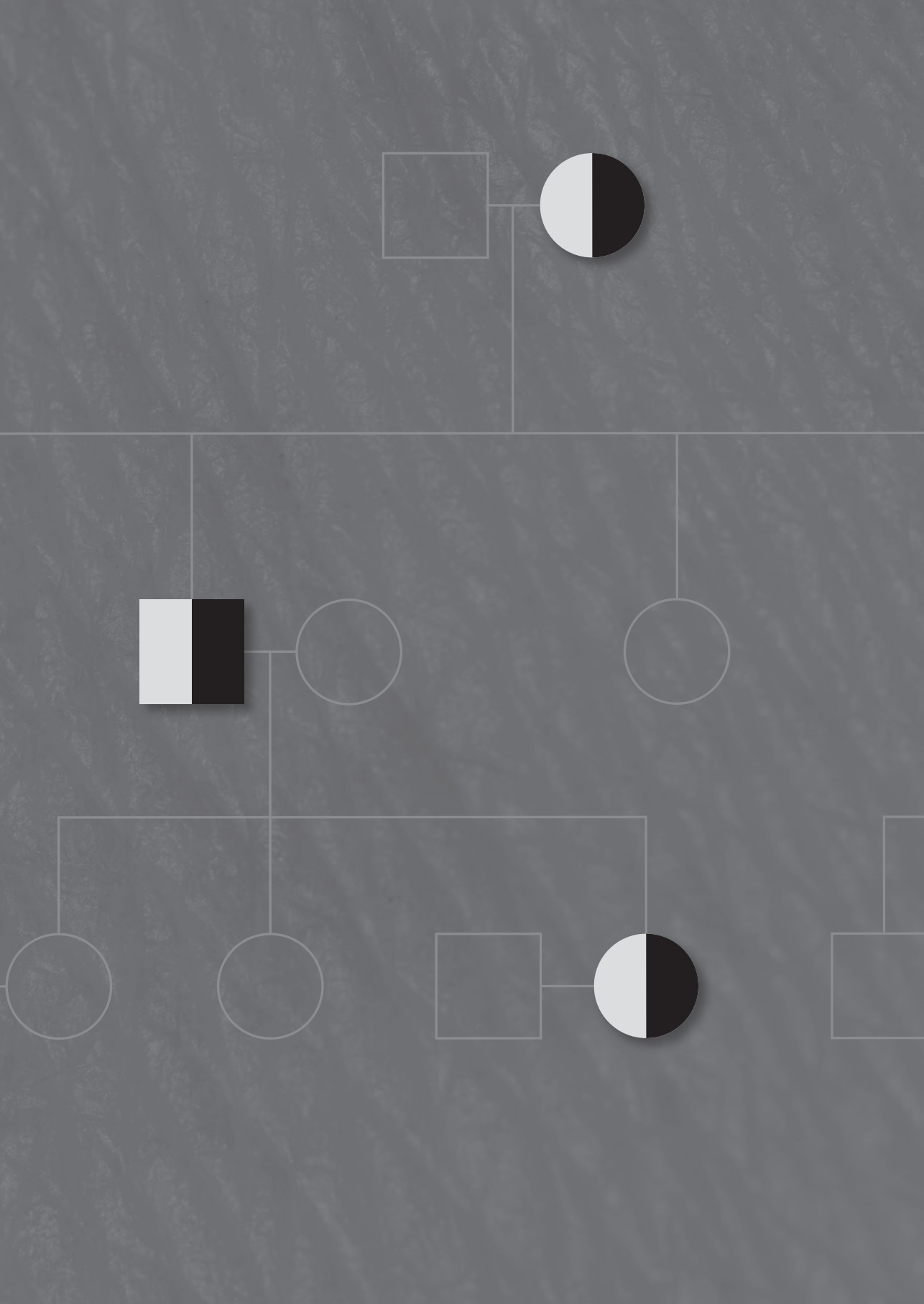

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General introduction

General introduction and outline of the thesis

Cutaneous malignant melanoma (CMM) is a malignant skin tumor that develops from melanocytes, the pigment producing cells, in the skin. In the last decades melanoma incidence rates have been increasing considerably worldwide. In the Netherlands the age-standardized incidence per 100.000 person-years increased from 11.3 in 1989 to 21.7 in 2008, with an estimated annual percentage increase of 4.1%.[1] By 2008 melanoma ranked the 8th most diagnosed cancer in man and 5th most diagnosed cancer in women.[2] Age-standardized mortality increased (albeit to a lesser extent), from 2.2 per 100.000 person-years in 1989 to 3.9 per 100.000 person-years in 2009, with an estimated annual percentage increase of 2.3%.[1]

One of the strongest risk factors for CMM is a familial predisposition. The characteristics and management of melanoma families are the subject of this thesis. This chapter will provide a background to the subject by discussing the epidemiology, clinical characteristics, and genetics of melanoma families, as well as the Dutch management guidelines that constituted the starting point for this thesis. In the final section, dermoscopy, which has been established as an integrated part of the everyday practice of skin surveillance, will be introduced. We will start however by providing a short overview of the different melanoma risk factors.

Melanoma risk factors

Several melanoma risk factors have been identified. Solar UV is the main environmental cause of melanoma.[3] At present it is known that the risk of melanoma is significantly increased by intermittent exposure: particularly irregular and intense exposure (with sunburn), while more regular (chronic) exposure is to some degree inversely associated with melanoma.[4] Recent studies suggest that chronic UV, intermittent UV and UV independent melanomas may represent (clinical, histological, epidemiological and molecular) different melanoma subtypes.[5]

Much of an individuals' risk of developing a CMM can be learned by inspection of the patients external characteristics. Hair colour (red vs. dark, relative risk of melanoma (RR) = 1.74 (1.41 – 2.14)), skin colour (fair vs. dark: RR = 2.06 (1.68 – 2.52)), and eye colour (blue vs. dark: RR = 1.47 (1.28 – 1.69)), are all associated with melanoma risk, most likely due to their correlation with sensitivity to ultraviolet light.[6] More strongly correlated with melanoma risk are the number of common nevi (RR = 6.89 (4.63 – 10.25) for 101-120 nevi vs. < 15) and number of atypical nevi (AN) (RR = 6.36 (3.80 – 10.33) for 5 vs. 0).[7] The risk of melanoma for patients with large congenital nevi is estimated to be about 2.5% to 5%, and highest in the first 5 to 10 years of life.[8]

The relative risk of melanoma for individuals with a positive family history of melanoma has been estimated to be 1.74 (1.41 – 2.14).[6] This risk increases considerably in families with many melanoma patients. An early study in 23 families with at least two family members

with CMM and two relatives with AN, reported a relative risk of CMM of 89 for relatives with AN and a relative risk of 229 for relatives with a previous CMM.[9] These high relative risks are illustrative of the fact that familial susceptibility is likely the strongest risk factor (in terms of effect size) for melanoma.

Familial melanoma

In 1820 Norris was the first to describe familial clustering of melanoma.[10] It took almost one and a half century before others published similar observations.[11] A positive family history of melanoma has been reported in 6% to 14% of melanoma patients.[12] Familial melanoma is defined as the occurrence of at least two first degree relatives with melanoma or three melanomas in second-degree relatives.[13] Because of the co-occurrence of AN in many of the first described melanoma families, the term Familial Atypical Multiple Mole-Melanoma (FAMMM) syndrome was adapted.[14] The correlation between AN and familial melanoma was later shown to be more complex however. AN regularly occur in the general population, with an estimated prevalence of 2% to 8% in whites.[15-17] In addition AN and CMM do not fully co-segregate within FAMMM families.[18] This has been well illustrated in families with a mutation in the high-penetrance melanoma susceptibility gene CDKN2A; relatives with AN have a higher probability of being CDKN2A mutation carrier, but mutation carriers may be devoid of AN and mutation negative relatives may have many AN.[19,20]

The genetics of melanoma susceptibility

So far, two high-penetrance melanoma susceptibility genes associated with an autosomal dominant inheritance have been identified. In 1994, CDKN2A(MIM# 600160), located in the 9p21 region, was the first melanoma susceptibility gene to be identified.[21] By using different first exons (1α and 1ß) respectively, it encodes two distinct proteins: p16INK4 and p14ARF. Both proteins are tumour suppressors involved in cell cycle regulation.

Pathogenic germline mutations in the tumor suppressor gene CDKN2A are detected in approximately 39% of families with ≥3 melanoma cases.[22] CDKN2A mutations have an estimated penetrance for CMM of 67% by the age of 80 years.[23] In the Netherlands the most prevalent CDKN2A germline mutation is a founder mutation (c.225-243del19) that is frequently found in the Leiden region, and has therefore been denominated the p16-Leiden mutation.[24] In addition to an increased melanoma risk, the p16-Leiden mutation is associated with a cumulative risk of pancreatic cancer of 17% by age 75.[25]
Mutations in the second high penetrance melanoma susceptibility gene, the oncogene CDK4 (MIM# 123829) have been detected only in few families (estimated 2%).[26] CDK4 germline mutations have a similar impact on melanoma risk as CDKN2A mutations.[27]
For the majority of families the genetic risk factor has not been fully clarified, but appears

to be the result of a combination of low (e.g. MC1R) and moderate (e.g. MITF) risk modifier genes and (possibly) some rare high penetrance genes.[28,29] Environmental and lifestyle factors (as described above) likely attribute to clustering of melanoma in (some) families, and modify expression of genetic risk factors.

In clinical genetics a distinction is made between families with a proven germline mutation (CDKN2A/CDK4): 'hereditary melanoma' and families without a (proven) germline mutation: 'familial melanoma'. In families without a proven pathogenic germline mutation, melanoma susceptibility is suggested by familial clustering of melanoma patients, but cannot be confirmed by genetic testing and therefore remains a clinical diagnosis based on pedigree studies alone.

Clinical characteristics of melanoma patients from melanoma families

Several studies indicate that melanoma patients from melanoma families have an earlier age of onset and an increased risk of multiple primary melanomas (MPM). There are also reports on a different distribution of histological tumour types in melanoma families; i.e. an increased proportion of superficial spreading melanomas, decreased proportion of nodular melanomas and a possible absence of acral lentiginous melanomas.[30-32] Data on the characteristic of melanoma patients specifically from families with a CDKN2A mutation are more scarce however. Reports on the age of diagnosis of the first melanoma in these families range from 36.3 to 43.3 years.[27,33,34] The proportion of mutation carriers who develop MPM in the literature ranges from 18.6% to 25.6%, but duration of follow-up was not reported in the referred studies.[33,35]

In **chapter 2** we report a study in which we investigated the clinical and histological characteristics of melanoma(patients) from CDKN2A mutated families in comparison with sporadic melanoma patients. In **chapter 4** some of the characteristics reported in chapter 2 were compared between CDKN2A mutated families and CDKN2A wild-type melanoma families.

Management

Melanoma patient survival is highly dependend on the stage at diagnosis. Early melanomas can be cured by a wide local excision with proper resection margins, but, even though promising new therapeutic options are emerging, prognosis is still poor for advanced disease.[36-38]

Survival outcomes are to a considerable extent predictable based on the histological characteristics of the primary tumor, and the presence of lymph node involvement (including sentinel node procedure) and (distant) metastases. One of the main histological predictive characteristics, first described by Alexander Breslow in 1970 is the tumor

(Breslow) thickness, which is the depth of the tumor from the surface of the lesion (stratum granulosum) to the deepest point of invasion, expressed in millimeters.[39] Breslow thickness strongly correlates with survival, which is illustrated by the fact that 10-year survival is 92% in case of melanomas ≤ 1.00 mm, and 50% if > 4.00 mm. The presence of ulceration and the mitotic rate are additional tumor characteristics correlated with outcome.[40]

Early detection is considered the most effective way to prevent melanoma mortality. For this reason, regular surveillance of individuals at high risk of melanoma, such as members of melanoma families, is widely advocated.[41,42]

Effectiveness of melanoma surveillance

In a 2009 review, the U.S. Preventive Services Task Force (USPSTF) reinforced their 2001 statement that, quote: "evidence is lacking that skin examinations by physicians is effective in reducing mortality or morbidity from skin cancer.".[43,44] This statement was predicated on the lack of evidence from randomized controlled studies. Although this statement addressed skin cancer (including squamous- and basal cell carcinoma) screening of the general population, the same argument is brought forward for specific melanoma screening or surveillance. Given the fact that an adequately powered, population-based randomized controlled trial of screening demonstrating mortality outcomes would require approximately 800.000 participants (based on US melanoma-related mortality rate), it is unlikely however that a randomized controlled study will ever be conducted.[42,43]

Several studies have reported that melanomas detected by physicians have a thinner Breslow thickness than those detected by patients themselves.[45-50] A few studies have reported the detection of thinner melanomas in the context of surveillance of melanoma families.[51-54] Recently convincing arguments for a beneficial effect of screening on melanoma survival came from an observational study concerning a population-based skin cancer screening project in Schleswig-Holstein, Germany, reporting a significantly different and more favourable trend in mortality rates compared to adjacent regions in the years following the screening period.[55]

In a recent article the Melanoma Prevention Working Group commented on the USPSTF statement that, quote: "…the evidence is compelling enough to support the efficacy of targeted screening programs for detecting thinner melanomas, as a proxy measure for reduced mortality.", and, quote: "…absolute proof is not necessary in the public health domain to implement a targeted screening program that has the immediate potential to save lives.".[42]

As noted above the effectiveness of surveillance in melanoma families has been investigated only in a few studies, mostly with limited numbers of surveillance detected melanomas and confined to specialized pigmented lesion clinics.[51-54] In **chapter 3** we investigated the effectiveness of surveillance in CDKN2A mutated families and also

address some issues related to the effectiveness of surveillance that have gained little attention so far in the literature. In **chapter 4** effectiveness of surveillance was assessed in families registered at the NFDHT, that were under surveillance throughout the Netherlands.

Management of melanoma families in the Netherlands

In the Netherlands, the first surveillance program for familial melanoma was initiated at the Leiden University Medical Center (LUMC) in 1981. Individuals that were invited to the program encompassed melanoma patients, their first degree relatives (parents, siblings and children) as well as their second degree relatives (grandparents, uncles, aunts, nieces, nephews and grandchildren). Starting from the age 12, these relatives are offered (a minimum of) annual total skin examinations.

In 1989 a national registry for familial melanoma was established at the Netherlands Foundation for the Detection of Hereditary Tumors (NFDHT) in order to promote the detection and surveillance of members of melanoma families throughout the Netherlands. Clinicians refer families suspected for familial melanoma to the registry. Genealogical studies are performed and all reported malignancies are verified by medical records. If criteria for familial melanoma are met, the registry monitors the continuity of the surveillance program for all family members with a history of melanoma and their unaffected first degree relatives by annually sending letters to the responsible clinician (mostly dermatologists). In return these clinicians report the results of surveillance and histo-pathologic examination.[54,56]

Starting from 2000 (predictive) DNA testing for CDKN2A (later complemented by CDK4) became available for members of melanoma families.

The segments of the 2005 Dutch melanoma guidelines that cover the management of melanoma families, and that were in effect when this thesis was initiated, are presented in Box 1.

As can be seen in box 1, the surveillance recommendations for FAMMM families in the 2005 guidelines were similar for all families that fulfilled the criteria of at least two first-degree relatives or three melanomas in second-degree relatives. Surveillance recommendations were independent of the presence or absence of a germline mutation in CDKN4/CDK4 in the family and family characteristics (e.g. the number of affected relatives). Given the fact that the chance of CDKN2A mutation detection was proven to be positively correlated with the number of melanoma patients in a family, it is anticipated that melanoma risk is higher in families with a high penetrance melanoma susceptibility gene-mutation (CDKN2A/CDK4) compared to CDKN2A/CDK4 wild-type families.[22] In addition, it is expected that melanoma risk in families is positively correlated with the number of melanoma patients. There is a lack of prospective studies however that confirm these notions, and it is therewith unclear whether all melanoma families need to be surveillanced with the same scrutiny.

Box 1 Guidelines with respect to the management of melanoma families from the Dutch Melanoma guidelines 2005 (appendix 3)[13]

Paragraph 3.1: Familial Dysplastic Nevus Syndrome (DNS)
(= FAMMM syndrome = Familial Atypical Multiple Mole / Melanoma Syndrome)

Melanoma with/without dysplastic melanocytic nevi nevocellulares in at least two first-degree relatives or three melanomas in second-degree relatives.

Note: Presence of dysplastic melanocytic nevi increases the probability of being a mutation carrier, but absence does not exclude being a carrier of the predisposition to melanoma.

Paragraph 5.2:
Risk level 2 (greatly increased):

Being a members of a family (up to the second degree) with familial DNS / FAMMM syndrome (see section 3.1).

management:
- Information (oral and written)
- Once a year or more frequent skin examinations (absolute indication)
- Check children from the age of twelve years

Another point of discussion has been the necessity for second degree relatives to be under surveillance, as the yield of surveillance may be relatively small. Surveillance of second degree relatives has not been explicitly recommended in melanoma guidelines from other countries.[57-59] In this thesis we present two studies that investigated melanoma detection rates in families with different CDKN2A mutation status and family characteristics (**chapter 4**) and in second degree relatives from CDKN2A mutated families (**chapter 5**).

'Overdiagnosis'

A recurrent point of discussion related to melanoma surveillance is the issue of 'misclassification' of benign or indolent melanocytic proliferations as CMM. This point gained attention upon the observation that the incidence of melanoma has increased dramatically, while the mortality from melanoma has not increased proportionately. In addition, the increased melanoma incidence is disproportionally attributable to thin lesions.[1,60] It has been argued that the 'melanoma epidemic' could (at least partially) be explained by increased public and physicians' melanoma awareness and screening/surveillance, which resulted in three (overlapping) phenomena; 1. overdiagnosis, i.e. detection of indolent melanocytic tumors, that would either never progress or progress slowly enough that the patient dies of other

causes, and 2. diagnostic drift, i.e. classification of melanocytic lesions as CMM, that years ago would have been diagnosed as benign melanocytic lesions, and 3. increase in false positives as a result of submission of increasing numbers of equivocal melanocytic proliferations to the pathologist.[60-62] Adversaries of this line of argumentation claim that, in stead, the epidemiological trends are mainly attributable to a real and steep increase in melanoma incidence due to behavioural changes regarding sun exposure. At the same time, mortality is believed to have been successfully constrained by strategies to advance melanoma diagnosis. The debate is ongoing.[63] In **chapter 6** we report an observation concerning the misclassification of melanocytic lesions as melanoma in the context of surveillance of CDKN2A mutated families.

Dermoscopy

Dermoscopy is a non-invasive technique in which oil immersion or polarised light are used to make the epidermis translucent and a lens is used for magnification to allow the visualization of structures not visible to the naked eye. Although the basic technique was already described in the late 19th century, it was not until the last two decades of the 20th century, after the introduction of handheld dermatoscopes, that dermoscopy gradually became integrated in the dermatological armamentarium.[64], Dermoscopy is primarily used to supplement the clinical (naked eye) evaluation of (pigmented) skin lesions, that are suspicious of malignancy. The basic approach to these lesions consists of two steps: 1. to distinguish melanocytic from non-melanocytic lesions, and 2. distinguish benign from malignant lesions. Several algorithms have been developed to facilitate a standardized assessment of (pigmented) lesions, including the pattern analysis, and more accessible simplified algorithms like the ABCD-method, Menzies method and seven point checklist.[65-68]

The impact of dermoscopy on clinical practice

Numerous studies have confirmed that dermoscopy improves the diagnostic accuracy for pigmented lesions.[80-92] In 2008 a meta-analysis of dermoscopy studies performed in a clinical setting, reported a statistically significant better sensitivity for the diagnosis of melanoma for dermoscopy (0.90) compared to naked eye examination alone (0.71), without a significant difference in specificity (dermoscopy: 0.90, naked eye examination: 0.81).[69] Strikingly, the findings of the only randomized controlled study on dermoscopy in a dermatologist setting, presented a rather opposite view; a 42% reduction in patients referred to excision, without a change in sensitivity.[70]

These contradictory findings may be related to the fact that the design of many dermoscopy studies possibly limited their applicability to clinical practice: clinicians judged (macro- and dermoscopic) images rather than life patients, study sets included only lesions that had been excised, and contained a disproportionate high number of

melanomas, dermoscopic images were not preceded by their accompanying macroscopic images, studies focussed on the impact of dermoscopy on the clinical (preferential) diagnosis, rather than management of lesions and many studies were performed in the setting of dermoscopy expert dermatologists. Performance of dermoscopy is likely to be highly dependent on the clinical context in which it is performed. As a consequence the actual impact of dermoscopy on the clinical dermatological practice is not fully clarified.
In **chapter 7** and **chapter 8** we describe two studies in which we investigated the impact of dermoscopy on clinical practice both in general dermatology clinics as well as in the context of surveillance of melanoma families in an expert pigmented lesion clinic. These two clinic settings are expected to differ both in respect to the characteristics of the presented lesions (symptomatic lesions versus early asymptomatic lesions against the background of atypical nevi) as to the degree of dermoscopy expertise.

Aims and outline of the thesis

The general aims of this thesis are threefold. Firstly, we aimed to verify and substantiate the clinical and histological characteristic of melanoma (patients) from melanoma families with a pathogenic germline mutation in CDKN2A. Secondly, we aimed to investigate the effectiveness and yield of surveillance of melanoma families with different CDKN2A mutation status and family characteristics, and to identify possible causes for failure of surveillance. Thirdly, we aimed to investigate the impact of dermoscopy on the management of suspicious lesions in relatives from melanoma families under surveillance in a tertiary pigmented lesion clinic.

Chapter 2 investigates the clinical and histological characteristics of melanoma (patients) from families with a germline mutation in CDKN2A in comparison to sporadic melanoma (patients)
Chapter 3 investigates the effectiveness of surveillance by comparison of tumour thickness of surveillance detected cases with pre-surveillance detected cases in CDKN2A mutated families. Mode of detection and length of the surveillance interval are analyzed to identify possible causes for failure of surveillance.
Chapter 4 investigates the effectiveness of surveillance in families registered at the NFDHT. The yield of surveillance in families with different family characteristics- and CDKN2A mutation status was investigated by estimation of the melanoma detection rates.
Chapter 5 investigates the yield of surveillance of second degree relatives from families with a founder mutation in CDKN2A by estimating the melanoma detection rate and studying the family dynamics of two- to first-degree relative transitions.
Chapter 6 reports an observation related to the issue of 'overdiagnosis' in the context of surveillance of CDKN2A mutated families.

Chapter 7 investigates the impact of dermoscopy on the preferential diagnosis and management decisions towards suspicious pigmented lesions in every day clinical practice of general dermatologists. This chapter is intended as a background for the findings reported in chapter 8.

Chapter 8 investigates the impact of dermoscopy on the preferential diagnosis and management decisions of suspicious pigmented lesions in high-risk patients from melanoma families.

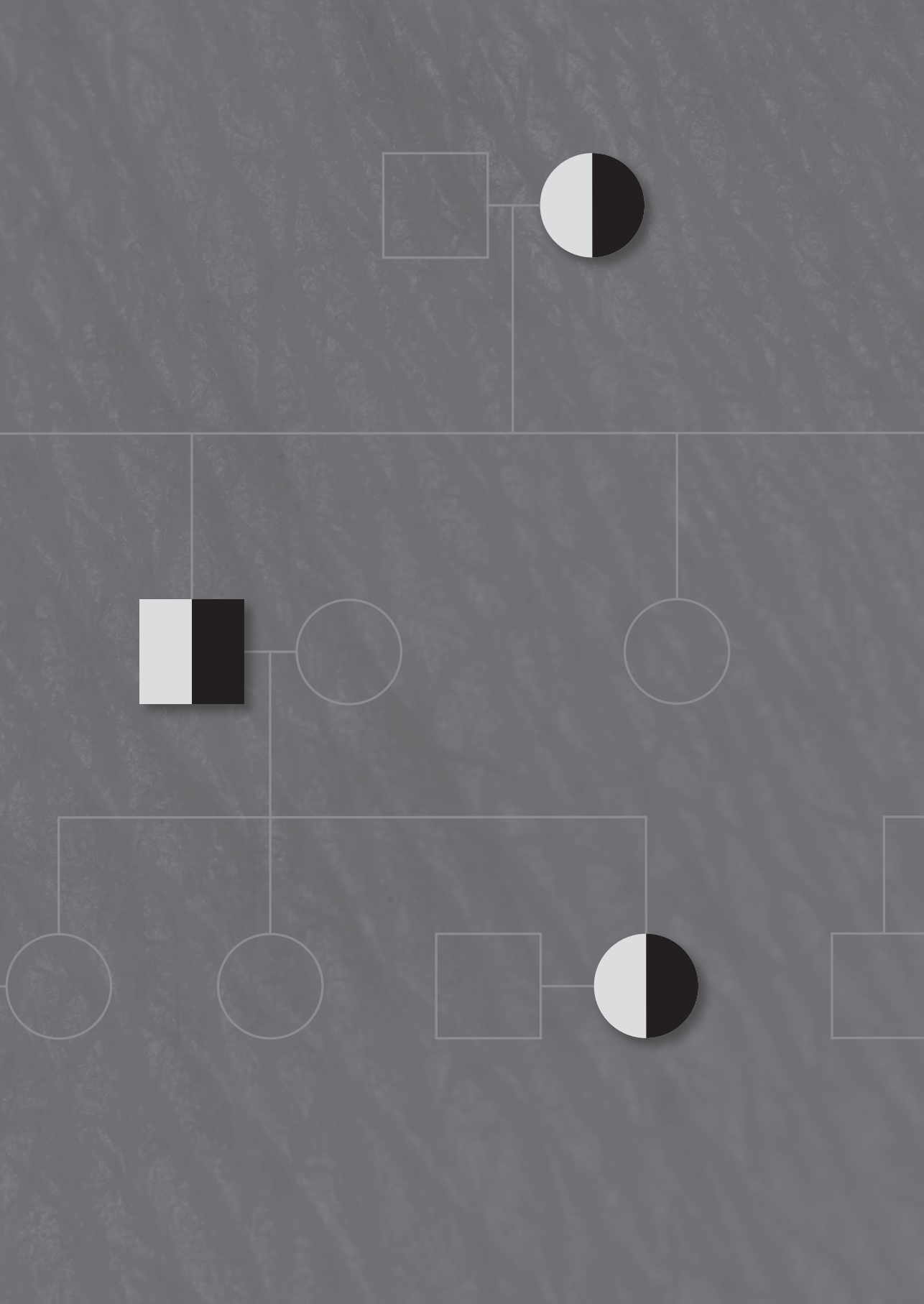
Chapter 9 summarizes and discusses the findings described in the preceding chapters.

Reference List

1. Hollestein LM, van den Akker SA, Nijsten T, Karim-Kos HE, Coebergh JW, de VE. Trends of cutaneous melanoma in The Netherlands: increasing incidence rates among all Breslow thickness categories and rising mortality rates since 1989. Ann.Oncol. 2012;23:524-30.
2. Integraal Kanker Centrum. Meest voorkomende kankersoorten in Nederland. http://www.cijfersoverkanker.nl/selecties/dataset_1/img507fdb687c352 (access date oktober 18 2012).
3. International Agency on the Research of Cancer. LARC Monographs on the evalutation of carcinogenic risks to humans: Solar and ultraviolet radiation. 55. 1992. Lyon, France.
4. Gandini S, Sera F, Cattaruzza MS, Pasquini P, Picconi O, Boyle P et al. Meta-analysis of risk factors for cutaneous melanoma: II. Sun exposure. Eur.J.Cancer 2005;41:45-60.
5. Whiteman DC, Pavan WJ, Bastian BC. The melanomas: a synthesis of epidemiological, clinical, histopathological, genetic, and biological aspects, supporting distinct subtypes, causal pathways, and cells of origin. Pigment Cell Melanoma Res. 2011;24:879-97.
6. Gandini S, Sera F, Cattaruzza MS, Pasquini P, Zanetti R, Masini C et al. Meta-analysis of risk factors for cutaneous melanoma: III. Family history, actinic damage and phenotypic factors. Eur.J.Cancer 2005;41:2040-59.
7. Gandini S, Sera F, Cattaruzza MS, Pasquini P, Abeni D, Boyle P et al. Meta-analysis of risk factors for cutaneous melanoma: I. Common and atypical naevi. Eur.J.Cancer 2005;41:28-44.
8. Shah KN. The risk of melanoma and neurocutaneous melanosis associated with congenital melanocytic nevi. Semin.Cutan.Med Surg 2010;29:159-64.
9. Tucker MA, Fraser MC, Goldstein AM, Elder DE, Guerry D, Organic SM. Risk of melanoma and other cancers in melanoma-prone families. J Invest Dermatol. 1993;100:350S-5S.
10. Norris, W. A case of fungoid disease. Edinb Med Surg J 16, 562-565. 1820.
11. CAWLEY EP, KRUSE WT, PINKUS HK. Genetic aspects of malignant melanoma. AMA.Arch.Derm.Syphilol. 1952;65:440-50.
12. Ang CG, Kelly JW, Fritschi L, Dowling JP. Characteristics of familial and non-familial melanoma in Australia. Melanoma Res. 1998;8:459-64.
13. Dutch Melanoma Society. *Guidelines Melanoma of the skin.* Alphen aan den Rijn: van Zuiden, 2005.
14. Lynch HT, Frichot BC, III, Lynch JF. Familial atypical multiple mole-melanoma syndrome. J.Med.Genet. 1978;15:352-6.
15. Crutcher WA, Sagebiel RW. Prevalence of dysplastic naevi in a community practice. Lancet 1984;1:729.
16. Lee G, Massa MC, Welykyj S, Choo J, Greaney V. Yield from total skin examination and effectiveness of skin cancer awareness program. Findings in 874 new dermatology patients. Cancer 1991;67:202-5.
17. Nordlund JJ, Kirkwood J, Forget BM, Scheibner A, Albert DM, Lerner E et al. Demographic study of clinically atypical (dysplastic) nevi in patients with melanoma and comparison subjects. Cancer Res. 1985;45:1855-61.
18. Clark WH, Jr., Reimer RR, Greene M, Ainsworth AM, Mastrangelo MJ. Origin of familial malignant melanomas from heritable melanocytic lesions. 'The B-K mole syndrome'. Arch.Dermatol. 1978;114:732-8.
19. Bishop JA, Wachsmuth RC, Harland M, Bataille V, Pinney E, MacK P et al. Genotype/phenotype and penetrance studies in melanoma families with germline CDKN2A mutations. J.Invest Dermatol. 2000;114:28-33.
20. Goldstein AM, Martinez M, Tucker MA, Demenais F. Gene-covariate interaction between dysplastic nevi and the CDKN2A gene in American melanoma-prone families. Cancer Epidemiol.Biomarkers Prev. 2000;9:889-94.
21. Kamb A, Gruis NA, Weaver-Feldhaus J, Liu Q, Harshman K, Tavtigian SV et al. A cell cycle regulator potentially involved in genesis of many tumor types. Science 1994;264:436-40.
22. Goldstein AM, Chan M, Harland M, Hayward NK, Demenais F, Bishop DT et al. Features associated with germline CDKN2A mutations: a GenoMEL study of melanoma-prone families from three continents. J.Med.Genet. 2007;44:99-106.
23. Bishop DT, Demenais F, Goldstein AM, Bergman W, Bishop JN, Bressac-de Paillerets B et al. Geographical variation in the penetrance of CDKN2A mutations for melanoma. J.Natl.Cancer Inst. 2002;94:894-903.
24. Gruis NA, van der Velden PA, Sandkuijl LA, Prins DE, Weaver-Feldhaus J, Kamb A et al. Homozygotes for CDKN2 (p16) germline mutation in Dutch familial melanoma kindreds. Nat.Genet. 1995;10:351-3.

25. de Snoo FA, Bishop DT, Bergman W, van L, I, van der DC, van Nieuwpoort FA et al. Increased risk of cancer other than melanoma in CDKN2A founder mutation (p16-Leiden)-positive melanoma families. Clin.Cancer Res. 2008;14:7151-7.
26. Goldstein AM, Chan M, Harland M, Gillanders EM, Hayward NK, Avril MF et al. High-risk melanoma susceptibility genes and pancreatic cancer, neural system tumors, and uveal melanoma across GenoMEL. Cancer Res. 2006;66:9818-28.
27. Goldstein AM, Struewing JP, Chidambaram A, Fraser MC, Tucker MA. Genotype-phenotype relationships in U.S. melanoma-prone families with CDKN2A and CDK4 mutations. J.Natl.Cancer Inst. 2000;92:1006-10.
28. Udayakumar D, Mahato B, Gabree M, Tsao H. Genetic determinants of cutaneous melanoma predisposition. Semin.Cutan.Med.Surg. 2010;29:190-5.
29. Yokoyama S, Woods SL, Boyle GM, Aoude LG, MacGregor S, Zismann V et al. A novel recurrent mutation in MITF predisposes to familial and sporadic melanoma. Nature 2011;480:99-103.
30. Greene MH, Clark WH, Jr., Tucker MA, Kraemer KH, Elder DE, Fraser MC. High risk of malignant melanoma in melanoma-prone families with dysplastic nevi. Ann.Intern.Med. 1985;102:458-65.
31. Kopf AW, Hellman LJ, Rogers GS, Gross DF, Rigel DS, Friedman RJ et al. Familial malignant melanoma. JAMA 1986;256:1915-9.
32. Nagore E, Botella-Estrada R, Garcia-Casado Z, Requena C, Serra-Guillen C, Llombart B et al. Comparison between familial and sporadic cutaneous melanoma in Valencia, Spain. J.Eur.Acad.Dermatol.Venereol. 2008.
33. Mantelli M, Barile M, Ciotti P, Ghiorzo P, Lantieri F, Pastorino L et al. High prevalence of the G101W germline mutation in the CDKN2A (P16(ink4a)) gene in 62 Italian malignant melanoma families. Am.J.Med.Genet. 2002;107:214-21.
34. Masback A, Olsson H, Westerdahl J, Sandberg T, Borg A, Jonsson N et al. Clinical and histopathological features of malignant melanoma in germline CDKN2A mutation families. Melanoma Res. 2002;12:549-57.
35. Borg A, Sandberg T, Nilsson K, Johannsson O, Klinker M, Masback A et al. High frequency of multiple melanomas and breast and pancreas carcinomas in CDKN2A mutation-positive melanoma families. J.Natl. Cancer Inst. 2000;92:1260-6.
36. Chapman PB, Hauschild A, Robert C, Haanen JB, Ascierto P, Larkin J et al. Improved survival with vemurafenib in melanoma with BRAF V600E mutation. N.Engl.J Med 2011;364:2507-16.
37. Hodi FS, O'Day SJ, McDermott DF, Weber RW, Sosman JA, Haanen JB et al. Improved survival with ipilimumab in patients with metastatic melanoma. N.Engl.J Med 2010;363:711-23.
38. Livingstone E, Zimmer L, Vaubel J, Schadendorf D. Current advances and perspectives in the treatment of advanced melanoma. J Dtsch.Dermatol.Ges. 2012;10:319-25.
39. Breslow A. Thickness, cross-sectional areas and depth of invasion in the prognosis of cutaneous melanoma. Ann.Surg 1970;172:902-8.
40. Balch CM, Gershenwald JE, Soong SJ, Thompson JF, Atkins MB, Byrd DR et al. Final version of 2009 AJCC melanoma staging and classification. J.Clin.Oncol. 2009;27:6199-206.
41. Katalinic A, Waldmann A, Weinstock MA, Geller AC, Eisemann N, Greinert R et al. Does skin cancer screening save lives?: An observational study comparing trends in melanoma mortality in regions with and without screening. Cancer 2012.
42. Curiel-Lewandrowski C, Chen SC, Swetter SM. Screening and Prevention Measures for Melanoma: Is There a Survival Advantage? Curr.Oncol.Rep. 2012;14:458-67.
43. Wolff T, Tai E, Miller T. Screening for skin cancer: an update of the evidence for the U.S. Preventive Services Task Force. Ann.Intern.Med. 2009;150:194-8.
44. Screening for skin cancer: recommendations and rationale. Am.J Prev.Med 2001;20:44-6.
45. Koh HK, Miller DR, Geller AC, Clapp RW, Mercer MB, Lew RA. Who discovers melanoma? Patterns from a population-based survey. J Am.Acad.Dermatol. 1992;26:914-9.
46. Epstein DS, Lange JR, Gruber SB, Mofid M, Koch SE. Is physician detection associated with thinner melanomas? JAMA 1999;281:640-3.
47. Brady MS, Oliveria SA, Christos PJ, Berwick M, Coit DG, Katz J et al. Patterns of detection in patients with cutaneous melanoma. Cancer 2000;89:342-7.
48. McPherson M, Elwood M, English DR, Baade PD, Youl PH, Aitken JF. Presentation and detection of invasive melanoma in a high-risk population. J Am.Acad.Dermatol. 2006;54:783-92.

49. Geller AC, Elwood M, Swetter SM, Brooks DR, Aitken J, Youl PH et al. Factors related to the presentation of thin and thick nodular melanoma from a population-based cancer registry in Queensland Australia. Cancer 2009;115:1318-27.
50. Kovalyshyn I, Dusza SW, Siamas K, Halpern AC, Argenziano G, Marghoob AA. The impact of physician screening on melanoma detection. Arch.Dermatol. 2011;147:1269-75.
51. Carey WP, Jr., Thompson CJ, Synnestvedt M, Guerry D, Halpern A, Schultz D et al. Dysplastic nevi as a melanoma risk factor in patients with familial melanoma. Cancer 1994;74:3118-25.
52. Hansson J, Bergenmar M, Hofer PA, Lundell G, Mansson-Brahme E, Ringborg U et al. Monitoring of kindreds with hereditary predisposition for cutaneous melanoma and dysplastic nevus syndrome: results of a Swedish preventive program. J.Clin.Oncol. 2007;25:2819-24.
53. Masri GD, Clark WH, Jr., Guerry D, Halpern A, Thompson CJ, Elder DE. Screening and surveillance of patients at high risk for malignant melanoma result in detection of earlier disease. J.Am.Acad.Dermatol. 1990;22:1042-8.
54. Vasen HF, Bergman W, van Haeringen A, Scheffer E, van Slooten EA. The familial dysplastic nevus syndrome. Natural history and the impact of screening on prognosis. A study of nine families in the Netherlands. Eur.J.Cancer Clin.Oncol. 1989;25:337-41.
55. Katalinic A, Waldmann A, Weinstock MA, Geller AC, Eisemann N, Greinert R et al. Does skin cancer screening save lives?: An observational study comparing trends in melanoma mortality in regions with and without screening. Cancer 2012.
56. Vasen HF, Gruis NA, Frants RR, Der Velden PA, Hille ET, Bergman W. Risk of developing pancreatic cancer in families with familial atypical multiple mole melanoma associated with a specific 19 deletion of p16 (p16-Leiden). Int.J.Cancer 2000;87:809-11.
57. Bichakjian CK, Halpern AC, Johnson TM, Foote HA, Grichnik JM, Swetter SM et al. Guidelines of care for the management of primary cutaneous melanoma. American Academy of Dermatology. J.Am.Acad.Dermatol. 2011;65:1032-47.
58. Marsden JR, Newton-Bishop JA, Burrows L, Cook M, Corrie PG, Cox NH et al. Revised U.K. guidelines for the management of cutaneous melanoma 2010; Br.J.Dermatol. 2010;163:238-56.
59. Clinical Practice Guidelines for the Management of Melanoma in Australia and New Zealand (2008) http://www.nhmrc.gov.au/_files_nhmrc/publications/attachments/cp111.pdf (access date oktober 18 2012)
60. Swerlick RA, Chen S. The melanoma epidemic. Is increased surveillance the solution or the problem? Arch. Dermatol. 1996;132:881-4.
61. Levell NJ, Beattie CC, Shuster S, Greenberg DC. Melanoma epidemic: a midsummer night's dream? Br.J Dermatol. 2009;161:630-4.
62. Welch HG, Black WC. Overdiagnosis in cancer. J Natl.Cancer Inst. 2010;102:605-13.
63. Erickson C, Driscoll MS. Melanoma epidemic: Facts and controversies. Clin.Dermatol. 2010;28:281-6.
64. Stolz W, Braun-Falco O, Bilen N *et al. Color Atlas of Dermoscopy*, 2nd edn. Berlin: Blackwell Wissenschafts, 2002.
65. Argenziano G, Fabbrocini G, Carli P, de G, V, Sammarco E, Delfino M. Epiluminescence microscopy for the diagnosis of doubtful melanocytic skin lesions. Comparison of the ABCD rule of dermatoscopy and a new 7-point checklist based on pattern analysis. Arch.Dermatol. 1998;134:1563-70.
66. Menzies SW, Ingvar C, Crotty KA, McCarthy WH. Frequency and morphologic characteristics of invasive melanomas lacking specific surface microscopic features. Arch.Dermatol. 1996;132:1178-82.
67. Pehamberger H, Steiner A, Wolff K. In vivo epiluminescence microscopy of pigmented skin lesions. I. Pattern analysis of pigmented skin lesions. J.Am.Acad.Dermatol. 1987;17:571-83.
68. Stolz W, Riemann A, Cognetta A, Pillet L, Abmayr W, Hölzel D et al. ABCD rule of dermatoscopy: a new practical method for early recognition of malignant melanoma. Eur.J.Dermatol. 1994;4:521-7.
69. Vestergaard ME, Macaskill P, Holt PE, Menzies SW. Dermoscopy compared with naked eye examination for the diagnosis of primary melanoma: a meta-analysis of studies performed in a clinical setting. Br.J.Dermatol. 2008.
70. Carli P, de G, V, Chiarugi A, Nardini P, Weinstock MA, Crocetti E et al. Addition of dermoscopy to conventional naked-eye examination in melanoma screening: a randomized study. J.Am.Acad.Dermatol. 2004;50:683-9.



2

Clinical and histologic characteristics of malignant melanoma in families with a germline mutation in CDKN2A

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Abstract

Background:
About 10% of cutaneous malignant melanomas (CMM) occur in individuals with a family history of melanoma. In 20% to 40% of melanoma families germline mutations in CDKN2A are detected. Knowledge of the clinicohistologic characteristics of melanomas and patients from these families is important for optimization of management strategies, and may shed more light on the complex interplay of genetic and environmental factors in the pathogenesis of melanoma.

Objective:
We sought to investigate the clinical and histologic characteristics of CMM in CDKN2A-mutated families.

Methods:
Clinical and histologic characteristics of 182 patients with 429 CMM from families with a founder mutation in CDKN2A (p16-Leiden mutation) were compared with 7512 patients with 7842 CMM from a population-based cancer registry.

Results:
Patients with p16-Leiden had their first melanoma 15.3 years younger than control patients. The 5-year cumulative incidence of second primary CMM was 23.4% for patients with p16-Leiden compared with 2.3% for control patients. The risk of a second melanoma was twice as high for patients with p16-Leiden who had their first melanoma before age 40 years, compared with older patients with p16-Leiden. Unlike control patients, there was no body site concordance of the first and second melanoma in patients with p16-Leiden and multiple primary melanomas. Patients with p16-Leiden had significantly more superficial spreading, and less nodular and lentiginous melanomas.

Limitations:
Ascertainment of patients with p16-Leiden was family based. The study was performed in families with a founder mutation, the p16-Leiden mutation.

Conclusion:
Our findings are consistent with a pathogenic pathway of melanoma development from nevi, starting early and ongoing throughout life, and not related to chronic sun exposure.

Introduction

Between 6% and 14% of cases of primary cutaneous malignant melanoma (CMM)) have been reported to occur in individuals with a family history of melanoma.[1] In these families two major melanoma susceptibility genes have been identified so far. The oncogene CDK4 (MIM# 123829) has been found in a few melanoma families (estimated 2%).[2] Germline mutations in CDKN2A (MIM# 600160) are far more prevalent and are found in approximately 20% to 40% of melanoma families.[3] CDKN2A encodes two distinct proteins: p16INK4 and p14ARF, which both function as tumor suppressors. The penetrance of CDKN2A mutations for melanoma has been estimated to be 0.67 by the age of 80 years.[4] In The Netherlands the p16-Leiden mutation (c.225-243del19) is the most prevalent CDKN2A germline mutation.[5]

Several studies have reported that patients with melanoma and a CDKN2A mutation have an earlier age of onset and have an increased risk of multiple primary melanomas (MPM).[6-9] The purpose of this study was to further substantiate and expand the knowledge of the clinical and histologic characteristics of patients with melanoma from CDKN2A-mutated families. Knowledge of these features may not only provide useful information for clinicians, but can also shed more light on the complex interplay of genetic and environmental factors in the pathogenesis of melanoma.

We investigated the clinical and histologic characteristics of malignant melanoma in families with the p16-Leiden mutation, by comparing 182 patients with melanoma from p16-Leiden families who had 429 melanomas with patients and melanomas from a population-based cancer registry.

Methods

P16-Leiden cases were collected from proven p16-Leiden-mutated families that were registered at The Netherlands Foundation for the Detection of Hereditary Tumors (NFDHT). The organization and methods of the NFDHT have been published elsewhere.[10,11] In brief, physicians from all parts of The Netherlands refer families suspected for familial melanoma to the registry. Genealogic studies are performed and all reported malignancies are verified by medical records. If familial melanoma is confirmed clinically, the registry monitors the continuity of the surveillance program for relatives and collects follow-up data on the results of surveillance and pathologic examination. In 2007 the NFDHT database contained 51 p16-Leiden families with 194 patients with confirmed primary CMM since 1970. The majority of patients (n = 144, 74%) had been treated for at least one of their melanomas at the Department of Dermatology at the Leiden University Medical

Center, which is a tertiary referral center for familial melanoma. Most data on patient mutation status for the study were collected at this department as described elsewhere.[12] In addition we used results from clinical genetic testing, which the NFDHT receives if patients consent.

Twelve melanoma patients that had tested negative for the p16-Leiden mutation in their family were excluded from the study. The remaining 182 patients consisted of 127 proven mutation carriers and 55 untested patients (from proven p16-Leiden families). As 8.6% (12/139) of gene-tested patients tested negative for the p16-Leiden mutation, we expected about 5 phenocopies among the 55 untested patients. We found this acceptable and decided to include the untested patients as p16-Leiden patients in the study. For each patient, follow-up data were collected on subsequent melanomas and life status.

Control patients were obtained from the Leiden Cancer Registry (LCR). The LCR is a population-based registry of all newly diagnosed malignancies, which covers the western part of The Netherlands with a population of approximately 1.7 million inhabitants. The registry has (near) complete coverage since 1989. For new patients in the registry, malignancies diagnosed before the start of the registry are also recorded. All patients with histologically confirmed primary cutaneous melanoma up to September 2007 were selected from the cancer registry. The patients known to be members of p16-Leiden families were excluded from the LCR database. Survival data of the control patients were obtained from the Central Bureau for Genealogy and from the municipal registries, which keep records of all deceased persons in The Netherlands. Survival data were completed until January 2006. The LCR contained a total of 7512 eligible patients. For control patients no data on CDKN2A mutation status were available. For all included patients, data were gathered concerning date of birth and gender. For all melanomas, data were collected concerning date of diagnosis, histologic type, and body site.

Statistical analyses

Body site was subdivided into head and neck, trunk, upper extremities, lower extremities, and not recorded. Histologic type was categorized as superficial spreading melanoma (SSM) or melanoma in situ (Mis), nodular melanoma (NM), lentigo maligna (LM) melanoma (LMM) or LM, and acral lentiginous melanoma (ALM) or ALM in situ. Other histologic types and melanomas that were not otherwise specified were excluded from all analyses that included the variable histologic type. In some analyses a dichotomous covariate was included to distinguish invasive from in situ melanomas. Age and year of diagnosis were analyzed as continuous variables. In some analyses age was divided in two categories. To distinguish the two study populations we used the term "p16-Leiden status", coded 1 for patients with p16-Leiden, and 0 for control patients. This term refers to being a member of a p16-Leiden-mutated family, rather than personal mutation status (see above).

Differences in gender distribution and age at diagnosis were calculated with the Pearson x^2 and Student t test, respectively. The cumulative incidence of second primary melanomas was calculated using a competing risk analysis[13] accounting for death as competing risk. Survival times were calculated from the date of first melanoma to the date of second melanoma, death, or last follow-up. For control patients, January 2006 was considered as the end of follow-up as data on life status were only available until this date. Melanomas diagnosed after January 2006 were therefore excluded from the competing risk analyses.

A Cox proportional hazard model was used to compare the hazard of developing a second primary melanoma in the two study populations, and to study risk factors for developing a second melanoma in the two populations separately. Because the risk of developing a second melanoma was age dependent in both populations, separate hazard ratios (HRs) were calculated for age below and above 40 years. Equality of HRs across age groups was tested using Cox regression with p16-Leiden status, age group, and their interaction. To identify risk factors for a second primary melanoma, the following covariates were tested in univariate analyses: age of diagnosis (<40 vs >40 years), age of diagnosis (<60 vs >60 years), gender, year of diagnosis, tumor type (SSM/Mis, NM, LMM/LM, ALM/ALM in situ), tumor localization (lower extremities, head/neck, trunk, upper extremities), and invasiveness of the first melanoma (in situ vs invasive melanoma). Covariates with a P value less than .10 in the univariate analyses were included in the multivariate analyses.

Body site concordance of the first and second primary melanoma was calculated with Cohen k statistics. To compare concordance in patients with p16-Leiden and control patients an analysis of variance was performed.
For comparison of tumor characteristics of p16-Leiden cases and control patients, a multivariate logistic regression analysis was performed with robust SEs to correct for within-patient correlations in relatives with MPM. Comparison of the age at diagnosis according to body site pattern in the two study populations was done by means of a 3-way analysis of variance.

All analyses were performed with software (SPSS 14.0, SPSS Inc, Chicago, IL, and R 2.5.1, R Development Core Team, Vienna, Austria). Statistical significance was determined at a = .05, and all tests for statistical significance were two-sided.

Results

Patient characteristics

In total 182 patients with p16-Leiden melanoma and 429 CMM, and 7512 control patients with melanoma and 7842 CMM were included in the analyses. Patient characteristics are presented in Table 1. The gender distribution was similar in both groups. Age at diagnosis

Table 1 Patient and tumor characteristics

	p16-Leiden melanoma patients (n = 182)		Control melanoma patients (n = 7512)		p-value
p16-Leiden mutation status					
Positive	127	(69.8%)	N/A		
Not tested	55	(30.2%)	N/A		
Gender					
Male	80	(44.0%)	2977	(39.6%)	0.24
Female	102	(56.0%)	4535	(60.4%)	
Age					
Mean (SD), Male	40.5 yrs	(13.0)	55.3 yrs	(16.7)	< 0.001
Mean (SD), Female	37.9 yrs	(13.5)	53.7 yrs	(18.0)	< 0.001
Mean (SD), All	39.0 yrs	(13.3)	54.3 yrs	(17.5)	< 0.001
Total No. of melanomas	429		7842		
No. of melanomas / patient					
1	108	(59.3%)	7239	(96.4%)	< 0.001
2	30	(16.5%)	236	(3.1 %)	
3-5	27	(14.8%)	34	(0.5%)	
> 5	17	(9.3%)	3	(0%)	
Cum. Incidence of second melanoma					
1-Year incidence	8.5%		1.0%		
2-Year incidence	12.2%		1.5%		
5-Year incidence	23.4%		2.3%		
10-Year incidence	34.8%		3.1%		
20-year incidence	41.4%		4.5%		
Tumor type					< 0.001
SSM/Mis	338	(88.9%)	4205	(66.2%)	
NM	29	(7.6%)	1129	(17.8%)	
LMM/LM	8	(2.1%)	973	(15.3%)	
ALM/ALMis	5	(1.3%)	47	(0.7%)	
Other	1		275		
NOS	48		1213		
Location					< 0.001
Male					
Head & Neck	27	(14.4%)	720	(23.4%)	
Trunk	102	(54.3%)	1396	(45.4%)	
Upper extremities	26	(13.8%)	486	(15.8%)	
Lower extremities	33	(17.6%)	473	(15.4%)	
Unknown	1		38		

Table 1 Continued

	p16-Leiden melanoma patients (n = 182)		Control melanoma patients (n = 7512)		p-value
Female					
Head & Neck	26	(10.9%)	828	(17.8%)	
Trunk	70	(29.4%)	1186	(25.4%)	
Upper extremities	45	(18.9%)	963	(20.7%)	
Lower extremities	97	(40.8%)	1684	(36.1%)	
Unknown	2		68		
Invasiveness					
In situ	94	(22.0%)	1641	(20.9%)	0.61
Invasive	334	(78.0%)	6201	(79.1%)	
Unknown	1				

N/A, not applicable; SD, standard deviation; MPM-patients, multiple primary melanoma patient; Cum. incidence, cumulative incidence; SSM/Mis, Superficial Spreading Melanoma or Melanoma in situ; NM, Nodular Melanoma; LMM/LM, Lentigo Maligna Melanoma or Lentigo Maligna; ALM/ALMis, Acrolentiginous Melanoma or Acrolentiginous Melanoma in situ; NOS, not otherwise specified

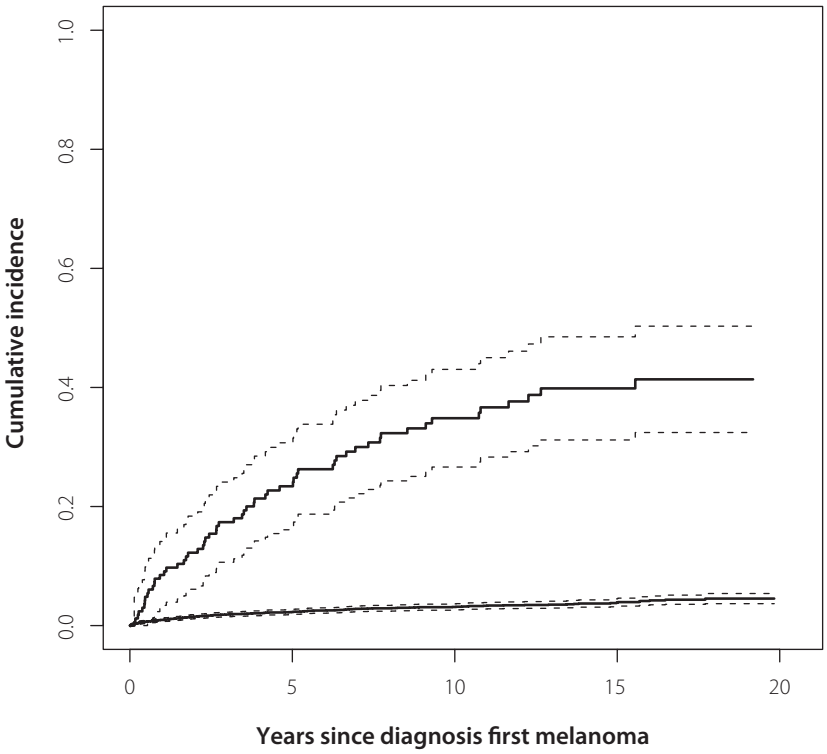
(of the first melanoma) was significantly younger in patients with p16-Leiden (mean: 39.0 years) compared with the control patients (mean: 54.3 years). The mean age difference was 15.3 years (95% confidence interval [CI] 13.3-17.3). In both populations women had their first melanoma at a slightly younger age than men, but the difference was only statistically significant in the control population (control: P <.001; p16-Leiden: P = .189).

Multiple primary melanomas

Patients with p16-Leiden were followed up for a median of 10.4 years (range 0-35.7), and control patients for 5.5 years (range 0-36.0). MPM developed in 40.7% of the patients with p16-Leiden and 3.6% of the control patients. Patients with p16-Leiden and MPM had more primary CMM per patient than control patients with MPM (median: 3.0 [range 2-19] vs 2.0 [range 2-10]).

The estimated 5-year cumulative incidence of a second primary tumor was 23.4% for patients with p16-Leiden and 2.3% for control patients (Table I). Patients with p16-Leiden were at a considerably higher risk of a second CMM than control patients (Fig 1). The HR was dependent on the age of diagnosis of the first melanoma, with a HR of 15.8 (95% CI 11.0-22.7, P < .001) for age younger than 40 years, and a HR of 7.5 (95% CI 4.5-12.7, P < .001) for age older than 40 years. These HRs were significantly different (P value for p16-Leiden status by age group interaction = .016).

Figure 1 Cumulative incidences of second melanomas with 95% confidence intervals in the p16-Leiden (upper line) and control (lower line) population



Risk factors for multiple melanomas

For patients with p16-Leiden, diagnosis of a melanoma before age 40 years was a significant risk factor for the development of a second CMM (HR 1.9, 95% CI 1.2-3.2, P = .011). For control patients localization of the first melanoma in the head and neck region was a statistically significant risk factor for a second CMM (HR 1.61, 95% CI 1.04-2.50, P = .032, "lower extremities" as reference category).

Body site concordance

The overall concordance of body site of the first and second melanoma was 36.1% for patients with p16-Leiden MPM (κ statistics 0.08, SE = 0.071, P = .250) and 52.5% for control patients with MPM (k statistics 0.36, SE = 0.041, P < .001). The difference in body site

concordance between the two populations was statistically significant (P < .001). In the control population concordance was dominated by melanomas in the head and neck region (73.7%) (Table 2).

Table 2 Concordance of body site for first and second primary cutaneous melanoma in patients with multiple melanoma

First primary ↓	Second primary									
	p16-Leiden MPM-patients n (%)					Control MPM-patients n (%)				
	H&N	T	UE	LE	Total	H&N	T	UE	LE	Total
H&N	1 (14%)	5 (71%)	1 (14%)	0 (0%)	7	56 (74%)	6 (8%)	6 (8%)	8 (11%)	76
T	5 (17%)	16 (55%)	5 (17%)	3 (10%)	29	7 (9%)	36 (47%)	19 (25%)	14 (18%)	76
UE	1 (11%)	3 (33%)	1 (11%)	4 (44%)	9	7 (15%)	17 (37%)	15 (33%)	7 (15%)	46
LE	3 (11%)	10 (37%)	6 (22%)	8 (30%)	27	9 (14%)	15 (23%)	10 (15%)	31 (48%)	65
Total	10 (14%)	34 (47%)	13 (18%)	15 (21%)	72	79 (30%)	74 (28%)	50 (19%)	60 (23%)	263

MPM, Multiple Primary Melanomas; H&N, Head and neck; T, Trunk; UE, Upper Extremities; LE, Lower Extremities

Tumor body site and histologic type

A multivariate analysis was performed to investigate differences between p16-Leiden cases and controls concerning tumor type and tumor localization, while adjusting for age of diagnosis, gender, incidence year, and invasiveness. A statistically significant difference in tumor type distribution was found. Patients with p16-Leiden had a smaller proportion of NM (odds ratio 0.38) and LMM/LM (odds ratio 0.20) than control patients (Table 3). There was no difference in tumor localization between patients with p16-Leiden and control patients.

Age at diagnosis related to body site

Patients with p16-Leiden developed melanomas on the head and neck and lower extremities at a younger age than melanomas on the trunk and upper extremities (Table 4). In the control patients head and neck tumors were diagnosed at an older age than tumors on all other body sites. The difference between age at diagnosis by body site pattern of patients with p16-Leiden and control patients was statistically significant (P < .001).

Table 3 Multivariate analysis for characteristics associated with p16-Leiden families

Covariate	OR (95% CI)	p-value
Age	0.96 (0.95 – 0.97)	< 0.001
Gender		
Male	1	
Female	0.70 (0.43 – 1.13)	0.14
Incidence year	0. 91 (0.89 – 0.94)	< 0.001
Invasiveness		
In situ	1	
Invasive	0.74 (0.54 – 1.01)	0.067
Tumor type		
SSM/Mis	1	
NM	0.38 0.25 – 0.57)	< 0.001
LMM/LM	0.20 (0.10 – 0.40)	< 0.001
ALM/ALMis	0.96 (0.38 – 2.44)	0.93
Localization		
Lower extremities	1	
Head/neck	1.35 (0.84 – 2.16)	0.22
Trunk	1.07 (0.79 – 1.43)	0.68
Upper extremities	1.07 (0.77 – 1.49)	0.67

OR, Odds ratio; 95% CI, 95% confidence interval; SSM/Mis, Superficial Spreading Melanoma or Melanoma in situ; NM, Nodular Melanoma; LMM/LM, Lentigo Maligna Melanoma or Lentigo Maligna; ALM/ALMis, Acrolentiginous Melanoma or Acrolentiginous Melanoma in situ

Control patients are the reference population.

Table 4 mean age at diagnosis for different tumor localizations in p16-Leiden and control patients

	p16-Leiden patients		**Control patients**	
Localization	Age* (SD)	n	Age* (SD)	n
Trunk	41.5 yrs (11.8 yrs)	72	50.0 yrs (15.5 yrs)	2495
Head/neck	36.1 yrs (14.6 yrs)	24	65.4 yrs (16.8 yrs)	1447
Upper extremities	41.6 yrs (13.9 yrs)	27	55.0 yrs (17.0 yrs)	1379
Lower extremities	36.3 yrs (13.7 yrs)	57	51.4 yrs (17.4 yrs)	2086
Unspecified	25.3 yrs (14.5 yrs)	2	52.4 yrs (16.6 yrs)	105
Total	39.0 yrs (13.3 yrs)	182	54.3 yrs (17.5 yrs)	7512

*For multiple melanoma patients the age at diagnosis of the first melanoma was taken. SD, standard deviation

Because patients with p16-Leiden had considerably less LMM/LM, which are known to be diagnosed at an older age and frequently occur in the head and neck region, we performed a subanalysis in which we excluded all LMM/LM. The difference in age by body site patterns remained highly statistically significant in this analysis ($P < .001$).

Discussion

We compared the clinical and histologic characteristics of 182 patients with 429 CMM from families with a germline mutation in CDKN2A, with a large control population from the Leiden population-based cancer registry (7512 patients with 7842 CMM). Patients with p16-Leiden melanoma had a younger age of onset and a highly increased risk of MPM, which was highest for patients who had their first melanoma before the age of 40 years. In contrast to control patients with MPM no body site concordance was found for first and second melanomas in patients with p16-Leiden MPM. Patients with p16-Leiden had a higher proportion of SSM/Mis and less NM and LMM/LM. Furthermore, a different age (at diagnosis) by body site pattern was found in the two populations.

The patients with p16-Leiden had a considerably younger age of onset of 39 years. Other studies reported a comparable age at diagnosis for CDKN2A mutation carriers, ranging from 36.3 to 43.3 years.[7-9]

With regard to the occurrence of MPM in patients with p16-Leiden our findings are in accordance with earlier reports, and contain some new observations. Like previous studies on CDKN2A-mutated families, we found a very high proportion of patients with MPM (40.7%) in the p16-Leiden population. It was strikingly higher than the 18.6%[8] and 25.6%[6] of patients with MPM reported in two other studies. The results are difficult to compare, however, because the duration of follow-up was not recorded in these studies. We estimated the 5-year cumulative incidence of second melanomas to be 23.4% in the p16-Leiden population, and 2.3% in the control population, which was similar to the 1.5% to 3.4% reported in other population-based studies.[14-16] In a clinic-based study, Ferrone et al[17] found a 5-year risk of 11.4%. This relatively high risk may have been because of the fact that their study was performed in a tertiary cancer center. Diagnosis of the first melanoma at a young age (<40 years) was associated with an almost doubled risk of MPM in the p16-Leiden population.

Unlike patients with p16-Leiden MPM, there was a statistically significant association between the body site of the first and second melanoma in control patients with MPM. In the control population overall body site concordance was 53%, which was similar to the 48% to 55% reported in earlier studies in comparable populations.[17-19] Like Giles et al,[18] we found concordance to be highest for tumors located in the head and neck area. The

absence of body site concordance in patients with p16-Leiden melanoma, in addition to the highly increased risk of MPM, underlines the importance of frequent and lifelong total-body skin examinations for these patients.

Patients with p16-Leiden had proportionally less NM and LMM/LM, but more SSM/Mis than control patients. There are earlier reports of a decreased proportion of NM in familial melanoma.[20] LMM/LM are diagnosed in extensively sun-damaged skin, usually in elderly people. Because patients with p16-Leiden had a much younger age of onset, less LMM/LM were expected in this patient population. But even though we adjusted for age, the difference remained strongly significant. An increased proportion of superficial spreading type melanomas in familial melanoma has been reported before.[21] It has been suggested that this is because a relatively large proportion of melanomas in patients with familial melanoma arises from (dysplastic) nevi as familial melanoma is associated with increased numbers of (dysplastic) nevi. Melanomas that are associated with nevi are usually of the superficial spreading type.[22] In a recent study Nagore et al[23] observed no acral melanomas among 41 familial melanoma cases. In our study the proportion of ALM/ALM in situ melanomas was similar in patients with p16-Leiden and control patients. The absence reported by Nagore et al[23] may be a result of small sample size in their study.

In accordance with previous studies,[24,25] we found that in the control population, head and neck melanomas were diagnosed at a considerably older age than melanomas on other body sites. Interestingly in the p16-Leiden population head and neck tumors were diagnosed at a younger age than tumors located on the trunk and upper extremities, even after exclusion of LMM/LM from the analysis.

Our findings in the p16-Leiden population are in accordance with the inherited increased susceptibility to melanoma that CDKN2A germline mutations are associated with, including the: (1) young age of onset; (2) high risk of MPM; and (3) absence of body site concordance of the first and second melanoma in patients with MPM. Besides this, our results also suggest that differences exist between the pathogen„esis of melanomas in patients with p16-Leiden and control patients. Several studies have brought forward that melanoma is a heterogeneous
disease.[24,26-28] Whiteman et al[27,28] proposed two distinct etiologic pathways, one associated with increased numbers of nevi, intermittent sun exposure, younger age at diagnosis, and location on the trunk (nevus pathway). The second pathway is associated with chronic sun exposure, fewer nevi, older age at diagnosis, and location in the head and neck region (ultraviolet B pathway). Our results in the control population are in accordance with this theory, as both pathways can be distinguished in this population. First of all, age at diagnosis was considerably higher for tumors in the head and neck region compared with tumors on the trunk, which is in support of the theory of a different origin for these two

body sites. Secondly, we found a statistically significant body site concordance in patients with MPM, with a high concordance for head and neck tumors. This clustering suggests a localized increased melanoma risk, which is in agreement with the divergent pathway theory. In the p16-Leiden population, the high proportion of superficial spreading type melanomas and lack of lentiginous melanomas suggest that melanomas in these patients develop predominantly through the nevus pathway. The young age at diagnosis of melanomas located in the head and neck region is difficult to interpret, however. Relatively high ultraviolet exposure at this body site might play a roll (synergy of the nevus and ultraviolet B pathways).

The ascertainment of patients with p16-Leiden was family based, which is a possible source of bias. Familial melanoma with a young age of onset or MPM are more likely to be identified and genetically tested. The differences between the p16-Leiden population and control population would probably have been smaller if patients with p16-Leiden had been ascertained from a patient population unselected for family history. Such a design has serious drawbacks, however, given the low prevalence of CDKN2A mutations in patients with general melanoma (0.2%-2.0%).[29,30] Moreover, we consider our results to be representative for patients from proven p16-Leiden families, as they present in clinic in daily practice.

In summary, we have verified and substantiated several characteristics of patients with familial melanoma and a CDKN2A mutation in a large case-control study and we have reported a number of new findings. Our findings are in concordance with the so-called divergent pathways hypotheses: familial melanomas tend to follow the nevus pathway. More studies are necessary to determine whether our results apply to founder populations with other CDKN2A mutations as well.

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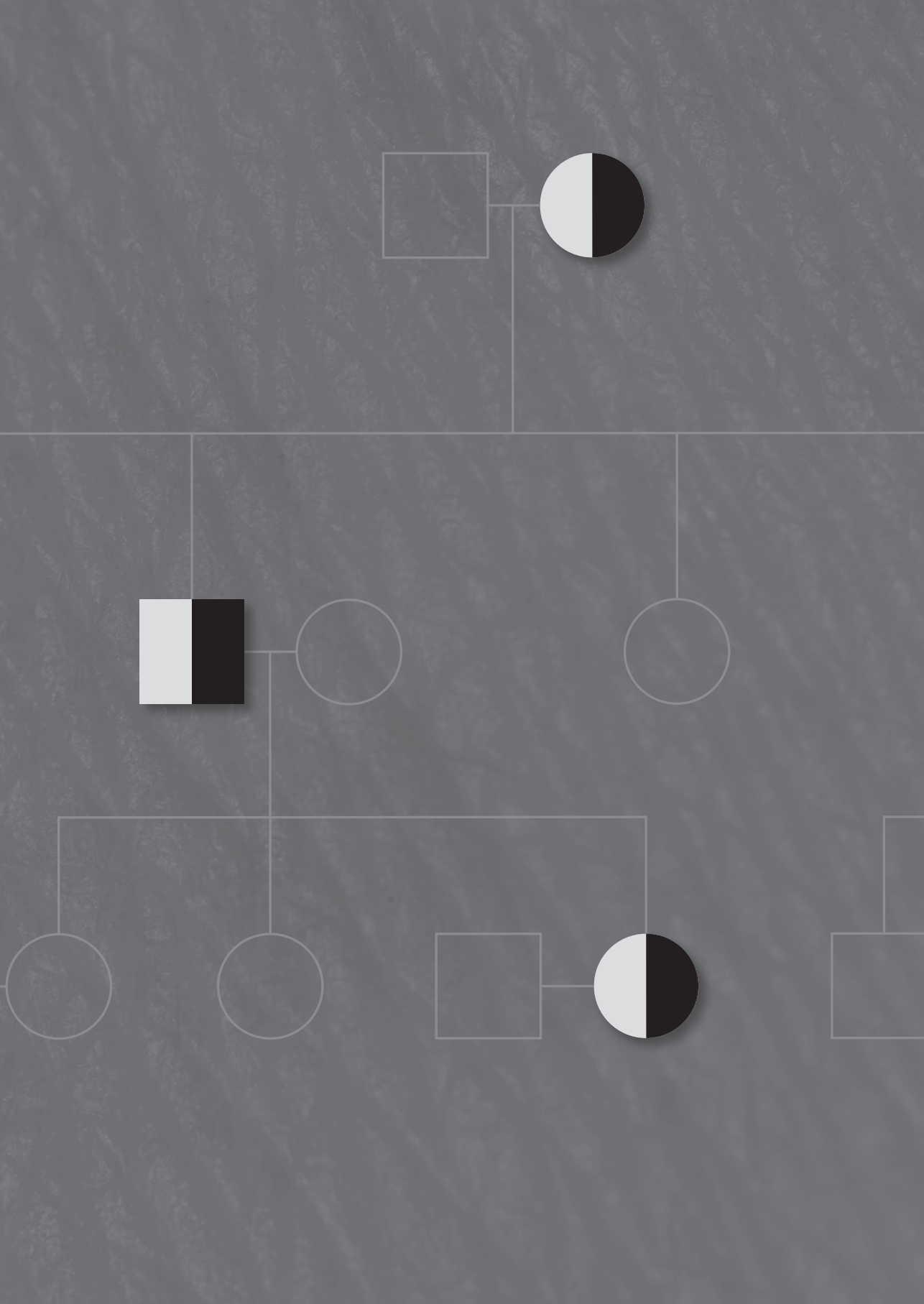
References

1. Ang CG, Kelly JW, Fritschi L, Dowling JP. Characteristics of familial and non-familial melanoma in Australia. Melanoma Res 1998;8:459-64.
2. Goldstein AM, Chan M, Harland M, Gillanders EM, Hayward NK, Avril MF, et al. High-risk melanoma susceptibility genes and pancreatic cancer, neural system tumors, and uveal melanoma across GenoMEL. Cancer Res 2006;66:9818-28.
3. Kefford RF, Newton Bishop JA, Bergman W, Tucker MA. Counseling and DNA testing for individuals perceived to be genetically predisposed to melanoma: a consensus statement of the melanoma genetics consortium. J Clin Oncol 1999;17:3245-51.
4. Bishop DT, Demenais F, Goldstein AM, Bergman W, Bishop JN, Bressac-de Paillerets B, et al. Geographical variation in the penetrance of CDKN2A mutations for melanoma. J Natl Cancer Inst 2002;94:894-903.
5. Gruis NA, van der Velden PA, Sandkuijl LA, Prins DE, Weaver-Feldhaus J, Kamb A, et al. Homozygotes for CDKN2 (p16) germline mutation in Dutch familial melanoma kindreds. Nat Genet 1995;10:351-3.
6. Borg A, Sandberg T, Nilsson K, Johannsson O, Klinker M, Masback A, et al. High frequency of multiple melanomas and breast and pancreas carcinomas in CDKN2A mutation-positive melanoma families. J Natl Cancer Inst 2000;92:1260-6.
7. Goldstein AM, Struewing JP, Chidambaram A, Fraser MC, Tucker MA. Genotype-phenotype relationships in US melanoma-prone families with CDKN2A and CDK4 mutations. J Natl Cancer Inst 2000;92:1006-10.
8. Mantelli M, Barile M, Ciotti P, Ghiorzo P, Lantieri F, Pastorino L, et al. High prevalence of the G101W germline mutation in the CDKN2A (P16(ink4a)) gene in 62 Italian malignant melanoma families. Am J Med Genet 2002;107:214-21.
9. Masback A, Olsson H, Westerdahl J, Sandberg T, Borg A, Jonsson N, et al. Clinical and histopathological features of malignant melanoma in germline CDKN2A mutation families. Melanoma Res 2002;12:549-57.
10. Vasen HF, Bergman W, van Haeringen A, Scheffer E, van Slooten EA. The familial dysplastic nevus syndrome: natural history and the impact of screening on prognosis; a study of nine families in The Netherlands. Eur J Cancer Clin Oncol 1989; 25:337-41.
11. Vasen HF, Gruis NA, Frants RR, Der Velden PA, Hille ET, Bergman W. Risk of developing pancreatic cancer in families with familial atypical multiple mole melanoma associated with a specific 19 deletion of p16 (p16-Leiden). Int J Cancer 2000; 87:809-11.
12. de Snoo FA, Bishop DT, Bergman W, van Leeuwen I, van der Drift C, van Nieuwpoort FA, et al. Increased risk of cancer other than melanoma in CDKN2A founder mutation (p16-Leiden) positive melanoma families. Clin Cancer Res 2008;14:7151-7.
13. Putter H, Fiocco M, Geskus RB. Tutorial in biostatistics: competing risks and multi-state models. Stat Med 2007;26: 2389-430.
14. Goggins WB, Tsao H. A population-based analysis of risk factors for a second primary cutaneous melanoma among melanoma survivors. Cancer 2003;97:639-43.
15. Kang S, Barnhill RL, Mihm MC Jr, Sober AJ. Multiple primary cutaneous melanomas. Cancer 1992;70:1911-6.
16. Slingluff CL Jr, Vollmer RT, Seigler HF. Multiple primary melanoma: incidence and risk factors in 283 patients. Surgery 1993;113:330-9.
17. Ferrone CR, Ben Porat L, Panageas KS, Berwick M, Halpern AC, Patel A, et al. Clinicopathological features of and risk factors for multiple primary melanomas. JAMA 2005;294:1647-54.
18. Giles G, Staples M, McCredie M, Coates M. Multiple primary melanomas: an analysis of cancer registry data from Victoria and New South Wales. Melanoma Res 1995;5:433-8.
19. Savoia P, Quaglino P, Verrone A, Bernengo MG. Multiple primary melanomas: analysis of 49 cases. Melanoma Res 1998; 8:361-6.
20. Greene MH, Clark WH Jr, Tucker MA, Kraemer KH, Elder DE, Fraser MC. High risk of malignant melanoma in melanoma-prone families with dysplastic nevi. Ann Intern Med 1985;102: 458-65.
21. Kopf AW, Hellman LJ, Rogers GS, Gross DF, Rigel DS, Friedman RJ, et al. Familial malignant melanoma. JAMA 1986;256:1915-9.
22. Bevona C, Goggins W, Quinn T, Fullerton J, Tsao H. Cutaneous melanomas associated with nevi. Arch Dermatol 2003;139: 1620-4.

23. Nagore E, Botella-Estrada R, Garcia-Casado Z, Requena C, Serra-Guillen C, Llombart B, et al. Comparison between familial and sporadic cutaneous melanoma in Valencia, Spain. J Eur Acad Dermatol Venereol 2008;22:931-6.
24. Lachiewicz AM, Berwick M, Wiggins CL, Thomas NE. Epidemiologic support for melanoma heterogeneity using the surveillance, epidemiology, and end results program. J Invest Dermatol 2008;128:1340-2.
25. Hoersch B, Leiter U, Garbe C. Is head and neck melanoma a distinct entity? A clinical registry-based comparative study in 5702 patients with melanoma. Br J Dermatol 2006;155: 771-7.
26. Curtin JA, Fridlyand J, Kageshita T, Patel HN, Busam KJ, Kutzner H, et al. Distinct sets of genetic alterations in melanoma. N Engl J Med 2005;353:2135-47.
27. Whiteman DC, Watt P, Purdie DM, Hughes MC, Hayward NK, Green AC. Melanocytic nevi, solar keratoses, and divergent pathways to cutaneous melanoma. J Natl Cancer Inst 2003;95: 806-12.
28. Whiteman DC, Parsons PG, Green AC. p53 Expression and risk factors for cutaneous melanoma: a case-control study. Int J Cancer 1998;77:843-8.
29. Aitken J, Welch J, Duffy D, Milligan A, Green A, Martin N, et al. CDKN2A variants in a population-based sample of Queensland families with melanoma. J Natl Cancer Inst 1999;91:446-52.
30. Begg CB, Orlow I, Hummer AJ, Armstrong BK, Kricker A, Marrett LD, et al. Lifetime risk of melanoma in CDKN2A mutation carriers in a population-based sample. J Natl Cancer Inst 2005;97:1507-15.



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Effectiveness and causes for failure of surveillance of CDKN2A-mutated melanoma families

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Abstract

Background:
For more than 25 years families with an increased susceptibility to melanoma have been under surveillance at our institution.

Objective:
We sought to investigate the effectiveness of surveillance for CDKN2A-mutated families and causes for failure of the program in patients with more advanced tumors.

Methods:
In a retrospective case-control study, Breslow thickness of melanomas diagnosed in relatives enrolled in the surveillance program were compared with melanomas of unscreened index patients. We investigated the influence of mode of detection and length of surveillance interval on outcome.

Results:
Surveillance melanomas (n = 226, median thickness: 0.50 mm) had a significantly lower Breslow thickness (multiplication factor: 0.61 [95% confidence interval 0.47-0.80], P \ .001) than index melanomas (n = 40, median thickness: 0.98 mm). Index melanomas were more likely diagnosed with a Breslow thickness greater than 1.0 mm (odds ratio: 3.1 [95% confidence interval 1.2-8.1], P = .022). In all, 53% of surveillance melanomas were diagnosed during regular screens, 7% during patients' first screen, 20% between regular screens, and 20% in patients who were noncompliant with the surveillance schedule. The majority of surveillance melanomas (58%) were detected within 6 months after the last screen. There was no correlation between tumor thickness and the length of the screening interval for tumors diagnosed within 24 months since the last screen.

Limitations:
The study is retrospective.

Conclusions:
Surveillance was associated with earlier detection of melanomas. Noncompliance was an important cause for failing surveillance. Shortening surveillance intervals may advance detection of tumors, but may paradoxically have little impact on prognosis.

Introduction

About 10% of primary cutaneous malignant melanomas have been reported to occur in families.[1] In 1994, germline mutations in the CDKN2A gene (MIM# 600160) were demonstrated in kindreds with hereditary melanoma.[2,3] CDKN2A encodes two distinct proteins: p16INK4 and p14ARF, both of which function as tumor suppressors. CDKN2A is the most prevalent high-penetrance melanoma susceptibility gene, mutations being detected in the germline in 20% to 40% of melanoma families.[4-6] Mutations in CDKN2A have an estimated penetrance of 67% by the age of 80 years.[4]

In 1981 the familial melanoma study group of the Leiden University Medical Center (LUMC) initiated a surveillance program for familial melanoma kindreds. Many of the first families that were screened at the LUMC were later shown to have a founder mutation in CDKN2A, consisting of a 19 base pair deletion in exon 2 (the p16-Leiden mutation).[7]

In 1989 we evaluated the surveillance program for these families, and reported that screen-detected melanomas (n = 31) had more favorable prognostic characteristics than those detected before the start of the surveillance program (n = 19).[8] We have noticed, however, that in spite of the surveillance program, some melanomas are detected relatively late. Possible explanations include: noncompliance with follow-up instructions; intervals between screens being too long to warrant early detection in all instances, because some melanomas grow rapidly[9]; failure to recognize melanomas because of an atypical clinical presentation[10]; or inadequate screening.

In the current study we compared the Breslow thickness of 226 melanomas of patients from p16-Leiden mutation positive families who were enrolled in the surveillance program with 40 melanomas of index patients from the same families, diagnosed before recognition of heredity for melanoma in these families. In addition we looked at the length of the surveillance intervals and the mode of detection of the melanomas.

Methods

The majority of families under surveillance at the LUMC were ascertained through the pigmented lesions clinic of the department of dermatology from 1980 onward. Family trees have been constructed for each kindred, initially at the clinic and later at The Netherlands Foundation for the Detection of Hereditary Tumors. Ascertainment of family data at the clinic[11] and The Netherlands Foundation for the Detection of Hereditary Tumors[8,12] has been described in detail elsewhere. Family members of clinically proven melanoma pedigrees were invited to the surveillance program, which consisted of an annual total skin examination. If a melanoma was diagnosed, surveillance was intensified

during the first 5 years after diagnosis (every 3 months during the first year, every 4 months during the second year, and every 6 months during the third to fifth year).

Before and after the identification of the p16-Leiden mutation in 1994, blood samples for research purposes have been collected from relatives who signed an informed consent form. Pedigree information was updated on a regular basis. We consider cancer data for all included families to be complete from 1970 onward. All melanomas diagnosed in family members who had been enrolled in the LUMC surveillance program were selected. These included tumors detected at the pigmented lesions clinic of the LUMC, and melanomas incidentally detected at other departments and by general practitioners. Melanomas detected before the start of the surveillance program in relatives who were under surveillance because of previous melanomas were also included. They were all termed "surveillance melanomas." Melanomas diagnosed in patients who had continued their surveillance at another institution were excluded. The first melanoma of the first two patients with melanoma from each family served as controls. They were detected before recognition of heredity for melanoma in these families, and termed "index melanomas." In total, 344 melanomas diagnosed in relatives from 37 families were eligible to the study.

For each patient, data were collected concerning date of birth and gender. For all melanomas, data on Breslow thickness, histologic type, and date of diagnosis were gathered and patient age at time of diagnosis was calculated. Screening intervals were calculated as the time between the last screen and melanoma detection. All tumors with missing data on Breslow thickness or histologic type, all in situ melanomas, and melanomas other than the superficial spreading histologic type (n = 132) were reviewed by one of us (W. J. M.). In all, 28 lesions were excluded from the study, because they were reclassified as benign (n = 22), unclassifiable (n = 5), or recurrent melanoma (n = 1). In situ melanomas and invasive melanomas with missing data on Breslow thickness that were unavailable for revision were excluded from the study.

We distinguished 4 modes of detection after enrollment in the surveillance program and surveillance melanomas were classified accordingly. Melanomas diagnosed at the first screen were termed "first-screen melanomas." If melanomas were detected at a subsequent screen, they were termed "regular-screen melanomas." Tumors that were detected between scheduled screens were termed "interval melanomas." The final category, "noncompliance melanomas," consisted of melanomas that were detected more than 2 months after the recommended screening interval. The margin of 2 months was taken because there have been waiting lists for the pigmented lesions clinic in the past (Fig 1).

Figure 1 Screening categories according to mode of ascertainment

Bold vertical lines = scheduled screening appointment; *dashed vertical line* = skipped screening appointment. *Arrows* indicate moment of diagnosis, and accompanying numbers refer to screening category: *1* = first-screen melanoma; *2* = regular-screen melanoma; *3* = interval melanoma; *4* = noncompliance melanoma.

Statistical analysis

Multivariate linear regression and binary logistic regression analyses were performed to calculate the effect of surveillance on Breslow thickness. Comparisons were made between surveillance melanomas and index melanomas, and among the 4 surveillance melanoma categories and index melanomas. In the linear regression analyses a log-transformed Breslow thickness was used. Because differences in the log-transformed variable translate to multiplication factors on the original scale, results are reported as multiplication factors on the original scale. In the logistic regression analyses Breslow thickness was analyzed as a categorical variable, coded 1 for Breslow thickness less than or equal to 1.00 mm, and 2 for greater than 1.00 mm. All analyses were adjusted for gender, age at diagnosis (in years), and year of diagnosis.

Many patients had multiple primary melanomas. We anticipated that these patients had their subsequent melanomas diagnosed at a more favorable prognostic stage than their first melanoma, not just because of surveillance, but also because of a change of the patients' and physicians' attitudes and behavior because of the previous (first) melanoma. For this reason we adjusted for melanoma rank, using a covariate coded 1 for first melanoma and 2 for all subsequent melanomas. In addition we used generalized estimating equations[13] to correct for within-patient correlations; this method uses sandwich estimators to calculate robust SEs. Correlation between the length of the screening interval and tumor thickness was calculated with linear regression analyses, with log-transformed Breslow thickness as dependent and the screening interval (in years) as covariate.

All analyses were performed with software (SPSS 14.0, SPSS Inc, Chicago, IL, and R 2.5.1, R Development Core Team, Vienna, Austria). The package geepack[14] was used for the calculation of adjusted SEs. Statistical significance was determined at a = .05, and all tests were two-sided. For analyses in which more than two groups were compared a Bonferroni correction for multiple testing was performed.

Results

In total, 266 melanomas from 114 patients were included (Table 1). These melanomas consisted of 40 index melanomas and 226 surveillance melanomas. Median Breslow thickness was 0.98 mm for index melanomas and 0.50 mm for surveillance melanomas (Table II). The mean thickness of surveillance melanomas was 0.61 times that of index melanomas (95% confidence interval [CI] 0.47-0.80, P < .001). The probability of being diagnosed with a Breslow thickness greater than 1.00 mm was significantly larger for index melanomas (odds ratio [OR] 3.1, CI 1.2-8.1, P = .022).

Table 1 Patient characteristics

	Patients (n = 114)	
Gender		
Male	50	(44%)
Female	64	(56%)
No. of melanomas / patient		
1	61	(54%)
2	22	(19%)
3-5	19	(17%)
6-10	8	(7%)
> 10	4	(4%)

Mode of detection of surveillance melanomas

Classification according to mode of detection was possible for 191 surveillance melanomas (85%) (Table 2). Tumors were classified as follows: 13 first-screen (7%), 102 regular-screen (53%), 38 interval (20%), and 38 noncompliance (20%) melanomas. Compliance was related to the number of melanomas for which patients had previously been given a diagnosis. The proportion of noncompliance melanomas was 46% among first melanomas, and 26% of first melanomas were regular-screen melanomas. For subsequent melanomas, patient compliance steadily increased (Table 2).

Screening interval

Most regular-screen (72%) and interval (68%) melanomas were diagnosed in patients who were under intensified surveillance because of a previous melanoma. The median interval between the last screen and moment of detection was 5 months for regular-screen melanomas, 3.5 months for interval melanomas, and 24 months for noncompliance

Table 2 Tumor characteristics according to screening category

	Index	Surveillance					
		First	Regular	Interval	Noncompliant	Unclassified	Total
Patients (n)	35	12	49	20	36	20	92*
Melanomas (n)	40	13	102	38	38	35	226
Age at diagnosis**							
1st melanoma (years)	42 (17 – 62)	32 (22 – 72)	34 (15 – 66)	33 (27 – 44)	39 (23 – 67)	27 (15 – 43)	36 (15 – 72)
all melanomas (years)	42 (17 – 62)	39 (22 – 72)	46 (22 – 72)	35 (25 – 76)	42 (23 – 67)	43 (15 – 64)	41 (15 – 76)
Year of diagnosis**	'84 ('68 – '00)	'85 ('81 – '04)	'97 ('73 – '06)	'98 ('72 – '06)	'98 ('84 – '06)	'92 ('77 – '06)	'97 ('72 – '06)
Screening interval (months)**	NA	NA	5.0 (1 – 14)	3.5 (0 – 11)	24 (12 – 159)	NA	NA
Diagnosis < 5 yrs after previous melanoma	NA	NA	73 (71.6%)	26 (68.4%)	4 (10.5%)	NA	118 (52.2%)
Melanoma rank							
1st melanoma	40	11 (18%)	16 (26%)	6 (10%)	28 (46%)	10	71
2nd melanoma	0	2 (6%)	19 (58%)	8 (24%)	4 (12%)	10	43
3rd melanoma	0	0	15 (63%)	6 (25%)	3 (13%)	5	29
> 3rd melanoma	0	0	52 (71%)	18 (25%)	3 (4%)	10	83
Breslow thickness							
Median	0.98 mm	0.69 mm	0.50 mm	0.49 mm	0.52 mm	0.50 mm	0.50 mm
Range	(0.30 – 12.00)	(0.30 – 1.80)	(Mis – 2.10)	(Mis – 3.90)	(Mis – 2.60)	(Mis – 3.10)	(Mis – 3.90)
Mean***	1.48 mm	0.75 mm	0.58 mm	0.76 mm	0.84 mm	0.70 mm	0.68 mm

*Numbers in row do not total 92 as some patients with multiple melanomas had melanomas belonging to different screening categories; **, Numbers in table represent median and range; NA, not applicable; Mis, melanoma in situ; ***, based on invasive melanomas only

melanomas (Table2). The Breslow thickness of surveillance melanomas was not correlated with the length of the screening interval for intervals less than 24 months (Table 3) (linear regression analysis, multiplication factor: 1.01/y, 95% CI 0.83-1.23, P = .917). If melanomas detected after an interval of more than 24 months were included in the analysis a significant correlation between screening interval and Breslow thickness was found (multiplication factor: 1.09/y, 95% CI 1.03-1.15, P = .003).

Table 3 Tumor thickness according to the time interval between the last screening and the moment of melanoma diagnosis

Time Interval	n	Median* (range)	n	Mean** (SD)
0 – 4 months	69	0.48 (Mis – 2.10)	57	0.62 (0.39)
5 – 8 months	43	0.55 (Mis – 3.90)	36	0.70 (0.64)
9 – 12 months	27	0.50 (Mis – 1.40)	23	0.53 (0.25)
13 – 18 months	11	0.50 (Mis – 1.20)	8	0.68 (0.27)
19 – 24 months	10	0.49 (Mis – 1.00)	8	0.61 (0.28)
> 24 months	17	0.48 (Mis – 2.60)	13	1.07 (0.77)
Total	177***	0.50 (Mis – 3.90)	145	0.67 (0.49)

Mis, melanoma in situ.
* Based on in situ and invasive melanomas; 95% CI, 95% confidence interval.
** Based on invasive melanomas only; SD, standard deviation.
*** One missing value for a noncompliance melanoma.

Detection of interval melanomas

Most interval melanomas (n = 21, 55%) were detected by patients themselves, with a median Breslow thickness of 0.55 mm (range: in situ-1.60 mm), after a median interval of 5 months (range: 1 - 11). Ten interval melanomas (26%) were diagnosed by physicians at an appointment for the excision of another pigmented lesion, judged to be suspicious at the last screen (median thickness: 0.40 mm [in situ - 0.90 mm], interval: 1 month [0-2]). Physicians consulted for another medical condition diagnosed 6 of the interval melanomas (16%, median thickness: 0.38 mm [in situ-3.90 mm], interval: 7.5 months [2-11]). One interval melanoma was detected in a research project (thickness: 2.00 mm, interval 3 months).

Tumor thickness according to mode of detection

The tumor thickness according to mode of detection is shown in Table 4. Regular-screen melanomas were significantly thinner than index melanomas (multiplication factor: 0.53, 95% CI 0.46-0.87, P <.001) and at borderline significance, first-screen melanomas were thinner than index melanomas (multiplication factor: 0.63, 95% CI 0.46-0.87, P = .0053, significance at a = .005, because of multiple testing) (Table 5). The probability of diagnosing

Table 4 Cumulative number and proportion of cases according to Breslow thickness

	Breslow thickness					
Category	**Mis**	**≤ 0.75mm**	**≤ 1.00mm**	**≤ 2.00mm**	**≤ 4.00mm**	**Total**
Index:	0 (0%)	13 (33%)	23 (58%)	32 (80%)	38 (95%)	40
Surveillance:						
- First screening	0 (0%)	10 (77%)	11 (85%)	13 (100%)	-	13
- Regular screening	17 (17%)	88 (86%)	96 (94%)	101 (99%)	102 (100%)	102
- Interval	8 (21%)	30 (79%)	33 (87%)	37 (97%)	38 (100%)	38
- Noncompliance	7 (18%)	24 (63%)	31 (82%)	36 (95%)	38 (100%)	38
- Not categorized	3 (9%)	29 (83%)	30 (86%)	33 (94%)	35 (100%)	35
Surveillance (all):	35 (16%)	181 (80%)	201 (89%)	220 (97%)	226 (100%)	226

Mis, melanoma in situ

a tumor with Breslow thickness greater than 1.00 mm was not significantly different between any of the screening categories and index melanomas (Table 5).

To further investigate possible differences between the different screening categories we performed a subanalysis with a cut-off point of 0.75 mm, as used in older versions of the American Joint Committee on Cancer staging system. The probability of being diagnosed with a tumor thickness greater than 0.75 mm was significantly larger for index melanomas than for regular-screen melanomas (OR 14.6, CI 4.4-48.2, $P < .001$) (Table 5), interval melanomas (OR 7.7, CI 2.0-29.3, $P = .0029$), and first-screen melanomas (OR 6.6, 95% CI 1.8-24.4, $P = .0047$). Noncompliance melanomas had a higher probability of being diagnosed with a Breslow thickness greater than 0.75 mm than regular-screen melanomas (OR 4.8, CI 1.8-13.2, $P = .0021$).

Table 5 Linear and logistic regression analyses

		Linear regression		Logistic regression					
				< 1.00 mm			< 0.75 mm		
Covariate	**MF**	**95% CI**	**p***	**OR**	**95% CI**	**p***	**OR***	**95% CI**	**p***
Screening category									
Index melanomas	1.0	-	-	1.0	-	-	1.0	-	-
First screen	0.63	(0.46 – 0.87)	0.0053	3.4	(0.7 – 16.0)	0.12	6.6	(1.8 – 24.4)	0.0047[$]
Regular screen	0.53	(0.40 – 0.70)	< 0.001[$]	5.7	(1.2 – 28.2)	0.031	14.6	(4.4 – 48.2)	< 0.001[$]
Interval	0.65	(0.44 – 0.96)	0.030	2.1	(0.4 – 11.6)	0.40	7.7	(2.0 – 29.3)	0.0028[$]
Noncompliance	0.69	(0.50 – 0.97)	0.031	2.1	(0.6 – 7.2)	0.27	3.0	(1.1 – 8.7)	0.041
Gender									
Male	1.0			1.0	-	-	1.0	-	-
Female	0.86	(0.73 – 1.01)	0.060	1.3	(0.6 – 2.8)	0.54	1.7	(1.0 – 3.1)	0.063
Age per 10 years	1.05	(1.00 – 1.11)	0.052	0.84	(0.60 – 1.18)	0.32	0.90	(0.75 - 1.10)	0.31
Date of diagnosis per 10 years	0.98	(0.88 – 1.09)	0.68	1.32	(0.79 – 2.23)	0.29	1.21	(0.85 – 1.72)	0.29
Melanoma rank									
Subsequent	1.0			1.0	-	-	1.0	-	-
First	0.93	(0.74 – 1.17)	0.56	1.7	(0.5 – 6.5)	0.43	0.8	(0.3 – 2.0)	0.56

MF, multiplication factor; OR, odds ratio; 95% CI, 95% confidence interval, *, significance level at $p < 0.005$ because of Bonferroni correction; [$], statistically significant

Discussion

We evaluated the effectiveness of our surveillance program for familial melanoma kindred with the p16-Leiden mutation in CDKN2A. The tumor thickness of 226 melanomas of relatives enrolled in the surveillance program was compared with 40 melanomas diagnosed in index patients.

Surveillance melanomas were significantly thinner than index melanomas, indicating that melanomas were detected in an earlier stage during the surveillance program. This is to some degree surprising, given the fact that only 53% of the surveillance melanomas were detected at a regular screen. Of surveillance melanomas, 7% were detected at first screens, 20% between regular screens, and 20% in patients who were not compliant with follow-up instructions at the time of diagnosis. Mode of ascertainment clearly influenced the effectiveness of surveillance. Only first-screen and regular-screen melanomas had a significantly lower Breslow thickness than index melanomas. There were no significant differences in the probability of being diagnosed with a tumor thickness greater than 1.00 mm among melanomas of any of the 4 surveillance melanoma categories and index melanomas. It is likely that this was caused by lack of statistical power, as significance was determined at a = .005 because of multiple testing. In a subanalysis with a cut-off point of 0.75 mm, all surveillance melanoma categories except for noncompliance melanomas were associated with a significantly smaller probability of being diagnosed with a tumor thickness greater than 0.75 mm compared with index melanomas.

The mean tumor thickness of regular-screen melanomas (0.58 mm) was comparable with those of screen-detected melanomas reported in other studies (0.52-0.56 mm).[8,15,16] Hansson et al[17] reported that 93% of 41 melanomas detected in the Swedish national preventive program for melanoma kindred had a tumor thickness less than 1.00 mm, which was comparable to the 89% in our study. These other studies did not specify, however, whether interval melanomas and melanomas in noncompliant patients were included.

First-screen melanomas had a higher mean tumor thickness than regular-screen melanomas, but the difference was not statistically significant. First-screen melanomas did have a significantly lower Breslow thickness than index melanomas, however. These findings are in accordance with earlier studies.[8,15,16]

As much as one fifth of melanomas were diagnosed in patients who were not compliant with follow-up instructions at the time of diagnosis. Moreover, almost half of patients were noncompliant at the time of diagnosis of their first melanoma. Noncompliance had a negative impact on melanoma detection as the probability of being diagnosed with a Breslow thickness greater than 0.75 mm was significantly greater for noncompliance melanomas compared with regular-screen melanomas (OR 4.8). Noncompliance has previously been reported to be a frequent problem in the follow-up of patients with a

primary melanoma,[18] and in the long-term (dermatoscopic) follow-up of patients with atypical pigmented lesions[19,20] as well.

A considerable proportion of melanomas (20%) was diagnosed between scheduled screens. The majority of these interval melanomas was detected by patients themselves after a median interval of 5 months since the last screen. The Breslow thickness of interval melanomas was comparable with that of regular-screen melanomas. This was probably facilitated by the fact that participants of the surveillance program were repeatedly educated about the characteristics of melanoma and instructed to perform regular skin self-examinations and promptly return to the clinic in case of symptomatic, changing, or new fast-growing lesions.

The large number of interval melanomas raises the question whether the standard screening interval of our surveillance program (12 months) is adequate. In this study the median screening interval of regular-screen melanomas was 5 months, because most tumors were diagnosed in patients who were under intensified surveillance because of a previous melanoma. Our results suggest that the majority of melanomas became detectable within 6 months after the preceding screen. Paradoxically we found that tumor thickness was not correlated with the length of the screening interval for intervals less than 24 months. This may have been a result of self-selection bias, however, as patients with a worrisome lesion are more likely to return to the clinic before the scheduled screen (interval melanomas) and are less likely to be noncompliant with follow-up instructions. It may also indicate that health education or increased awareness as a result of earlier melanomas enabled patients to determine themselves when to return. Alternatively this finding could be explained by the growth pattern of melanomas. It has been postulated that most melanomas (except for nodular melanomas) initially only exhibit radial expansion, without substantial vertical expansion.[21]

Based on this theory it could be argued that melanomas can be detectable for a long time, before a substantial increase in their Breslow thickness occurs.
Our study had a retrospective design and as a consequence classification of melanomas into different screening categories was dependent on completeness of data in patient charts. To limit the number of misclassifications we were very restrictive in categorizing doubtful cases and therefore 15% of surveillance melanomas were not further categorized.

Our results suggest a number of ways to improve the surveillance program. First, it is potentially very rewarding to increase efforts to improve patients' compliance with follow-up instructions. Second, early detection of clinically atypical and fast-growing melanomas may be promoted by instructing patients to report to the clinic in case of any changing or new (fast-growing) lesion. As a final point, we believe it is debatable whether our standard screening interval should be shortened from 12 to 6 months. On the one hand the majority of melanomas seemed to be detectable within 6 months after the preceding screen, so a shorter interval would advance melanoma detection. In addition, compliance with follow-up instructions may improve with shorter screening intervals.[19]

On the other hand it is unknown whether shortening of the screening interval would result in detection of tumors in a more favorable stage.[22] Moreover, adequate health education and promotion of skin self-examination may be a more cost-effective alternative than decreasing the screening interval. Further studies will be required to answer these questions.

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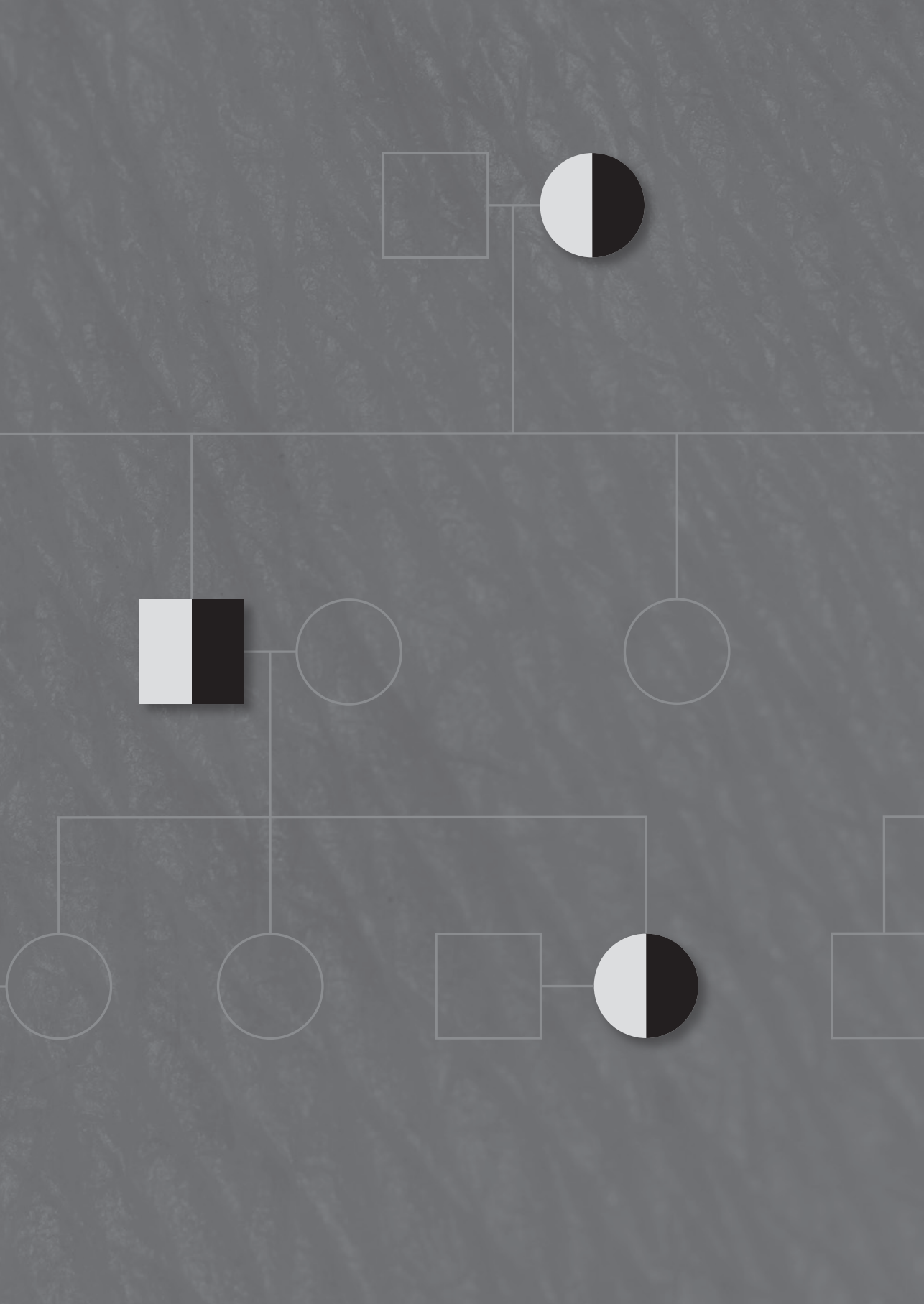
References

1. Ang CG, Kelly JW, Fritschi L, Dowling JP. Characteristics of familial and non-familial melanoma in Australia. Melanoma Res 1998;8:459-64.
2. Hussussian CJ, Struewing JP, Goldstein AM, Higgins PA, Ally DS, Sheahan MD, et al. Germline p16 mutations in familial melanoma. Nat Genet 1994;8:15-21.
3. Kamb A, Gruis NA, Weaver-Feldhaus J, Liu Q, Harshman K, Tavtigian SV, et al. A cell cycle regulator potentially involved in genesis of many tumor types. Science 1994;264:436-40.
4. Bishop DT, Demenais F, Goldstein AM, Bergman W, Bishop JN, Bressac-de Paillerets B, et al. Geographical variation in the penetrance of CDKN2A mutations for melanoma. J Natl Cancer Inst 2002;94:894-903.
5. de Vries E, Houterman S, Janssen-Heijnen ML, Nijsten T, van de Schans SA, Eggermont AM, et al. Up-to-date survival estimates and historical trends of cutaneous malignant melanoma in the south-east of The Netherlands. Ann Oncol 2007;18:1110-6.
6. Goldstein AM, Chan M, Harland M, Hayward NK, Demenais F, Bishop DT, et al. Features associated with germline CDKN2A mutations: a GenoMEL study of melanoma-prone families from three continents. J Med Genet 2007;44:99-106.
7. Gruis NA, van der Velden PA, Sandkuijl LA, Prins DE, Weaver-Feldhaus J, Kamb A, et al. Homozygotes for CDKN2 (p16) germline mutation in Dutch familial melanoma kindreds. Nat Genet 1995;10:351-3.
8. Vasen HF, Bergman W, van Haeringen A, Scheffer E, van Slooten EA. The familial dysplastic nevus syndrome: natural history and the impact of screening on prognosis; a study of nine families in The Netherlands. Eur J Cancer Clin Oncol 1989; 25:337-41.
9. Liu W, Dowling JP, Murray WK, McArthur GA, Thompson JF, Wolfe R, et al. Rate of growth in melanomas: characteristics and associations of rapidly growing melanomas. Arch Derma„tol 2006;142:1551-8.
10. Koch SE, Lange JR. Amelanotic melanoma: the great masquerader. J Am Acad Dermatol 2000;42:731-4.
11. de Snoo FA, Bishop DT, Bergman W, van Leeuwen I, van der Drift C, van Nieuwpoort FA, et al. Increased risk of cancer other than melanoma in CDKN2A founder mutation (p16-Leiden) positive melanoma families. Clin Cancer Res 2008;14:7151-7.
12. Vasen HF, Gruis NA, Frants RR, Der Velden PA, Hille ET, Bergman W. Risk of developing pancreatic cancer in families with familial atypical multiple mole melanoma associated with a specific 19 deletion of p16 (p16-Leiden). Int J Cancer 2000; 87:809-11.
13. Zeger SL, Liang KY. Longitudinal data analysis for discrete and continuous outcomes. Biometrics 1986;42:121-30.
14. Hæjsgaard S, Halekoh U, Yan J. The R package geepack for generalized estimating equations. J Stat Software 2005;15: 1-11.
15. Masri GD, Clark WH Jr, Guerry D, Halpern A, Thompson CJ, Elder DE. Screening and surveillance of patients at high risk for malignant melanoma result in detection of earlier disease. J Am Acad Dermatol 1990;22:1042-8.
16. Carey WP Jr, Thompson CJ, Synnestvedt M, Guerry D, Halpern A, Schultz D, et al. Dysplastic nevi as a melanoma risk factor in patients with familial melanoma. Cancer 1994;74:3118-25.
17. Hansson J, Bergenmar M, Hofer PA, Lundell G, Mansson-Brahme E, Ringborg U, et al. Monitoring of kindreds with hereditary predisposition for cutaneous melanoma and dysplastic nevus syndrome: results of a Swedish preventive program. J Clin Oncol 2007;25:2819-24.
18. Kittler H, Weitzdorfer R, Pehamberger H, Wolff K, Binder M. Compliance with follow-up and prognosis among patients with thin melanomas. Eur J Cancer 2001;37:1504-9.
19. Argenziano G, Mordente I, Ferrara G, Sgambato A, Annese P, Zalaudek I. Dermoscopic monitoring of melanocytic skin lesions: clinical outcome and patient compliance vary according to follow-up protocols. Br J Dermatol 2008;159:331-6.
20. Schiffner R, Schiffner-Rohe J, Landthaler M, Stolz W. Long-term dermoscopic follow-up of melanocytic nevi: clinical outcome and patient compliance. Br J Dermatol 2003;149:79-86.
21. Clark WH Jr, Elder DE, Guerry D, Epstein MN, Greene MH, Van Horn M. A study of tumor progression: the precursor lesions of superficial spreading and nodular melanoma. Hum Pathol 1984;15:1147-65.
22. Richard MA, Grob JJ, Avril MF, Delaunay M, Thirion X, Wolkenstein P, et al. Melanoma and tumor thickness - challenges of early diagnosis. Arch Dermatol 1999;135:269-74.



4

Is surveillance effective, and who should be under surveillance? Results of a national surveillance program for familial melanoma

Submitted

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Abstract

Objective:
To investigate the efficacy of a program launched in 1989 by the foundation for the Detection of Hereditary Tumours (NFDHT) aimed at the surveillance of members of melanoma families and to compare the melanoma detection rate in families with different clinical and genetic characteristics.

Patients and Methods:
From the NFDHT 72 families were selected. A total of 450 individuals were followed for 15 years between 1992 and 2008 at 85 hospitals throughout the Netherlands..

Results:
During follow-up 52 invasive melanomas were diagnosed in 37 individuals. Ten year cumulative melanoma incidence was 10.2% (95% CI: 6.9 – 13.5). Family members with a history of melanoma had a higher probability of being diagnosed with melanoma than their first degree relatives without a history of melanoma (HR: 3.9, 95% CI 2.0 – 7.7).
Median Breslow thickness of surveillance-detected invasive melanomas was 0.50 mm compared to 0.94 mm in pre-surveillance index melanomas. None of the patients with surveillance-detected melanomas died of melanoma during follow-up (median: 4.2 yrs).
Melanoma detection rate was higher in families with a germline CDKN2A mutation compared to CDKN2A wildtype families (HR: 3.6, 95% CI: 1.4 – 9.0) and borderline non-significantly higher for families with ≥ 3 affected family members compared to 2-case families (HR: 2.2, 95% CI: 0.9 – 5.0).

Conclusion:
Our findings are in support of a beneficial effect of surveillance on tumour thickness at diagnosis and survival. Members of CDKN2A mutated families may need more stringent surveillance than members of CDKN2A wildtype families.

Introduction

Being a member of a family with a hereditary predisposition for cutaneous malignant melanoma (CMM) is one of the main risk factors for melanoma. Up to 10% of all melanoma cases have been reported to occur in families.[1]

The prognosis of melanoma patients is highly dependent on the stage at diagnosis.[2] Early melanomas can be cured by a local excision with adequate margins, but for metastatic melanoma the outcome is generally poor. Early detection is considered the most effective way to prevent melanoma mortality. In 1989 a national registry for familial melanoma was established at the Netherlands Foundation for the Detection of Hereditary Tumours (NFDHT) in order to promote the detection and surveillance of members of melanoma families throughout the Netherlands. Regular surveillance, consisting of a minimum of annual total skin examinations complemented by skin self-examinations, has been recommended for all family members with a history of melanoma and their first degree relatives (parent, siblings and children).

So far, two high-penetrance melanoma susceptibility genes associated with an autosomal dominant inheritance have been identified. Pathogenic germline mutations in the tumour suppressor gene CDKN2A (MIM# 600160) are detected in approximately 39% of families with ≥3 melanoma cases, and have an estimated penetrance of 67% by the age of 80 years.[3,4] Mutations in the oncogene CDK4 (MIM# 123829) have been detected only in few families (estimated 2%). For the majority of families the genetic background has not been fully clarified, but appears to be mostly the result of a combination of low (e.g. MC1R) and intermediate (e.g. MITF) risk modifier genes, and (possibly) some rare high penetrance genes.[5,6] In families without a pathogenic germline mutation, melanoma susceptibility is suggested by familial clustering of patients with melanoma, but cannot be confirmed by genetic testing and therefore remains a clinical diagnosis.

Besides genetic differences, clinical experience learns that melanoma pedigrees might also be characterized by clinical differences concerning the number of melanoma patients and the age of melanoma diagnosis of the affected relatives. It is probable that genetic and clinical family characteristics are related to the magnitude of melanoma risk of family members. To the best of our knowledge no prospective studies on these issues have been published.

The goal of this study was to investigate the efficacy of surveillance and to compare the detection rate of melanomas in families with different clinical and genetic characteristics. Analyses were based on prospective NFDHT data of 450 individuals from 72 melanoma families that were followed for 15 years between 1992 and 2008 at 85 hospitals throughout the Netherlands.

Patients and methods

Data collection

The organization and methods of the NFDHT have been published elsewhere.[7,8] In brief, physicians from all parts of the Netherlands refer families suspected for familial melanoma to the registry. Genealogical studies are performed and all reported malignancies are verified by medical records. A genetic predisposition to melanoma is suspected if a family consists of a patient with invasive CMM with at least one first degree relative with invasive CMM or two additional relatives with invasive CMM. If these criteria are met, the registry monitors the continuity of the surveillance program for all family members with a history of melanoma and their unaffected first degree relatives by annually sending letters to the responsible clinician (mostly dermatologists). In return these clinicians report the results of surveillance and histo-pathologic examination.

From the NFDHT 72 families were selected based on the studied clinical and genetic familial characteristics (see below). Families were eligible if they contained a minimum of 2 first degree relatives with confirmed invasive CMM. All family members with: 1. age 12 years or older, 2. either having a personal history of melanoma (affected relative), or being an unaffected first degree relative of a melanoma patient. 3. registration at the NFDHT, and 4. having been subjected to at least one skin examination prior to the end date of the study (1-1-2008), were included in the study. Family-, patient-, tumour- and follow-up data were ascertained from the NFDHT database. For all individuals that were lost to follow-up, letters were sent to their clinician, and in case of no reply, to their general practitioner to obtain additional follow-up data.

Analyses

Cumulative melanoma incidence was calculated using the Kaplan Meier method. Survival times were calculated from the date of registration at the NFDHT until the date of melanoma diagnosis or last date of follow-up (censored). For patients with multiple melanomas only the first surveillance detected melanoma was included in the analyses.
Breslow tumour thicknesses of surveillance detected CMM were compared to pre-surveillance index CMM, which consisted of the first melanoma of the first two CMM patients from each family. For this purpose multivariate linear regression and binary logistic regression analyses were used. In the linear regression analyses a log-transformed Breslow thickness was used. Since differences in the log-transformed variable translate to multiplication factors on the original scale, results are reported as multiplication factors on the original scale. We adjusted for gender, age at diagnosis, order (first or subsequent melanoma) and year of diagnosis. Generalized estimating equations (GEE) were used to correct for within-patient correlations.[9]

We compared the melanoma incidence in families with different genetic and clinical characteristics by investigating: 1. Familial CDKN2A mutation status; coded as positive,

negative (CDKN2A wildtype) or unknown (in case no genetic testing had been performed). All members from a single family were coded equally for this covariate, independent of personal test results. 2. The number of affected relatives (melanoma patients); families were categorized as 2-case families in case of 2 first degree relatives with melanoma, without additional first or second degree relatives with melanoma. In case of one or more additional first or second degree relatives with melanoma, families were categorized as ≥3-case families. 3. The age at melanoma diagnosis of the youngest melanoma patient in the family; families were divided in two categories: youngest melanoma diagnosis at age younger or older than 30 years.

Correlation of these three family characteristics with cumulative melanoma incidence were analysed using multivariate Cox regression analyses with covariates: gender, age, history of melanoma, family CDKN2A mutation status, number of affected relatives, and age of youngest melanoma patient < or ≥30 yrs. To adjust for changes in familial characteristics during follow-up, the follow-up of patients was split up into disjoint (hence independent) follow-up intervals with the moment of change in familial characteristics as stopping time, and analyzed using "delayed entry". Two time dependent covariates were used, one for the number of affected relatives and one for age of the youngest melanoma patient. Generalized estimating equations (GEE) were used to correct for within-family correlations.[9] Based on these analyses adjusted cumulative melanoma incidences were calculated for different risk categories.

All analyses were performed with SPSS 17.0 and R 2.13.0. The package geepack was used for the calculation of adjusted standard errors in R.[10] Statistical significance was determined at $\alpha = .05$, and all tests were two-sided**.** For analyses in which more than two groups were compared a Bonferroni correction for multiple testing was performed.

Results

Data characteristics

In total 450 family members (197 males, 253 females) were included in the study. They consisted of 124 patients with a history of melanoma and 326 unaffected first degree relatives of patients with a history of melanoma. Mean age at registration was 39.7 years (range 12.0 – 80.4 yrs, SD: 15.7 yrs). Median follow-up was 6.3 years (range 0.1 – 15.1 yrs). Follow-up was complete for 336 patients (75%). Eleven patients died during follow-up (details below). For 6 members of CDKN2A mutated families participation in the surveillance program was discontinued after they were tested negative for the mutation.

Data on CDKN2A mutation status was available for thirty-four families (47%). In 15 families (92 patients) the p16-Leiden mutation (c.225-243del19) in the CDKN2A gene was detected.[11] In 19 families (138 patients) melanoma patients were tested negative for a mutation in

CDKN2A. For the remaining 38 families (220 patients) no data on the CDKN2A gene mutation status was available. There were no families with pathogenic CDKN2A mutations other than the p16-Leiden mutation, unclassified variants or CDK4 mutations included in the study. At the time of registration, 49 families (68%) classified as 2-case families, and 23 families (32%) as ≥3-case families. During follow-up eight 2-case families (16%) became ≥3-case families as additional family members were diagnosed with melanoma. In 32 (44%) of the 72 families the youngest melanoma patient was diagnosed with melanoma before the age of 30 years.

Mean age at diagnosis of (first) melanomas was 38.1 years (SD: 12.9, range: 16.5 – 69.3 yrs) in CDKN2A mutated families, 43.2 years (SD: 14.3 yrs, range: 16.4 – 78.2 yrs) in gene-tested CDKN2A wildtype families, and 42.8 years (SD: 13.0, range: 19.7 – 75.6 yrs) in untested families (n = 203, including all first melanomas of included family members and index melanomas). The differences in mean age at diagnosis according to CDKN2A mutation status was not statistically significant (p = 0.11, using a one-way ANOVA test). Of the 203 first melanomas, 2.5% (n = 5) were diagnosed before age 20 years and 3.4% (n = 7) after age 70 years.

Results of surveillance

During follow-up 37 patients (8%) were diagnosed with a total of 52 invasive melanomas (table 1). Twenty-nine patients were diagnosed with a single melanoma, four patients with two melanomas, one patient with three melanomas, and three patients with four melanomas. In addition five in situ melanomas were diagnosed, which were not included in the analyses.

Twenty-three (62%) of the 37 patients that were diagnosed with invasive melanoma during follow-up, had a history of melanoma prior to registration at the NFDHT. Ten year cumulative melanoma incidence during surveillance was 10.2% (95% CI: 6.9 – 13.5).

The median Breslow thickness of surveillance-detected melanomas (n = 51, 1 missing value) was 0.50 mm (range 0.25 – 2.60 mm), compared to 0.94 mm (range 0.18 – 6.00 mm) for index melanomas (n = 124). The Breslow thickness of surveillance-detected melanomas was significantly thinner than that of index melanomas (multiplication factor 0.65, 95% CI 0.44 – 0.96, p = 0.033).

Of the surveillance-detected melanomas 22% (n = 11) had a Breslow-thickness > 1.00 mm, compared with 49% (n = 61) of index melanomas. The higher proportion of melanomas > 1.00 mm among index cases was not statistically significant (odds ratio 2.50, 95% CI 0.61 – 10.29, p = 0.204).

Eleven patients (2.4%) died during follow-up. For seven patients cause of death was metastasized melanoma. These patients all had their melanoma before registration and

Table 1 Characteristics of patients with surveillance detected melanomas

Patient number	Gender	CDKN2A mutation[a]	Number of melanomas[b]	Age at registration	Age at diagnosis	Breslow thickness[c]
60005	Male	Unknown	1	49,3	59,6	Unknown[e]
60008	Female	Unknown	1	49,0	53,7	0,40
60020	Male	Unknown	1	15,9	21,1	1,00
60021	Female	Unknown	2	17,7	26,0[1] / 27,9[2]	0,62[1] / 1,05[2]
80007	Male	Unknown	1	42,0	43,7	0,49
130033	Male	No	1	63,0	76,5	0,27
240003	Male	Unknown	1	48,0	55,4	0,40
390014	Female	Yes	1	43,3	49,2	0,40
410007	Female	No	1	60,0	62,3	1,40
420001	Male	No	2	40,4	43,4[1] / 46,6[2]	0,50[1] / 0,70[2]
480019	Male	Unknown	1	40,9	44,3	0,50
530099	Female	No	1	37,0	37,7	0,40
630025	Female	Unknown	1	29,0	30,4	0,50
770004	Female	Yes	1	27,9	35,9	0,50
880006	Male	Unknown	1	47,8	54,7	0,32
900005	Female	Yes	4	33,3	38,4[1] / 38,4[1] / 40,0[2] / 40,4[3]	0,83[1] / 0,72[1] / 1,00[2] / 0,40[3]
900006	Male	Yes	4	33,1	35,3[1] / 39,2[2] / 39,2[2] / 42,1[3]	0,85[1] / 2,60[2] / 2,00[2] / 0,50[3]
1030001	Male	Yes	1	38,5	43,3	0,45
1050001	Male	No	1	61,6	70,6	0,35
1330003	Female	Unknown	1	52,6	56,0	0,35
1660002	Female	Unknown	1	47,0	56,0	0,45
1890002	Female	Unknown	1	49,1	54,0	0,50
1890028	Female	Unknown	2	51,4	53,8[1] / 54,2[2]	0,79[1] / 0,67[2]
1910006	Male	Unknown	1	44,6	49,8	0,40
1920018	Female	Unknown	1	35,1	37,3	1,10
2110001	Male	Yes	4	43,1	43,1[1,d] / 43,7[2] / 46,5[3] / 47,1[4]	0,50[1] / 0,70[2] / 0,60[3] / 0,75[4]
2190002	Female	Yes	1	52,8	55,0	0,42
2190016	Male	Yes	2	57,6	60,1[1] / 62,8[2]	0,50[1] / 1,20[2]
2580005	Female	Unknown	1	28,5	30,8	0,60
2630102	Male	Yes	1	43,7	46,9	0,30
2650019	Female	Unknown	1	33,0	33,4	0,25
2770101	Female	Yes	1	52,7	56,7	0,25
2840003	Male	Yes	1	33,9	35,2	0,26

Table 1 Continued

Patient number	Gender	CDKN2A mutation[a]	Number of melanomas[b]	Age at registration	Age at diagnosis	Breslow thickness[c]
2840103	Female	Yes	3	26,2	27,9[1] / 28,7[2] / 29,3[3]	0,32[1] / 1,40[2] / 0,92[3]
3030101	Male	No	1	47,6	48,6	1,94
3080065	Male	Yes	1	62,9	63,2	1,60
3160009	Female	Yes	1	19,4	20,2	0,55

[a] ; Familial CDKN2A mutation status, [b] ; Number of melanomas during follow-up, [c] ; Breslow thickness in millimetres, [d] ; The first melanoma of patient 2110001 was diagnosed one month after registration, [e] ; Because of low clinical suspicion this lesions was removed by curettage, and the pathologist was unable to determine the Breslow thickness, [1] ; 1st melanoma, [2] ; 2nd melanoma, [3] ; 3rd melanoma, [4] ; 4th melanoma

none of them was diagnosed with a subsequent primary melanoma during follow-up. Two patients died from oesophagus carcinoma. For two patients cause of death was unknown, but neither of them was diagnosed with melanoma during follow-up. None of the 37 patients that were diagnosed with melanoma during follow-up died of melanoma (median follow-up after diagnosis: 4.2 yrs, range 0.0 – 11.4 yrs, follow-up complete for 33 patients, 4 patients lost to follow-up).

Risk factors

In table 2 the results of the multivariate Cox proportional hazard analyses for family members with a history of melanoma and first degree relatives are presented separately. The analyses showed a significantly increased melanoma detection rate for family members with a history of melanoma who were member of a family in which a relative had been diagnosed with melanoma before age 30 (hazard ratio (HR) 3.6, table 2). An increased melanoma detection rate was also seen for family members with a history of melanoma from CDKN2A-mutated families compared to families without a CDKN2A mutation (HR 5.9), though this finding was not statistically significant after correction for multiple testing. For first degree relatives, no significant personal or familial risk factors could be identified.

An additional Cox proportional hazard analysis combining the data of affected relatives and first degree relatives (table 3) identified a personal history of melanoma (HR 3.93) as the main predictor of melanoma detection. Ten year cumulative melanoma incidence was 23.3% (95% CI: 13.9 – 32.7) for family members with a history of melanoma and 6.0% (95% CI: 2.7 – 9.3) for first degree relatives. In addition, melanoma detection rate was significantly higher in family members from families with the p16-Leiden mutation compared with family members from families without a CDKN2A mutation (HR 3.6). Adjusted ten year cumulative melanoma incidence was 16.2% for CDKN2A mutated families, 7.6% for untested families, and 4.8% for CDKN2A wildtype families (figure 1).

Table 2 Multivariate analyses of personal and familial risk factors for melanoma of affected relatives and first degree relatives.

	Affected relatives[#]			First degree relatives		
Covariate	**HR**	**(95% CI)***	**P**	**HR**	**(95% CI)***	**P**
Family size ≥ 3 cases	0.7	(0.3 – 1.7)	0.44	1.5	(0.5 – 4.4)	0.47
Age youngest relative < 30 yrs	3.6	(1.4 – 9.4)	0.01[$]	0.6	(0.2 – 1.5)	0.29
No CDKN2A mutation in family	1.0					
CDKN2A mutation in family	5.9	(1.2 – 30.2)	0.039[%]	2.4	(0.7 – 8.3)	0.17
Familial CDKN2A status unknown	2.7	(0.5 – 13.9)	0.23	1.0	(0.3 – 4.0)	0.99
Male gender	2.0	(0.8 – 5.0)	0.10	0.4	(0.2 – 1.2)	0.12
Age (years)	1.0	(1.0 – 1.1)	0.97	1.0	(1.0 – 1.1)	0.58

[#] Affected relatives = relatives with a history of melanoma; * HR (95% CI); Hazard Ratio (95% Confidence Interval), [$] statistically significant, [%] not statistically significant after adjustment for multiple testing (significance level at 0.017

Table 3 Multivariate analyses of personal and familial risk factors for melanoma: affected and first degree relatives combined.

Covariate	**HR**	**(95% CI)***	**P**
Personal history of melanoma	3.9	(2.0 – 7.7)	< 0.001[$]
Family size ≥ 3 cases	1.0	(0.5 – 2.1)	0.99
Age youngest relative < 30 yrs	1.8	(0.9 – 3.5)	0.090
No CDKN2A mutation in family		1.0	
CDKN2A mutation in family	3.6	(1.4 – 9.0)	0.006[$]
Familial CDKN2A status unknown	1.6	(0.6 – 4.3)	0.33
Male gender	1.2	(0.7 – 2.2)	0.56
Age (years)	1.0	(1.0 – 1.0)	0.81

* HR (95% CI); Hazard Ratio (95% Confidence Interval), [$] statistically significant

A sub-analysis was performed in which members from CDKN2A mutated families were excluded (table 4). In this analysis a personal history of melanoma again came up as the main predictor of melanoma detection. There were borderline non-significant differences in melanoma detection rate between patients from ≥3-case families and 2-case families (p = 0.074), with an adjusted ten year cumulative melanoma incidence of 9.7% in ≥3-case families and 4.6% in 2-case families.

Table 4 Multivariate analyses of personal and familial risk factors for melanoma in families without a CDKN2A mutation and untested families (families with a CDKN2A mutation excluded).

Covariate	HR	(95% CI)*	P
Personal history of melanoma	4.2	(1.8 – 9.7)	< 0.001[$]
Family size ≥ 3 cases	2.2	(0.9 – 5.0)	0.074
Age youngest relative < 30 yrs	0.8	(0.4 – 1.5)	0.42
No CDKN2A mutation in family	1.0		
CDKN2A mutation in family	excluded		
Familial CDKN2A status unknown	1.9	(0.8 – 4.7)	0.15
Male gender	1.3	(0.6 – 3.0)	0.49
Age (years)	1.0	(1.0 – 1.0)	0.82

* HR (95% CI); Hazard Ratio (95% Confidence Interval), [$] statistically significant

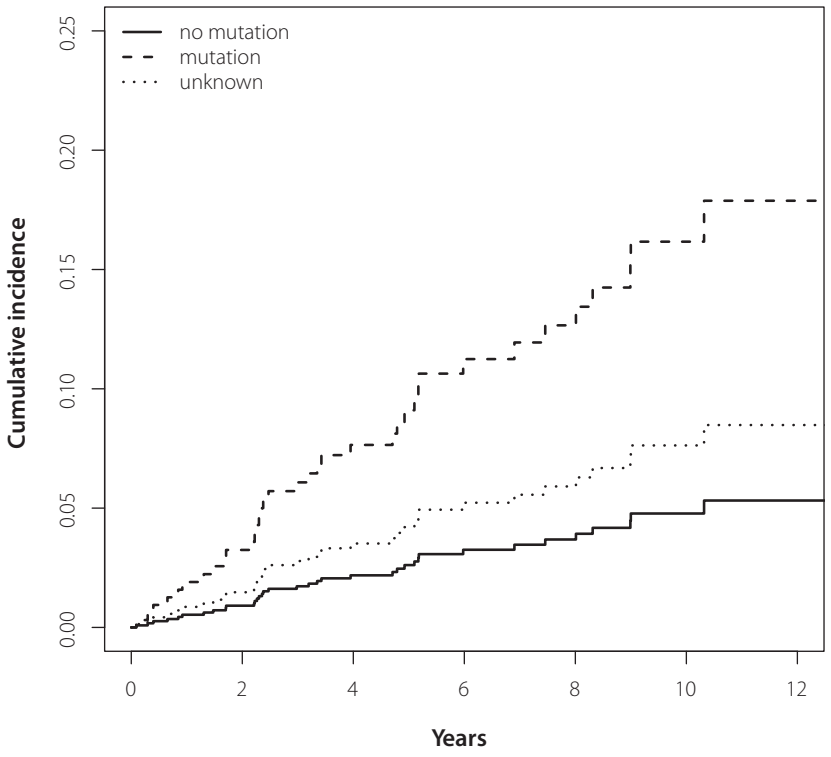
Discussion

In this study we report the results of surveillance of 72 families selected from the NFDHT Dutch national registry for familial melanoma. Overall 450 individuals were followed for 15 years between 1992 and 2008 at 85 hospitals throughout the Netherlands. Only few prospective studies have investigated the effectiveness and safety of surveillance of melanoma families and to the best of our knowledge this is the first prospective study to compare melanoma detection rates in families with different clinical and genetic characteristics.

The 10 year cumulative melanoma incidence during surveillance was 10.2%, which affirms the high melanoma risk and necessity of adequate surveillance of these families. Family members with a history of melanoma at the moment of entering surveillance were more likely to be diagnosed with melanoma during surveillance than their unaffected first degree relatives (HR: 3.9, 95% CI 2.0 – 7.7).

Patient survival is strongly correlated with Breslow tumour thickness, which is illustrated by the fact that 10-year survival is 92% in case of melanomas ≤ 1.00 mm, and 50% if > 4.00 mm.[2] In our study, surveillance-detected invasive CMM's had a statistically significant, almost 40%, thinner Breslow thickness (median 0.50 mm) than melanomas of pre-surveillance index patients (median 0.94 mm). Moreover, the proportion of melanomas with a Breslow thickness > 1.00 mm was 22% in surveillance-detected cases, compared to 49% in index cases, though the difference was not statistically significant after correction

Figure 1 Cumulative melanoma incidence during surveillance according to familial CDKN2A mutation status



No mutation; CDKN2A wildtype, Mutation: p16-Leiden mutation (c.225-243del19) in CDKN2A, Unknown; family has not been genetically tested.

for possible confounders. The median thickness of surveillance-detected melanomas in our study was comparable to other studies (0.50 mm – 0.56 mm) with equally or more stringent surveillance regiments.[12,13,14,15,7] None of the patients with surveillance-detected melanomas died of melanoma during follow-up. This finding needs to be viewed with some reserve as the duration of follow-up was limited (median 4.2 years), 12% of melanoma patients were lost to follow-up and lead time bias may have affected our results. The overall picture of our findings suggests that surveillance results in the detection of a considerable number of CMM, mostly diagnosed at an early stage and with a generally good prognosis.

Less than 40% of melanoma families are characterized by mutations in the high penetrance melanoma susceptibility genes CDKN2A and CDK4, and clinical characteristics like the number of affected relatives and age of melanoma diagnoses, differ considerably between pedigrees. As the clinical significance of these genetic and clinical differences have not been fully clarified, it is uncertain if and how they should affect surveillance guidelines. We compared melanoma detection rates during surveillance in clinically and genetically different families in order to support the development of a tailored surveillance program. The melanoma detection rate was significantly higher in families with a germline mutation in CDKN2A compared to CDKN2A wildtype families. Adjusted ten year cumulative melanoma incidence was 16.2% in CDKN2A mutated families compared to 4.8% in CDKN2A wildtype families. This finding is in accordance with a relatively higher melanoma risk in families with a high-penetrance melanoma susceptibility gene mutation compared to families lacking such mutation and in which melanoma risk is most likely the result of low and intermediate risk modifier genes. The relatively high melanoma detection rate in CDKN2A mutated families may also, to some extent, be attributable to the selection of high risk individuals for surveillance as a result of genetic testing. In our dataset 6 patients (6.5%) from CDKN2A mutated families were released from surveillance during follow-up when they were tested negative for the mutation.

In a subanalysis of CDKN2A wildtype and untested families a borderline non-significant two-fold increased melanoma detection rate (HR: 2.2, 95% CI: 0.9 – 5.0) in ≥3-case compared to 2-case families was found. It seems plausible that the familial melanoma risk is reflected in the number of affected relatives. Statistical non-significance of this finding may be due to lack of power, but more studies are needed to confirm these results.

Our study had several limitations. Only 47% (34/72) of families were genetically tested. Our data suggest that families with more affected relatives were more likely to be tested (the proportion of ≥3-case families in p16-Leiden mutation families, CDKN2A wildtype families and untested families were 40% (6/15), 42% (8/19) and 24% (9/38) respectively). This selection bias most likely resulted in an underestimation of the difference in melanoma detection rate between CDKN2A mutated and wildtype families. We performed sub-analyses (data not shown) in which only gene-tested families were included, but this had little effect on the results.

Prevention of melanomas due to excision of changing and suspicious nevi as happens regularly in daily practice, could not be accounted for in this study design, and may have resulted in an underestimation of the efficacy of surveillance. To limit the effect of overdiagnosis, melanomas in situ were excluded from the analyses.

Concluding, our findings are in support of a beneficial effect of surveillance on tumour Breslow thickness at diagnosis and survival of members of melanoma families. Our results suggest that surveillance should start during puberty and may need to be continued

beyond seventy. Surveillance may need to be more frequent in melanoma patients compared to their first degree relatives without a history of melanoma and CDKN2A mutated families may need more stringent surveillance than CDKN2A wild-type families. Compared to other melanoma families, CDKN2A wild-type two-case families appear to be at a relatively low risk. More studies are needed to facilitate the development of a tailored, cost-effective surveillance program for familial melanoma.

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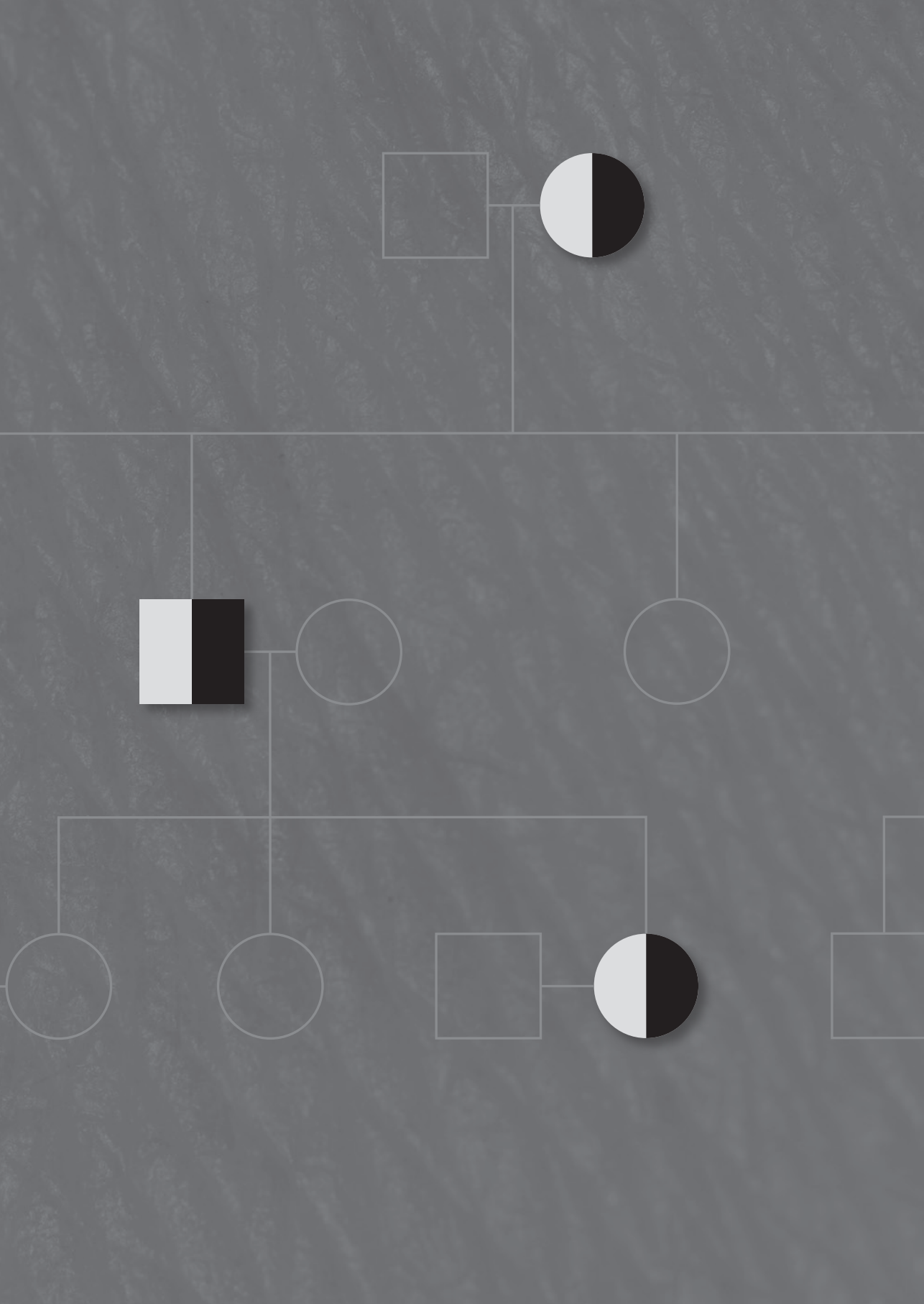
References

1. Ang CG, Kelly JW, Fritschi L, et al: Characteristics of familial and non-familial melanoma in Australia. *Melanoma Res* 1998;8:459-464
2. Balch CM, Gershenwald JE, Soong SJ, et al: Final version of 2009 AJCC melanoma staging and classification. *J Clin Oncol* 2009;27:6199-6206
3. Bishop DT, Demenais F, Goldstein AM, et al: Geographical variation in the penetrance of CDKN2A mutations for melanoma. *J Natl Cancer Inst* 2002;94:894-903
4. Goldstein AM, Chan M, Harland M, et al: Features associated with germline CDKN2A mutations: a GenoMEL study of melanoma-prone families from three continents. *J Med Genet* 2007;44:99-106
5. Udayakumar D, Mahato B, Gabree M, et al: Genetic determinants of cutaneous melanoma predisposition. *Semin Cutan Med Surg* 2010;29:190-195
6. Yokoyama S, Woods SL, Boyle GM, et al: A novel recurrent mutation in MITF predisposes to familial and sporadic melanoma. *Nature* 2011;480:99-103
7. Vasen HF, Bergman W, van Haeringen A, et al: The familial dysplastic nevus syndrome. Natural history and the impact of screening on prognosis. A study of nine families in the Netherlands. *Eur J Cancer Clin Oncol* 1989;25:337-341
8. Vasen HF, Gruis NA, Frants RR, et al: Risk of developing pancreatic cancer in families with familial atypical multiple mole melanoma associated with a specific 19 deletion of p16 (p16-Leiden). *Int J Cancer* 2000;87:809-811
9. Zeger SL, Liang KY: Longitudinal data analysis for discrete and continuous outcomes. *Biometrics* 1986;42:121-130
10. Højsgaard S, Halekoh U, Yan J.: The R Package geepack for Generalized Estimating Equations. *Journal of Statistical Software* 2005;15:1-11
11. Gruis NA, van der Velden PA, Sandkuijl LA, et al: Homozygotes for CDKN2 (p16) germline mutation in Dutch familial melanoma kindreds. *Nat Genet* 1995;10:351-353
12. Carey WP, Jr., Thompson CJ, Synnestvedt M, et al: Dysplastic nevi as a melanoma risk factor in patients with familial melanoma. *Cancer* 1994;74:3118-3125
13. van der Rhee JI, de Snoo FA, Vasen HF, et al: Effectiveness and causes for failure of surveillance of CDKN2A-mutated melanoma families. *J Am Acad Dermatol* 2011;65:289-296
14. Hansson J, Bergenmar M, Hofer PA, et al: Monitoring of kindreds with hereditary predisposition for cutaneous melanoma and dysplastic nevus syndrome: results of a Swedish preventive program. *J Clin Oncol* 2007;25:2819-2824
15. Masri GD, Clark WH, Jr., Guerry D, et al: Screening and surveillance of patients at high risk for malignant melanoma result in detection of earlier disease. *J Am Acad Dermatol* 1990;22:1042-1048



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Suveillance of second degree relatives from melanoma families with a CDKN2A germline mutation

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Abstract

Background:
Life time melanoma risk of mutation carriers from families with a germline mutation in the CDKN2A gene is estimated to be 67%. The necessity to include family members in a melanoma surveillance program is widely endorsed, but there is no consensus on which family members should be invited.

Methods:
In a retrospective follow-up study we investigated the yield of surveillance of first- and second-degree relatives of melanoma and pancreatic cancer patients from 21 families with the 'p16-Leiden' CDKN2A mutation. Melanoma incidence rates were compared with the general population.

Results:
Three-hundred and fifty four first-degree relatives and 391 second-degree relatives were included. Forty-five first-degree relatives and 11 second-degreesecond-degree relatives were diagnosed with melanoma. Most (72%) of second-degree relatives diagnosed with melanoma, had become a first-degree relative before diagnosis, due to the occurrence of a melanoma in a parent or sibling. Overall, melanoma incidence rate was 2.1 per 1000 person years [95% confidence interval (CI), 1.2-3.8] in family members still being second-degreerelatives at diagnosis, compared with 9.9 per 1000 person years (95% CI, 7.4-13.3) in first-degree relatives. The standardized morbidity ratio for melanoma of second-degree relatives compared with the general population was 12.9 (95% CI, 7.2-23.4).

Conclusion:
Second-degree relatives from families with the p16-Leiden mutation in CDKN2A have a considerably increased melanoma risk compared to the general population.

Impact:
This study provides justification for the surveillance of second-degree relatives from families with a CDKN2A germline mutation.

Introduction

Familial melanoma is one of the strongest risk factors for cutaneous melanoma. Approximately 10% of melanoma cases are found in families with two or more patients with melanoma.[1] In up to 40% of families with three or more melanoma cases a mutation in the high penetrance melanoma susceptibility gene CDKN2A (MIM# 600160) is found.[2] With respect to melanomagenesis, CDKN2A has an incomplete penetrance that has been estimated to be 0.67 by age 80 years.[3] In the Netherlands, by far the most prevalent CDKN2A germline mutation is a specific founder mutation (c.225-243del19),[4] known as the p16-Leiden mutation. The p16-Leiden mutation is associated with a very high melanoma risk, comparable with other CDKN2A mutations, and with a cumulative risk of pancreatic cancer of 17% by age 75.[5]

Because of the increased melanoma risk and expected benefit of surveillance,[6,7] regular surveillance of members of familial melanoma families is widely advocated.[8,9] In the Netherlands, the first surveillance program for familial melanoma was initiated at the Leiden University Medical Center (LUMC, Leiden, the Netherlands) in 1981. Individuals that were invited to the program encompassed patients with melanoma, their first-degree relatives (parents, siblings and children) as well as their second-degree relatives (grandparents, uncles, aunts, nieces, nephews and grandchildren). Assuming an autosomal dominant pattern of inheritance (as was later proven to be the case for CDKN2A germline mutations), first and second-degree relatives have a 50% and 25% chance of carrying the genetic risk factor.

The 1981 surveillance guidelines were based on the estimation that life time melanoma risk of mutation carriers approximated 100%, implying an almost 25% life time melanoma risk for second-degree relatives. Since 1981, the recommendation for surveillance of second-degree relatives has remained unchanged.[10]

Since the year 2000 (predictive, CDKN2A) DNA testing of asymptomatic family members is available in the Netherlands. Predictive DNA testing facilitates the selective offering of surveillance to those family members at highest risk of melanoma and therefore has the potential to greatly increase surveillance (cost-) effectiveness. An earlier study at our institution suggested however that the majority of relatives either do not opt for genetic testing, or at an age (i.e., average age 48 years) that lies considerably beyond the young age of onset (i.e. median age of melanoma diagnosis 39 years) in CDKN2A mutated families.[11,12] As a consequence second-degree relatives have continued to present for surveillance.

To the best of our knowledge, surveillance of second-degree relatives has not been addressed in (familial) melanoma guidelines from other countries and is unusual for other types of hereditary cancer syndromes as well. Given this discrepancy between Dutch and foreign guidelines, and to evaluate current surveillance recommendations, we performed

a retrospective follow-up study to evaluate the yield of surveillance of second-degree relatives in families with a founder mutation in CDKN2A (the p16-Leiden mutation).

Materials and Methods

Family ascertainment and data collection

Families were ascertained through the pigmented lesion clinic of the department of dermatology of the Leiden University Medical Centre (LUMC) from 1980 onward. At the clinic family trees were constructed and family members were invited to participate in annual total skin examinations. Since 1985, a decade before the identification of the p16-Leiden mutation, blood samples have been collected for research purposes. Patients consented to this, knowing that carrier information would not be transmitted back to them. Clinicians were also kept unaware of gene carrier status.

Pedigree information was updated on a regular basis. Follow-up data were collected, both during clinic visits, and in several research projects. Confirmation of melanoma diagnoses was gathered through pathology reports and medical records. In 2007 at the event of an earlier study, melanomas of mutation negative family members and all tumors with missing data, in situ melanomas and lentigo malignas were revised by one of us, a member of the pathology panel of the Dutch Melanoma Working Party.[7] Further details on family ascertainment and the collection of follow-up data have been described elsewhere.[5]

Inclusion

Inclusion of family members was based on the presence of the p16-Leiden mutation in their family and independent of personal CDKN2A mutation status (mutated, wildtype or unknown). In the study model all relatives with melanoma or pancreatic cancer were regarded (probable) mutation-carriers and mutation status of all other relatives was regarded to be unknown.

First and second-degree relatives of family members with a medical history of invasive melanoma, melanoma in situ or pancreatic cancer, were included. A minimum age of 12 years was required. To minimize selection bias, not only family members undergoing regular skin check-ups and participants in research projects at the LUMC dermatology department, but also relatives that had not visited the LUMC clinic thus far, were included in the study. Data on nonvisiting family members were obtained from their parents, siblings or children that did visit the clinic and was collected through questionnaires at the occasion of an earlier study.[5] No data were available on the extent of participation of family members at skin examinations by general practitioners or clinicians at other hospitals than the LUMC.

Analysis of melanoma incidence

Melanoma incidence was analyzed in two ways. In the first analysis we used a model in which calculation of follow-up was independent of actual participation in the surveillance program. Follow-up times for all relatives started as soon as their family fulfilled the criteria for familial melanoma, which was defined as: a minimum of two first-degree relatives with either invasive melanoma, or one with invasive melanoma and one with pancreatic cancer, or three (non-first-degree) relatives with invasive melanoma or two with invasive melanoma and one with pancreatic cancer. End of follow-up was defined as: (i) occurrence of an event, i.e. the diagnosis of an invasive or in situ melanoma; (ii) end of follow-up due to closure of the study (Jan 1 2004, based on completeness of data collected for an earlier study), lost to follow-up, death, a diagnosis of pancreatic cancer (indicating a probable p16-Leiden mutation carrier) or having a child or grandchild diagnosed with melanoma or pancreatic cancer (indicating a high likelihood of being a p16-Leiden mutation carrier). An additional reason for end of follow-up for second-degree relatives consisted of the reclassification as first-degree relative. Subsequent follow-up of these relatives was included in the calculation of the melanoma incidence rate of first-degree relatives. For relatives with multiple primary melanomas, only the first melanoma was included in the analysis.

A second analysis was conducted to calculate the melanoma incidence in first and second-degree relatives during actual participation in the surveillance program at the LUMC dermatology department. It was anticipated that this sub-population was enriched for individuals with an increased likelihood of being diagnosed with melanoma, as it was expected that family members with a suspicious nevus or large numbers of (atypical) nevi were more likely to attend surveillance. For this sub-analysis we used data on clinic visits spanning from 1993 till 2004, as from 1993 onwards clinic appointments were recorded in the LUMC hospital information system.

Statistical analysis

Person years were calculated separately for each category (first- and second-degree relatives) and used to compute overall incidence rates and incidence rates per 10 years age groups. For second-degree relatives that became first-degree relatives during follow-up, subsequent follow-up was added to the person years for first-degree relatives, as described earlier. The overall HR for first-degree relatives to develop a melanoma compared with second-degree relatives corrected for age was estimated using a Cox proportional hazard analysis.

Standardized morbidity ratios (SMR) were calculated to estimate the increase in melanoma risk in first and second-degree relatives compared to the general population. The SMR was computed as the ratio of the observed number of cases in either the first- or second-degree relatives over the number of cases expected based on the case rate in the general population, after standardization for age and incidence year distribution. General population data were based on the reported melanoma incidences (with (near) completeness

of data both for invasive and in situ melanomas) per gender and 5-years age categories between 1989 and 2003 at the Netherlands Cancer Registry.[13]

To analyze the probability of second-degree relatives to become a first-degree relative before melanoma diagnosis a multi-state analysis was conducteded with age as time scale.[14] Four states are considered: (i) second-degree relative without melanoma, (ii) first-degree relative without melanoma, (iii) second-degree relative with melanoma, and (iv) first-degree relative with melanoma. Transitions are possible from state 1 to 2 and 3, and from state 3 to 4. Non-parametric estimates of the transition hazards were obtained using the package mstate, version 0.2.6 for R.[15] Subsequently, state occupation probabilities (of the four states) over time were calculated, given second-degree relative without melanoma at t=0, based on the Aalen-Johansen estimator.
Analyses were performed with STATA (version 11), SPSS (version 17) and R (version 2.15.0).

Results

Data characteristics

A total of 21 families was included in the study. Twelve families were included with two first-degree relatives with melanoma, 2 families with three (non-first-degree) relatives with melanoma, 5 families with two first-degree relatives, one with melanoma and one with pancreatic cancer, and 2 families with two relatives with melanoma and one or more relatives with pancreatic cancer.

On the basis of the pedigrees of these 21 families there were 789 eligible family members. Of these, 667 could be included: 354 first-degree relatives (including 78 relatives that turned from second into first-degree relative during follow-up) and 391 second-degree relatives. Data characteristics are reported in table 1.

A total of 56 relatives (45 first-degree, 11 second-degree relatives) were diagnosed with melanoma during follow up; 50 invasive and 6 in situ melanomas (5 in first-degree, 1 in second-degree relatives). Three additional lesions initially diagnosed as in situ melanoma were excluded, as they were reclassified as benign melanocytic lesions after histologic revision. Median age of melanoma diagnosis was 39 years (range 15 – 72) in first-degree relatives and 26 years (range 16 – 44) in second-degree relatives.

In case of the first-degree relatives, 13 patients were diagnosed with melanoma (29%) at their first clinic visit, 22 patients (49%) during surveillance, and for 10 patients (22%) the moment of melanoma detection could not be verified. For second-degree relatives the moment of melanoma detection was; 1 (9%) first clinic visit, 9 (82%) during surveillance and 1 (9%) unverifiable.

Table 1 Data characteristics

Characteristics	First-degree relatives	Second-degree relatives	Total
Total number of relatives in the pedigrees	364 (286 + 78*)	503	867 (789 *)
Number of relatives included in the study	354 (97%)	391 (78%)	745 (667*)
Total follow-up (person years)	4531	5280	9811
Number of relatives with complete follow-up	319	331	650
Length of follow-up median (range)	12.8 y (0.0 – 36.4)	14.9 y (0.0 – 25.7)	-
Age at inclusion median (range)	33 y (12 – 78)	19 y (12 – 71)	-
Gender	177 males (49.7%)	203 males (52.2%)	-

Legend: * Seventy-eight individuals changed from second- to first-degree relative during follow-up. Their person years as second-degree relative were added to the totals of second-degree relatives and person years as first-degree relative to the totals of the first-degree relatives.

Melanoma incidence

Overall, melanoma incidence rate was 9.9 per 1000 person years (95% CI, 7.4 to 13.3) in first-degree relatives and 2.1 per 1000 person years (95% CI, 1.2 to 3.8) in second-degree relatives (Table 2). Overall HR of first-degree relatives to develop a melanoma compared with second-degree relatives, adjusted for age, was found to be 5.1 (95% CI = 2.6 to 10.0, $P < 0.001$).

Overall SMR for melanoma compared with the general population was 101.0 (95% CI, 55.9 – 182.3) in first-degree relatives (observed: 45, expected: 0.76), and 12.9 (95% CI, 7.2 – 23.4) in second-degree relatives (observed: 11, expected: 0.53).

Melanoma detection rates during surveillance

We conducted a subanalysis of relatives that had been under surveillance at the LUMC dermatology clinic. A total of 128 of 277 first-degree relatives (46%) and 113 of 286 second-degree relatives (40%) with follow-up data between 1993 and 2004, attended the surveillance program at least once within this period.

The number of clinic visits per year was 1.12 for first-degree relatives (705 clinic visits in 627 person years) and 0.91 for second-degree relatives (536 clinic visits in 588 person years). Median age of first and second-degree relatives was 32.6 years and 28.8 years, respectively.

Table 2 Incidence rates of melanoma in first- and second-degree relatives according to age

A. Incidence rates of melanoma in first-degree relatives:

Age band	No. of melanoma case patients	No. of person years	Incidence rate / 1000 person years (95% CI)
10-19	4	407.1	9.8 (3.7-26.2)
20-29	7	801.5	8.7 (4.2-18.3)
30-39	14	967.1	14.5 (8.6-24.4)
40-49	8	777.3	10.3 (5.1-20.6)
50-59	6	682.5	8.8 (3.9-19.6)
60-69	5	574.0	8.7 (3.6-20.9)
70-79	1	253.8	3.9 (0.6-28.0)
80-89	0	53.1	0 (0-69.0)
90-99	0	14.6	0 (0-252.7)
TOTAL	45	4531.0	9.9 (7.4-13.3)

B. Incidence rates of melanoma in second-degree relatives:

Age band	No. of melanoma case patients	No. of person years	Incidence rate / 1000 person years (95% CI)
10-19	1	878.5	1.1 (0.2-8.1)
20-29	6	1588.9	3.8 (1.7-8.4)
30-39	2	1551.4	1.3 (0.3-5.2)
40-49	2	838.6	2.4 (0.6-9.5)
50-59	0	267.4	0 (0-13.8)
60-69	0	112.3	0 (0-32.8)
70-79	0	38.2	0 (0-96.6)
80-89	0	5.0	0 (0-737.8)
TOTAL	11	5280.2	2.1 (1.2-3.8)

Melanoma incidence rate was calculated to be 22.3 / 1000 person years (95% CI, 13.2 – 37.7) in first-degree relatives and 8.5 / 1000 person years (95% CI, 3.5 – 20.4) in second-degree relatives. Overall HR of first-degree relatives to develop a melanoma compared with second-degree relatives, adjusted for age, was found to be 2.6 (95% CI, 0.9 to 7.6, p = 0.070).

In family members that had not been under surveillance at the LUMC between 1993 and 2004, in this period one melanoma was diagnosed in first-degree relatives (n = 149) and one melanoma in second-degree relatives (n = 173).

Figure 1

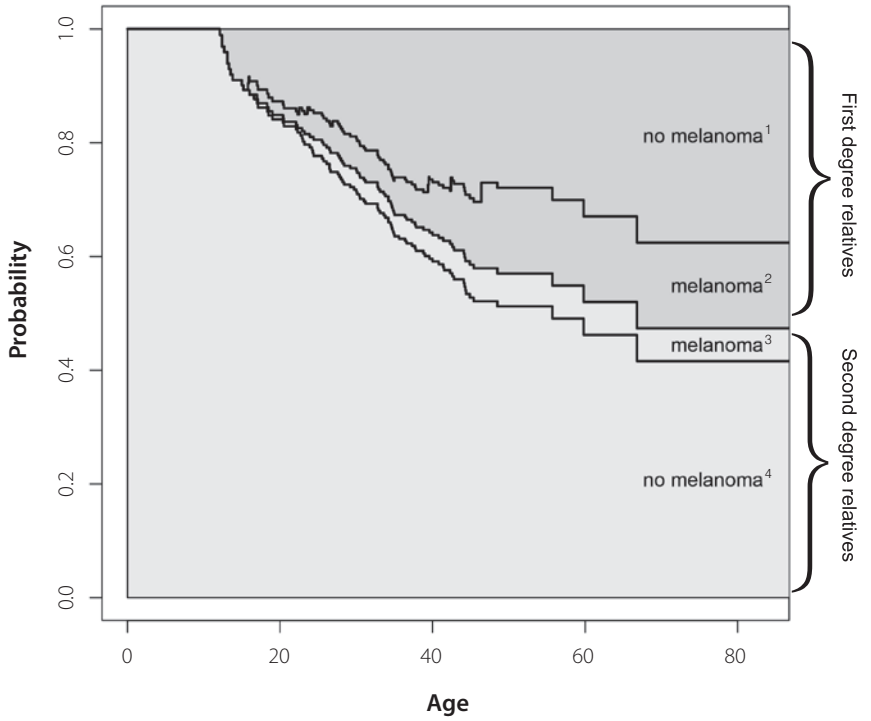

Figure 1 represents the probability of second-degree relatives to belong to either of four states according to their age (12 – 80 years): [1] second-degree relatives who have become first-degree relatives (but were not diagnosed with melanoma); [2] second-degree relatives who have become first-degree relatives and were subsequently diagnosed with melanoma; [3] second-degree relatives diagnosed with melanoma (as second-degree relative); [4] second-degree relatives (who remain second-degree relative and were not diagnosed with melanoma).

Second to first-degree relative transition

During follow up 20% (78/391) of the second-degree relatives became first-degree relatives as the result of a new diagnosis of melanoma or pancreatic cancer in one of their family members. Median age of transition from second to first-degree relative was 30 years (range 12-67).

Besides the 11 second-degree relatives that were diagnosed with melanoma while (still) being a second-degree relative (median age of diagnosis: 26 years (range 16 – 44), there were 11 relatives, who started out as second-degree relative at inclusion, and were diagnosed with melanoma after they had become a first-degree relatives (median age of diagnosis: 35 years (range 16 – 46). There were no differences between these two groups

concerning age at inclusion; (median 15.1 yrs (range 12.0 – 28.4) versus 17.4 yrs (range 12.0 – 39.3), number of first-degree relatives (parent and siblings); 4 relatives (range 3 – 6) versus 3 relatives (range 1 – 8), or number of first-degree relatives older than themselves; 2 relatives (range 1 – 4) versus 3 relatives (range 1 – 4).

In Fig. 1 the probabilities of second-degree relatives to become a first-degree relative and to develop a melanoma as a first or second-degree relative, according to age, are presented. Overall 20.8% of individuals that entered follow-up as second-degree relatives were estimated to be diagnosed with melanoma at age 80 years, 72.2% of whom had been transformed to first-degree relative, before melanoma diagnosis.

Discussion

The goal of this study was to evaluate the yield of surveillance of second-degree relatives in melanoma families with the p16-Leiden mutation in CDKN2A. In the Netherlands, historically all first and second-degree relatives of patients with melanoma with familial melanoma have been recommended to undergo regular skin examinations. Given the expected high life time risk and relatively simple, noninvasive screening procedure at hand, inclusion of second-degree relatives in the surveillance program seemed logical at the time. An intensive literature study suggested that inclusion of second-degree relatives is unusual in other countries and for other types of hereditary cancer, [16,17] but we did not encounter evidence against inclusion of second-degree relatives either.

We report a melanoma incidence rate of 2.1 / 1000 person years for second-degree relatives. The relative risk of first-degree relatives (incidence rate: 9.9 / 1000 person years) compared with second-degree relatives was 5.1, which was considerably higher than anticipated on the basis of Mendelian inheritance (expected RR ≈ 2). To a certain extent this can be explained by our finding that 72% of individuals that entered the study as second-degree relatives that were later diagnosed with melanoma, became a first-degree relative before their melanoma diagnosis. This implies that the majority of melanomas diagnosed in (initially) second-degree relatives were diagnosed after their parent or sibling became melanoma patient, and would not have been missed if only first-degree relatives would have been under surveillance. If transition of second-degree relatives to first-degree relatives was neglected, the overall proportion of second-degree relatives diagnosed with melanoma at age 80 years was estimated to be 20.8%, which is similar to the 17% (*a priori*) risk that would be expected on Mendelian inheritance (25% of the penetrance for melanoma of proven mutation carriers (67%)).[3]

The melanoma detection rate of second-degree relatives that had been under surveillance at the LUMC dermatology clinic was considerably higher (8.5 / 1000 person years) than the estimate for the second-degree relatives population as a whole (2.1 / 1000

person years). It is likely that family members with larger number of (atypical) nevi, who have a higher probability of being a mutation carrier and a higher melanoma risk,[18,19] are more likely to participate in surveillance. However, we have no data on nevus phenotype to support this supposition. In addition we expect that relatives with a suspicious lesion (i.e. possible melanoma) are more likely to participate in surveillance. This notion is supported by the fact that 25% of the melanomas in our study were detected in family members who presented at the clinic for the first time. Early diagnosis may have accounted for part of the higher melanoma detection rates in second-degree relatives that were under surveillance. This is supported by an earlier reported that surveillance of these families was associated with lower tumor Breslow thickness.(7)

The melanoma risk of second-degree relatives was calculated to be 12.9 -fold that of the general population, which is considerably higher than the estimates for individuals with established risk factors such as > 5 atypical naevi (relative risk (RR): 6.4) or with > 100 melanocytic nevi (RR: 6.9).[20]

On the basis of the incidence rates from our study, the number of patients needed to be screened annually to detect one melanoma (number needed to screen; NNS), was 101 in first, and 476 in second-degree relatives. For second-degree relatives that participated in the LUMC surveillance program at the time of diagnosis, the NNS was 118. A recent population-based skin cancer screening intervention study in the German state of Schleswig-Holstein involving 360288 screenees, reported a NNS for malignant melanoma of 620.[21] Taking into consideration that this study involved considerably older subjects (mean age 50 years) that were screened only once, the yield of surveillance of second-degree relatives in our study was considerably higher.

The ultimate goal of melanoma surveillance and screening is to reduce morbidity and mortality. With the lack of evidence from randomized controlled studies, there is still considerable debate on this subject.[22,23] As there are considerable data that suggests screening for melanoma does save lives, offering surveillance to selected high risk populations seems justified.[8]

Retrospective follow-up studies like ours are at risk of several biases. We dealt with possible family selection bias by excluding probands from the analyses. Consistency of (retrospective and prospective) data could be confirmed in an additional analysis (Cox proportional hazard analysis with delayed entry, data not shown) as from the time families were included in the analysis, melanoma incidence rates were constant over time (a straight line fitting within the 95% confidence intervals) for both first and second-degree relatives. This was also the case for the probability of second-degree relatives to become first-degree relatives. In an attempt to correct for selection bias of persons at increased risk of melanoma (as described earlier) relatives that had not been screened at the LUMC were included in the analysis.

Our study may have been prone to overdiagnosis and misclassification bias.[24,25] However, as all in situ melanomas and melanomas of proven mutation negative family members have been revised at the event of an earlier study, we may have reduced misclassification to a minimum. At the same time surveillance is likely to prevent melanomas as a result of the practice to excise changing and clinically suspicious nevi. Taking these consideration into account our results should be viewed with some reserve. It is our overall impression however that our data are sound given the fact that the overall cumulative melanoma incidence of second-degree relatives at age 80 years approximated the expected life-time risk based on data from the literature (see above).

In conclusion, this study provides insights in the family dynamics of surveillance and estimates of melanoma incidence rates and relative risks of second-degree relatives from CDKN2A mutated families that facilitate the discussion on the selection of relatives for surveillance. We belief our results provide justification for the surveillance of second-degree relatives from these very high-risk melanoma families. Further research is necessary to sort out whether these findings equally apply to families without (or other germline-) mutations in the high-penetrance melanoma susceptibility gene CDKN2A.

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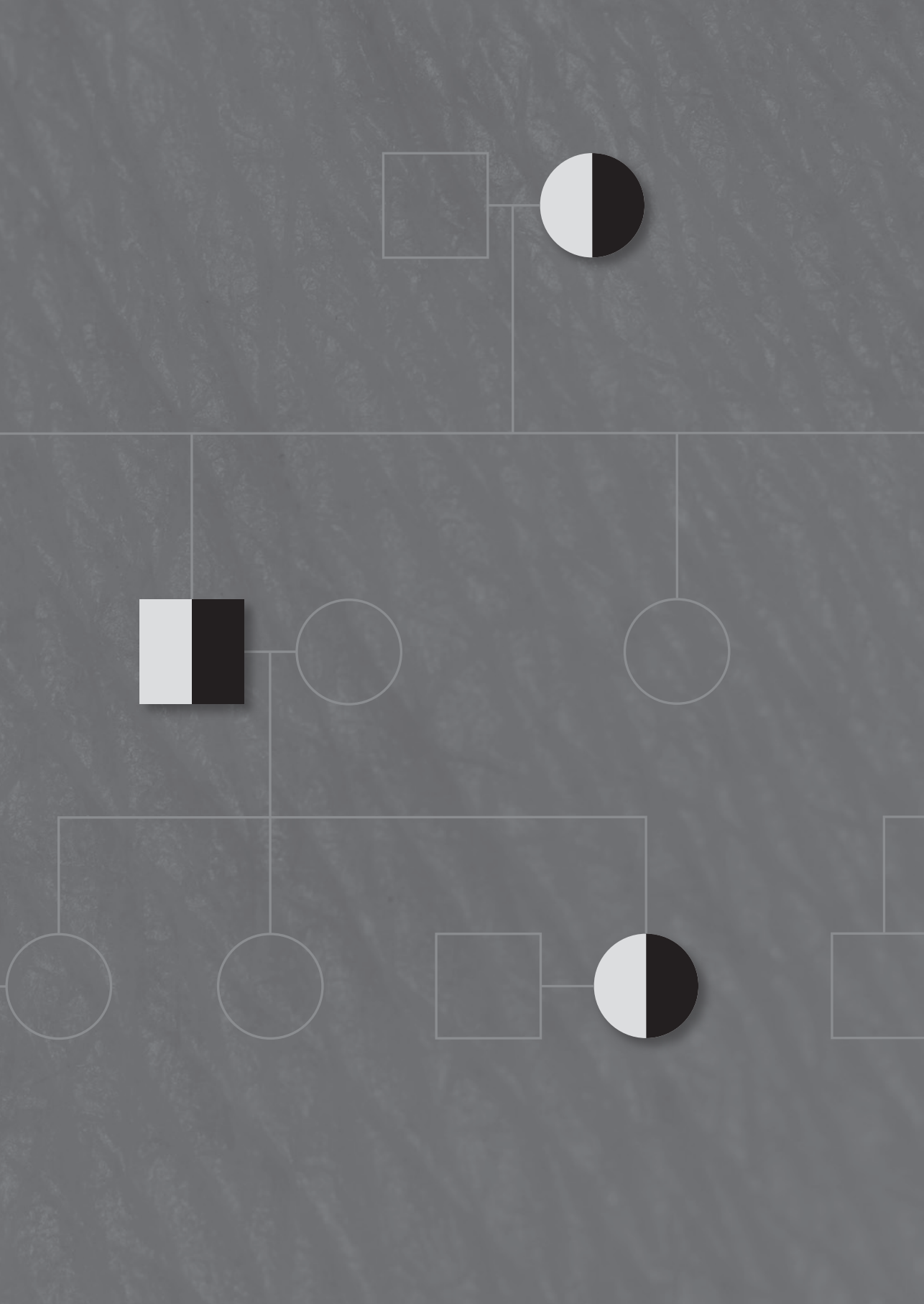
Reference List

1. Ang CG, Kelly JW, Fritschi L, Dowling JP. Characteristics of familial and non-familial melanoma in Australia. Melanoma Res. 1998;8:459-64.
2. Goldstein AM, Chan M, Harland M, Hayward NK, Demenais F, Bishop DT et al. Features associated with germline CDKN2A mutations: a GenoMEL study of melanoma-prone families from three continents. J.Med. Genet. 2007;44:99-106.
3. Bishop DT, Demenais F, Goldstein AM, Bergman W, Bishop JN, Bressac-de Paillerets B et al. Geographical variation in the penetrance of CDKN2A mutations for melanoma. J.Natl.Cancer Inst. 2002;94:894-903.
4. Gruis NA, van der Velden PA, Sandkuijl LA, Prins DE, Weaver-Feldhaus J, Kamb A et al. Homozygotes for CDKN2 (p16) germline mutation in Dutch familial melanoma kindreds. Nat.Genet. 1995;10:351-3.
5. de Snoo FA, Bishop DT, Bergman W, van L, I, van der DC, van Nieuwpoort FA et al. Increased Risk of Cancer Other Than Melanoma in CDKN2A Founder Mutation (p16-Leiden)-Positive Melanoma Families. Clin.Cancer Res. 2008;14:7151-7.
6. Hansson J, Bergenmar M, Hofer PA, Lundell G, Mansson-Brahme E, Ringborg U et al. Monitoring of kindreds with hereditary predisposition for cutaneous melanoma and dysplastic nevus syndrome: results of a Swedish preventive program. J.Clin.Oncol. 2007;25:2819-24.
7. van der Rhee JI, de Snoo FA, Vasen HF, Mooi WJ, Putter H, Gruis NA et al. Effectiveness and causes for failure of surveillance of CDKN2A-mutated melanoma families. J Am.Acad.Dermatol. 2011;65:289-96.
8. Curiel-Lewandrowski C, Chen SC, Swetter SM. Screening and Prevention Measures for Melanoma: Is There a Survival Advantage? Curr.Oncol.Rep. 2012;14:458-67.
9. Katalinic A, Waldmann A, Weinstock MA, Geller AC, Eisemann N, Greinert R et al. Does skin cancer screening save lives?: An observational study comparing trends in melanoma mortality in regions with and without screening. Cancer 2012.
10. Dutch Melanoma Society. *Guidelines Melanoma of the skin*. Alphen aan den Rijn: van Zuiden, 2005.
11. de Snoo FA, Riedijk SR, van Mil AM, Bergman W, ter Huurne JA, Timman R et al. Genetic testing in familial melanoma: uptake and implications. Psychooncology. 2008;17:790-6.
12. van der Rhee JI, Krijnen P, Gruis NA, de Snoo FA, Vasen HF, Putter H et al. Clinical and histologic characteristics of malignant melanoma in families with a germline mutation in CDKN2A. J Am.Acad.Dermatol. 2011;65:281-8.
13. Integraal Kankercentrum Nederland. http://www.cijfersoverkanker.nl/selecties/dataset_1/img5207707bb5daa. (access date oktober 18 2012).
14. Putter H, Fiocco M, Geskus RB. Tutorial in biostatistics: competing risks and multi-state models. Stat.Med. 2007;26:2389-430.
15. de Wreede LC, Fiocco M, Putter H. The mstate package for estimation and prediction in non- and semi-parametric multi-state and competing risks models. Comput.Methods Programs Biomed. 2010;99:261-74.
16. Marsden JR, Newton-Bishop JA, Burrows L, Cook M, Corrie PG, Cox NH et al. Revised U.K. guidelines for the management of cutaneous melanoma 2010. Br.J Dermatol. 2010;163:238-56.
17. Clinical Practice Guidelines for the Management of Melanoma in Australia and New Zealand. http://www.nhmrc.gov.au/_files_nhmrc/publications/attachments/cp111.pdf . (access date oktober 18 2012).
18. Bishop JA, Wachsmuth RC, Harland M, Bataille V, Pinney E, MacK P et al. Genotype/phenotype and penetrance studies in melanoma families with germline CDKN2A mutations. J.Invest Dermatol. 2000;114:28-33.
19. Demenais F, Mohamdi H, Chaudru V, Goldstein AM, Newton Bishop JA, Bishop DT et al. Association of MC1R variants and host phenotypes with melanoma risk in CDKN2A mutation carriers: a GenoMEL study. J Natl. Cancer Inst. 2010;102:1568-83.
20. Gandini S, Sera F, Cattaruzza MS, Pasquini P, Abeni D, Boyle P et al. Meta-analysis of risk factors for cutaneous melanoma: I. Common and atypical naevi. Eur.J.Cancer 2005;41:28-44.
21. Waldmann A, Nolte S, Geller AC, Katalinic A, Weinstock MA, Volkmer B et al. Frequency of excisions and yields of malignant skin tumors in a population-based screening intervention of 360,288 whole-body examinations. Arch.Dermatol. 2012;148:903-10.
22. Curiel-Lewandrowski C, Kim CC, Swetter SM, Chen SC, Halpern AC, Kirkwood JM et al. Survival is not the only valuable end point in melanoma screening. J.Invest Dermatol. 2012;132:1332-7.

23. Wolff T, Tai E, Miller T. Screening for skin cancer: an update of the evidence for the U.S. Preventive Services Task Force. Ann.Intern.Med. 2009;150:194-8.
24. van der Rhee JI, Mooi WJ, Kukutsch NA, de Snoo FA, Bergman W. Iatrogenic melanoma. Comment on: Melanoma epidemic: a midsummer night's dream? Br.J Dermatol. 2010;162:457-8.
25. Welch HG, Black WC. Overdiagnosis in cancer. J Natl.Cancer Inst. 2010;102:605-13.



6

Iatrogenic melanoma. Comment on: Melanoma epidemic: a midsummer night's dream?

British Journal of Dermatology 2010 Feb 1;162(2):457-8

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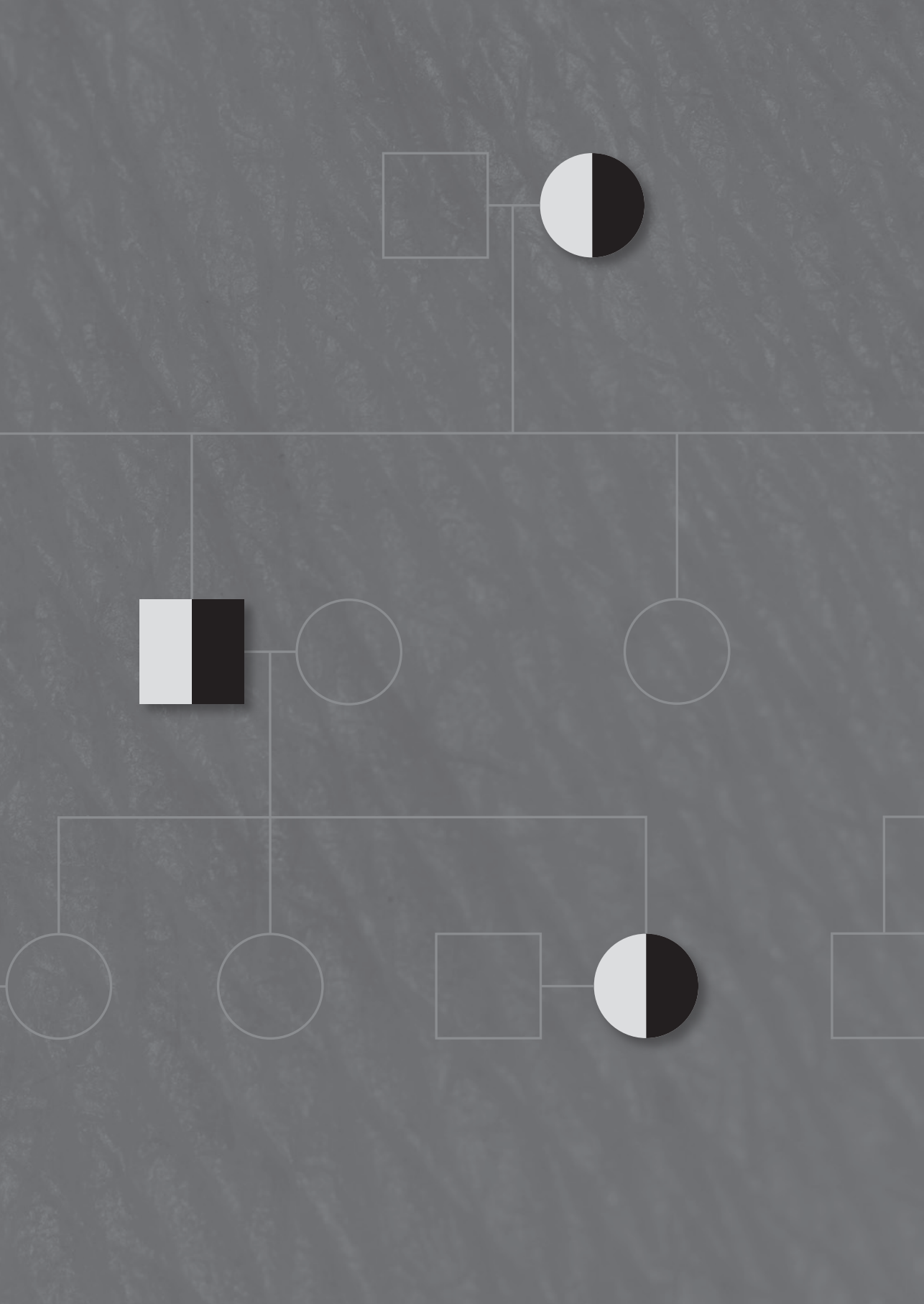
MADAM, We read with great interest the recent article about the controversy over the explanation of the melanoma epidemic.[1] The past decades have witnessed a substantial increase in the reported incidence of cutaneous malignant melanoma (CMM) without a proportional rise in melanoma mortality in most European countries. The paper suggests that the large increase is likely to be due to diagnostic drift which classifies benign lesions as stage 1 melanoma.[1,2]

Histology is the gold standard for the diagnosis of CMM, but the assessment of small and thin melanocytic lesions, that constitute a growing proportion of lesions submitted for histology, is problematic, and interobserver agreement is moderate at best.[3,4] Histological indicators of malignancy have largely been derived from larger lesions, and it is unknown if they are equally applicable for small lesions. As the consequences of overdiagnosis are generally limited to a small local re-excision and increased patient stress, whereas underdiagnosis results in an increased chance of recurrence and death, judgement tends to be skewed towards malignancy.[2,4] In our clinic, members of melanoma families have been under surveillance since 1981. In many of these families, a mutation (p16-Leiden) in the high-penetrance melanoma susceptibility gene CDKN2A has been identified.[5] During surveillance of 37 families with a p16-Leiden mutation, melanomas have been diagnosed in 105 genetically tested relatives, 12 of whom (11%) were noncarriers. These 12 noncarriers had a total of 13 melanomas. As part of a study on the effect of surveillance (manuscript in preparation) the slides of 126 melanomas were reviewed. These consisted of all in situ melanomas (n = 63), and invasive melanomas with missing data or of a nonsuperficial spreading histological type (n = 52) that had been diagnosed in mutation carriers within these 37 families. All melanomas of the 12 noncarriers that were available for histological review (seven in situ and four invasive melanomas from 10 patients) were added to the set. Slides were revised by a pathologist who is a member of the Dutch melanoma panel (W.J.M.). Revisions were performed blinded for the patients' mutation status. After disclosure of the mutation status a disproportionately high proportion of (in situ) melanomas reclassified as benign melanocytic lesions turned out to be cases of noncarriers. Eight (seven in situ and one invasive) of the 11 melanomas of noncarriers were reclassified as benign (73%), compared with only 13 of the other 115 cases (11%). In seven of 10 mutation-negative relatives a history of melanoma was therefore not confirmed.

These results touch on two important issues. Firstly, the value of genetic testing for CDKN2A mutations has been discredited because of a reported increased melanoma incidence among mutation-negative relatives.[6] Our data show that overdiagnosis may account for a significant proportion of this observation. Secondly, increased screening and surveillance of individuals with a low a priori melanoma risk may result in removal of increased numbers of small and histologically equivocal lesions, some of which will be overdiagnosed as cancers and (especially in the case of individuals with a single relative with melanoma) will contribute to the chance of an inappropriate picture of familial clustering.

References

1. Levell NJ, Beattie CC, Shuster S, Greenberg DC. Melanoma epidemic: a midsummer night's dream? Br J Dermatol 2009; 161:630–4.
2. Swerlick RA, Chen S. The melanoma epidemic. Is increased surveillance the solution or the problem? Arch Dermatol 1996; 132:881–4.
3. A nationwide survey of observer variation in the diagnosis of thin cutaneous malignant melanoma including the MIN terminology. CRC Melanoma Pathology Panel. J Clin Pathol 1997; 50:202–5.
4. Brochez L, Verhaeghe E, Grosshans E et al. Inter-observer variation in the histopathological diagnosis of clinically suspicious pigmented skin lesions. J Pathol 2002; 196:459–66.
5. Gruis NA, van der Velden PA, Sandkuijl LA et al. Homozygotes for CDKN2 (p16) germline mutation in Dutch familial melanoma kindreds. Nat Genet 1995; 10:351–3.
6. Kefford R, Bishop JN, Tucker M et al. Genetic testing for melanoma. Lancet Oncol 2002; 3:653–4.



7

The impact of dermoscopy on the management of pigmented lesions in everyday clinical practice of general dermatologists: a prospective study

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Summary

Background:
Dermoscopy greatly improves the clinical diagnosis of pigmented lesions. Few studies have investigated, however, how dermoscopy is guiding management decisions in everyday clinical practice. In addition, most studies have been performed in the setting of dermoscopy experts working in pigmented lesion clinics.

Objectives:
To assess the impact of dermoscopy on clinical diagnosis and management decisions for pigmented lesions in everyday practice of general dermatologists.

Methods:
We performed a prospective study in general dermatology clinics in community hospitals run by dermatologists with intermediate dermoscopy experience and expertise. Each clinician independently included suspicious lesions from consecutive patients. Pre-and postdermoscopy diagnoses and management decisions were recorded. Pathology was used as reference diagnosis.

Results:
In total, 209 suspicious lesions were included in the study by 17 dermatologists. Fourteen lesions were histologically proven in situ or invasive malignant melanomas. Based on clinical diagnoses, dermoscopy improved sensitivity from 0.79 to 0.86 (P =1.0). All 14 melanomas were intended to be excised based on naked eye examination alone, independent of dermoscopic evaluation. Specificity increased from 0.96 to 0.98 (P =0.22). Dermoscopy resulted in a 9% reduction of the number of excisions.

Conclusions:
Dermoscopy reduced the number of excisions, but did not improve the detection of melanomas. Our results suggest that in everyday clinical practice of general dermatologists the main contribution of dermoscopy is a reduction of unnecessary excisions.

Introduction

Several studies have demonstrated that dermoscopy is better at discriminating between melanoma and benign pigmented lesions than naked eye examination (NEE).[1–3] However, very few studies have investigated how dermoscopy is guiding management decisions in everyday clinical practice.[4–7]

The first dermoscopy studies were performed predominantly in experimental settings. Clinicians judged lesions based on macroscopic and dermoscopic images instead of live patients, study sets often contained a disproportionately high number of melanomas (high pretest probability) and in some studies dermoscopic images were not preceded by their accompanying macroscopic images.[8–12]

More recent studies have evaluated dermoscopy in more realistic clinical settings.[3] Most of these studies have focused on the ability of dermoscopy to improve the clinical diagnosis of pigmented lesions.[1–3,8–17]

Several authors have suggested that ultimately the purpose of dermoscopy is to improve the ability to determine whether lesions need to undergo a biopsy procedure.[5,18–21] In other words, dermoscopy improves the detection of melanomas only if melanomas that would not have been biopsied based on NEE are biopsied because of their dermoscopic characteristics. Dermoscopy improves the malignant /benign ratio of excised lesions if it results in leaving benign lesions in situ that would have been biopsied based on NEE.

Most previous dermoscopy studies have been performed in the setting of specialized pigmented lesion clinics (PLCs) run by dermoscopy experts. To our knowledge there are no studies on the impact of dermoscopy on the clinical practice of general dermatologists with intermediate experience and excellence in dermoscopy. It is not unlikely that most (potential) dermoscopy users belong to this specific group of clinicians. The aim of this study was to assess prospectively the impact of dermoscopy on the clinical diagnosis and management of pigmented lesions in everyday clinical practice of general dermatologists.

Materials and methods

Participants in the study were dermatologists working in general dermatology clinics in community hospitals located in different parts of the Netherlands. They all had been performing dermoscopy for at least 6 months and had recently participated in a full-day dermoscopy course covering the basic dermoscopic characteristics of melanocytic and nonmelanocytic lesions, the ABCD rule for dermoscopy,[22] pattern analysis[23] and the more recently described (vascular) patterns and structures.[24–26]

For the current study they were instructed to include 20 consecutive eligible lesions of patients visiting their regular clinics. Patients were either newly referred (in the Dutch health system dermatological service is accessible only after referral by a general practitioner) or already under treatment at the department of the participating dermatologist. The initial or primary reason for patients to attend the dermatologist was irrelevant for the eligibility of lesions.

Lesions were eligible if they fulfilled the following criteria: (i) they had to be suspicious pigmented lesions, (ii) for which the participant would normally also apply dermoscopy, and (iii) if a patient had more than one eligible lesion the most suspicious lesion had to be selected. After participants identified an eligible lesion, they first had to evaluate it on the basis of NEE and to record the NEE preferential diagnosis and management strategy as if there were no opportunity to perform dermoscopy afterwards. NEE was guided by the ABCDE criteria, the ugly duckling sign and symptoms reported by patients.[27] Subsequently dermoscopy was performed and the preferential diagnosis and management strategy based on the combined NEE and dermoscopic evaluation were recorded (Fig. 1).

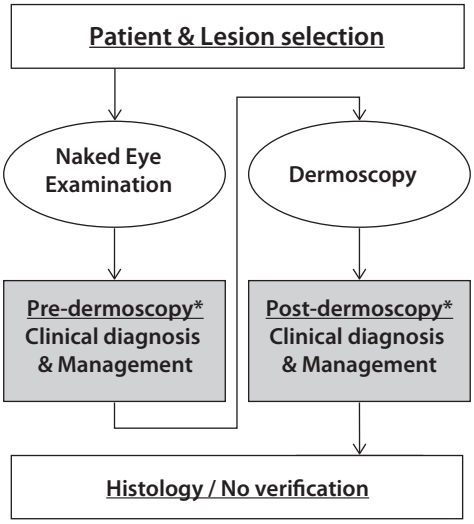
In cases where a biopsy was performed, participants were requested to send us a copy of the pathology report and to indicate whether they performed the biopsy for diagnostic purposes or for other reasons (e.g. cosmetic). Participants were requested to add a description of their method of sampling, in order to check whether they had followed instructions to include lesions in a consecutive order.

Data analysis

Preferential diagnoses were categorized as 'melanoma' or 'nonmelanoma'. Management strategies were also grouped into two categories: (i) 'intervention': a diagnostic (punch, shave or excisional) biopsy with the primary intention of histological verification and treatment of a possible melanoma and (ii) 'no intervention': follow-up, no follow-up, or an intervention for a nondiagnostic reason (e.g. cosmetic). For biopsied lesions histological diagnosis was used as the reference diagnosis.

True positives (TP) were defined as lesions classified as melanoma, and confirmed as melanoma on histological examination. True negatives (TN) were defined as lesions that were classified as 'nonmelanoma', with a subsequent diagnosis other than melanoma on histological examination or left unbiopsied because there was no suspicion of melanoma. False positives (FP) were defined as lesions classified as melanoma, but not diagnosed as melanoma on histology, or not biopsied (after dermoscopic evaluation). False negatives (FN) were defined as lesions that were classified as 'nonmelanoma', but were diagnosed as melanoma on histology. Sensitivity was computed as TP /(TP+FN) and specificity as TN /(TN+FP). Sensitivity and specificity were also calculated from a management perspective, with the clinical diagnosis 'nonmelanoma' being exchanged for 'no intervention' and the

Figure 1 Study design



*Pre- and post dermoscopy clinical diagnosis and management decisions were compared.

clinical diagnosis 'melanoma' for 'intervention'. To compare sensitivity and specificity before and after dermoscopy a statistical analysis was performed, using the McNemar test. Analyses were performed with SPSS 14.0 (SPSS, Chicago, IL, U.S.A.), and statistical significance was determined at a =0.05, and two-sided.

The impact of dermoscopy on management was analysed according to the two management categories as defined above ('intervention' and 'no intervention'), in two ways. The impact of dermoscopy on the detection of melanomas was calculated as the proportion of histologically confirmed melanomas that would not have been biopsied (management category: 'intervention') without the use of dermoscopy. In addition to this we calculated the proportional reduction of the number of 'interventions' due to dermoscopy.

Results

Data characteristics

Seventeen general dermatologists with a median experience in dermoscopy of 7.5 years (range 6 months–14 years) participated in the study. Twelve clinicians (71%) reported a methodology that implied consecutive sampling. Five clinicians (29%) stated they had followed inclusion instructions, but gave no detailed description of their method of sampling.

Participants judged a mean number of 12 lesions (range 4– 20), with a total of 209 lesions. Data on clinical diagnosis and management were complete for 207 (99%) and 196 lesions (94%), respectively. In total, 99 lesions were biopsied: 72 for diagnostic purposes, 20 for other reasons (e.g. cosmetic) and seven cases in which the distinction could not be made due to incomplete data. Among the 72 lesions that were excised for diagnostic purposes there were ten invasive and four in situ melanomas (Table 1). In addition, there was one borderline lesion, described in the histology report as 'a dysplastic naevus with severe atypia, melanoma in situ not excluded'. There were no melanomas among the other 27 biopsied lesions.

Table 1 Characteristics of histologically proven melanomas diagnosed in the general dermatology setting

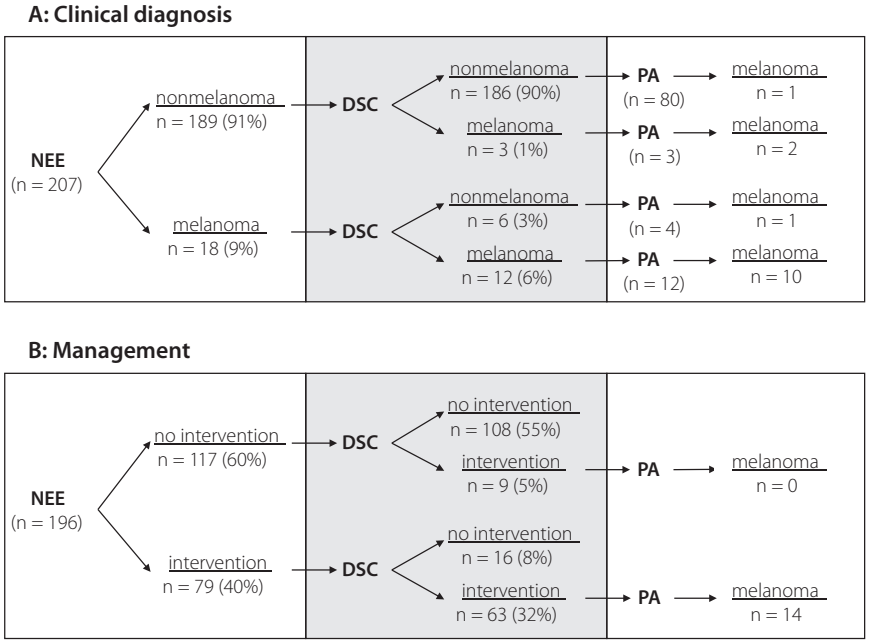
Histological type	n	Breslow thickness
SSM	4	0.15 mm, 0.85 mm, 0.98 mm and 2.8 mm
Mis	4	not applicable
LMM	1	not available
NM	1	1.1 mm
NOS	4	0.40 mm, 0.95 mm, 0.98 mm and 1.1 mm
Total	14	

SSM, superficial spreading melanoma; Mis, melanoma in situ; LMM, lentigo maligna melanoma; NM, nodular melanoma; NOS, not otherwise specified

The impact of dermoscopy on the clinical diagnosis

Based on NEE 18 lesions were classified as melanomas (Fig. 2a). After dermoscopy only 12 of these were still regarded to be melanomas, 10 of which were confirmed by histopathology. The other two lesions were diagnosed as a dysplastic naevus and a collision tumour consisting of a seborrhoeic keratosis (SK) and a basal cell carcinoma (BCC). Of the six lesions no longer classified as melanomas after dermoscopy, four were biopsied.

Figure 2 The effect of dermoscopy on (**A**) the clinical diagnosis and (**B**) management decisions



NEE, naked eye examination; DSC, dermoscopy; PA, pathology

One was diagnosed as a BCC, one as a common naevus and one as a collision tumour consisting of a BCC, an SK and a sebaceous adenoma. The fourth lesion, which was clinically diagnosed as a common naevus after dermoscopy, proved to be a malignant melanoma on histology (Breslow thickness 1.1 mm). The two lesions that were not biopsied were clinically diagnosed as lentigo maligna before dermoscopy, but were regarded as benign lentigines after dermoscopy and left in situ.

After dermoscopy three lesions regarded as 'nonmelanoma' by NEE were reclassified as melanoma. Two of these were confirmed by histology (Breslow thickness 0.40 mm and an in situ melanoma) and one was diagnosed as a dysplastic naevus. There was one lesion that was incorrectly diagnosed as a dysplastic naevus, both before and after dermoscopy, but turned out to be a melanoma (Breslow thickness 0.95 mm).

Sensitivity was calculated to be 0.79 (11 /14) for NEE alone and 0.86 (12 /14) for NEE aided by dermoscopy. Specificity was 0.96 (186 /193) before and 0.98 (190 /193) after dermoscopy

had been performed. Statistical analysis demonstrated that the improvements of sensitivity and specificity by the addition of dermoscopy were not statistically significant (P =1.0 and P =0.22, respectively).

The impact of dermoscopy on management decisions

In 13% (n = 25) of lesions management changed after dermoscopy had been performed (Fig. 2b): for 16 lesions (8%) a diagnostic biopsy was abandoned and for nine lesions (5%) a diagnostic biopsy was induced. Histological evaluation of these nine lesions demonstrated three common naevi, three dysplastic naevi, one benign lentigo, one congenital naevus and the borderline lesion that was described before. The predermoscopy management for this borderline lesion was noted as 'follow-up'. Dermoscopy had no influence on the management of the 14 histologically confirmed melanomas, as all were intended to be excised (diagnostic biopsy) based on the NEE, before dermoscopy had been performed.

Before dermoscopy 40% (79 /196) of included lesions were intended to be excised (diagnostic biopsy). After dermoscopy 37% (72 /196) of the lesions were excised. The malignant /benign ratio of excised lesions decreased from 1 : 5.6 (14 /79) before to 1:5.1 lesions (14 /72) after dermoscopy had been performed. Dermoscopy resulted in a reduction of the total number of diagnostic biopsies of 9% (7 /79). Neither sensitivity nor specificity (P =1.0 and P =0.23) was increased by dermoscopy, if calculated based on management decisions instead of clinical diagnoses.

Discussion

To our knowledge this is the first prospective study that has evaluated how dermoscopy influences the clinical diagnosis and is guiding management decisions made by general dermatologists in their routine daily practice.

Sensitivity increased after addition of dermoscopy to the NEE, although not statistically significantly. The sensitivities of NEE (0.79) and dermoscopy (0.86) in our study were comparable with the summary estimates of sensitivity in a recent meta-analysis of clinical dermoscopy studies by Vestergaard et al.[3] (0.71 and 0.90, respectively).

There were two melanomas in our study that had wrongfully been classified as benign lesions based on NEE, but were correctly classified as melanoma due to dermoscopy. In one instance, however, a melanoma, correctly classified based on NEE, was reclassified as a benign melanocytic lesion after dermoscopy had been performed. This did not affect the decision to excise this particular lesion, but it illustrates the danger of false reassurance due to dermoscopy.

Dermoscopy did not improve the detection of melanomas, as all 14 melanomas were intended to be excised before dermoscopy was performed. Dermoscopy did, however, result in the decision to excise a dysplastic naevus with severe atypia, that would have

been left in situ to be followed up if dermoscopy had not been performed. This excision may have prevented the development of an invasive melanoma, but this is, of course, speculative.

Specificity slightly improved as a result of performing dermoscopy, but the difference was not statistically significant. Our estimates of specificity (0.96 before and 0.98 after dermoscopy) were higher than the summary estimates in the meta-analysis of Vestergaard et al.[3] (0.81 and 0.90, respectively). The difference can be explained by the fact that our study was based on all suspicious lesions for which dermoscopy was used, including those that were not biopsied. Many of the studies in the meta-analysis only included lesions that were biopsied. Our results were comparable with a study by Stanganelli et al.[14] (specificity of 0.99 before and 1.00 after dermoscopy) that included unbiopsied lesions in their analyses as well.

Dermoscopy reduced the number of excisions by 9%, which is considerably lower than the figure reported in other studies.[4–6] In a randomized study Carli et al.[4] reported that 38% fewer excisions were performed in the dermoscopy study arm compared with the NEE arm. Two prospective studies that investigated the influence of dermoscopy on the management of lesions preselected for excision by NEE found a reduction of the number of excisions of 40% and 70%.[5,6]

The a priori possibility for dermoscopy to reduce the number of (unnecessary) excisions in our study was probably limited, due to the fact that the malignant /benign ratio of (intended to be) excised lesions was very low before dermoscopy had been performed (1 : 5.6). There are a number of possible explanations for this. Most previous dermoscopy studies were performed in PLCs. A considerable proportion of patients seen at PLCs have a high a priori melanoma risk and are therefore regularly screened. As a consequence it is likely that the spectrum of melanomas diagnosed in general dermatology clinics differs from those in PLCs. Melanomas presented in general dermatology clinics may be in a more advanced stage, with more clear-cut clinical characteristics, making it easier to diagnose them based on NEE alone. This explanation is weakened, however, by the fact that two[4,6] of the three earlier mentioned studies that reported a considerable reduction of the number of excisions were performed in a similar patient population as our study: patients who had been referred by general practitioners (in the third study the patient population was not described). In addition, the median Breslow thickness of melanomas in our study and two of the three PLC-setting studies were comparable (Breslow thickness was not reported in one study). An alternative explanation would be that general dermatologists have a higher threshold for performing biopsies of lesions, possibly because they are used to managing patients with a relatively low a priori melanoma risk, compared with the patient population seen by expert dermoscopists in PLCs. In addition, the management decisions made by general dermatologists, with less expertise in dermoscopy, are likely to be less dependent on dermoscopy. As a relatively

large proportion of the lesions that were included in our study were melanomas (7%), our results suggest that general dermatologists perform dermoscopy only if they have a (relatively) high level of suspicion of melanoma. As a final note, the smaller reduction of the number of biopsies could not be explained by cosmetic interventions, as these were excluded from the analyses.

In conclusion, we found a comparable impact of dermoscopy on sensitivity and specificity in everyday clinical practice of general dermatologists as was recently reported in a meta-analysis based on studies that were mostly performed in expert PLC settings.[3] From a management perspective, however, dermoscopy did not improve the detection of melanomas. The reduction of the number of excisions was considerably less than has been reported in dermoscopy-expert PLC settings. In our study in the case of one melanoma dermoscopy resulted in false reassurance, changing the clinical diagnosis from melanoma to naevus. It is of great importance that more studies are performed to evaluate the risk of FN due to dermoscopy in nonexpert settings. Unfortunately we had no histological verification of lesions that were initially regarded as melanoma, but were not excised because of their dermoscopic characteristics. This limited our ability to detect possible negative effects of dermoscopy on the detection of melanomas. More studies focusing not only on clinical diagnoses, but also on the management decisions, with a larger number of melanomas are needed, further to determine the benefits and safety of dermoscopy in nonexpert settings.

Acknowledgments

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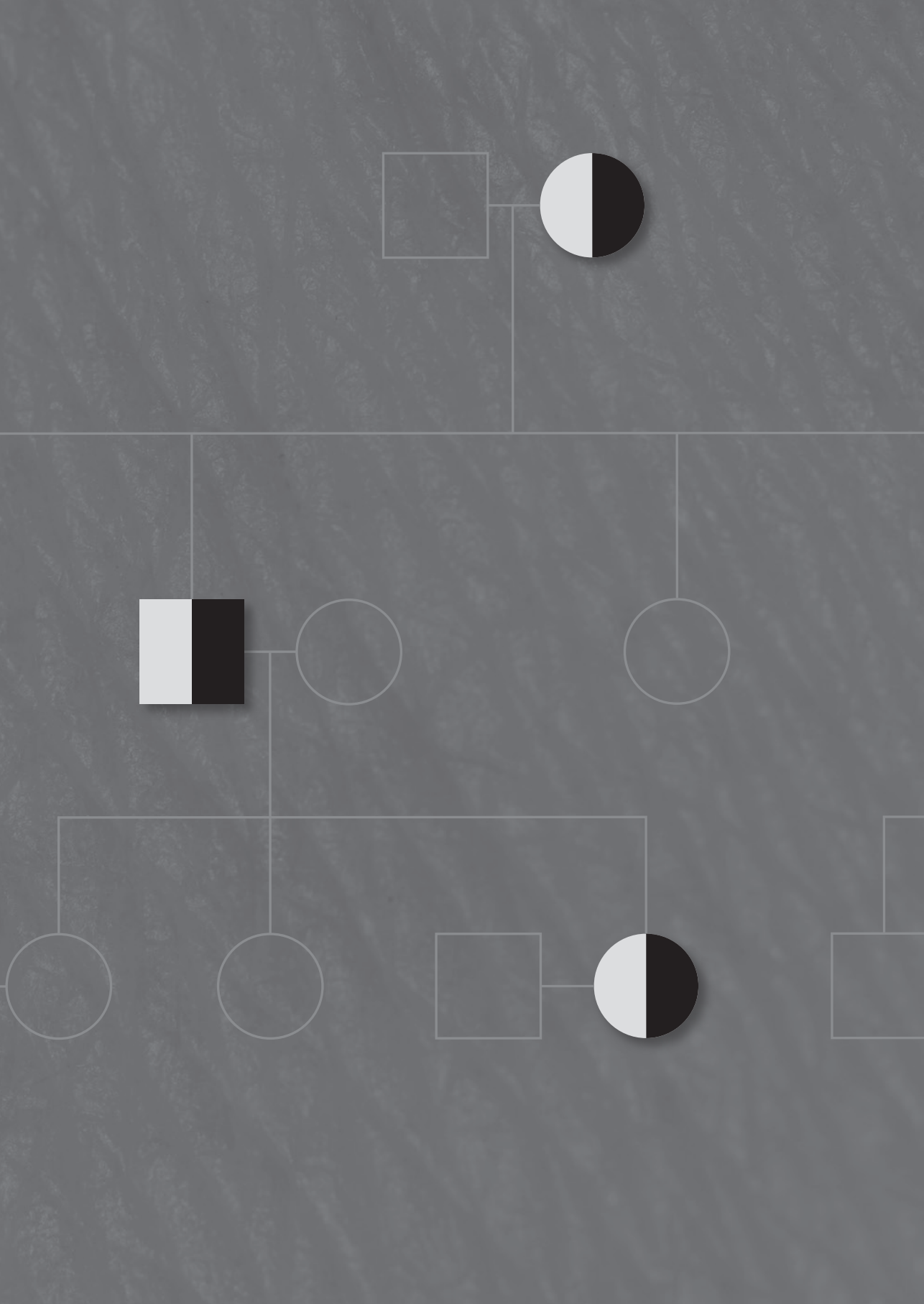
References

1. Bafounta ML, Beauchet A, Aegerter P et al. Is dermoscopy (epiluminescence microscopy) useful for the diagnosis of melanoma? Results of a meta-analysis using techniques adapted to the evaluation of diagnostic tests Arch Dermatol 2001; 137:1343–50.
2. Kittler H, Pehamberger H, Wolff K et al. Diagnostic accuracy of dermoscopy. Lancet Oncol 2002; 3:159–65.
3. Vestergaard ME, Macaskill P, Holt PE, Menzies SW. Dermoscopy compared with naked eye examination for the diagnosis of primary melanoma: a meta-analysis of studies performed in a clinical setting. Br J Dermatol 2008; 159:669–76.
4. Carli P, de Giorgi V, Chiarugi A et al. Addition of dermoscopy to conventional naked-eye examination in melanoma screening: a randomized study. J Am Acad Dermatol 2004; 50:683–9.
5. Argenziano G, Soyer HP, Chimenti S et al. Impact of dermoscopy on the clinical management of pigmented skin lesions. Clin Dermatol 2002; 20:200–2.
6. Carli P, Mannone F, de Giorgi V et al. The problem of false-positive diagnosis in melanoma screening: the impact of dermoscopy. Melanoma Res 2003; 13:179–82.
7. Argenziano G, Puig S, Zalaudek I et al. Dermoscopy improves accuracy of primary care physicians to triage lesions suggestive of skin cancer. J Clin Oncol 2006; 24:1877–82.
8. Binder M, Schwarz M, Winkler A et al. Epiluminescence microscopy. A useful tool for the diagnosis of pigmented skin lesions for formally trained dermatologists. Arch Dermatol 1995; 131:286–91.
9. Westerhoff K, McCarthy WH, Menzies SW. Increase in the sensitivity for melanoma diagnosis by primary care physicians using skin surface microscopy. Br J Dermatol 2000; 143:1016–20.
10. Lorentzen H, Weismann K, Petersen CS et al. Clinical and dermatoscopic diagnosis of malignant melanoma. Assessed by expert and non-expert groups. Acta Derm Venereol (Stockh) 1999; 79:301–4.
11. Carli P, de Giorgi V, Naldi L et al. Reliability and inter-observer agreement of dermoscopic diagnosis of melanoma and melanocytic naevi. Dermoscopy Panel. Eur J Cancer Prev 1998; 7:397–402.
12. Binder M, Puespoeck-Schwarz M, Steiner A et al. Epiluminescence microscopy of small pigmented skin lesions: short-term formal training improves the diagnostic performance of dermatologists. J Am Acad Dermatol 1997; 36:197–202.
13. Cristofolini M, Zumiani G, Bauer P et al. Dermatoscopy: usefulness in the differential diagnosis of cutaneous pigmentary lesions. Melanoma Res 1994; 4:391–4.
14. Stanganelli I, Serafini M, Bucch L. A cancer-registry-assisted evalua•tion of the accuracy of digital epiluminescence microscopy associated with clinical examination of pigmented skin lesions. Dermatology 2000; 200:11–16.
15. Benelli C, Roscetti E, Pozzo VD et al. The dermoscopic versus the clinical diagnosis of melanoma. Eur J Dermatol 1999; 9:470–6.
16. Bono A, Bartoli C, Cascinelli N et al. Melanoma detection. A prospective study comparing diagnosis with the naked eye, dermatoscopy and telespectrophotometry. Dermatology 2002; 205:362–6.
17. Bono A, Tolomio E, Trincone S et al. Micro-melanoma detection: a clinical study on 206 consecutive cases of pigmented skin lesions with a diameter < or = 3 mm. Br J Dermatol 2006; 155:570–3.
18. Carli P, de Giorgi V, Giannotti B. Dermoscopy and early diagnosis of melanoma: the light and the dark. Arch Dermatol 2001; 137:1641–4.
19. Soyer HP, Argenziano G, Talamini R et al. Is dermoscopy useful for the diagnosis of melanoma? Arch Dermatol 2001; 137:1361–3.
20. Bystryn JC. Epiluminescence microscopy: a reevaluation of its purpose. Arch Dermatol 2001; 137:377–9.
21. de Giorgi V, Crocetti E, Carli P. Cutaneous melanoma. Lancet 2005; 365:2003–5.
22. Stolz W, Riemann A, Cognetta A et al. ABCD rule of dermatoscopy: a new practical method for early recognition of malignant melanoma. Eur J Dermatol 1994; 4:521–7.
23. Pehamberger H, Steiner A, Wolff K. In vivo epiluminescence microscopy of pigmented skin lesions. I. Pattern analysis of pigmented skin lesions. J Am Acad Dermatol 1987; 17:571–83.
24. Argenziano G, Zalaudek I, Corona R et al. Vascular structures in skin tumors: a dermoscopy study. Arch Dermatol 2004; 140:1485–9.
25. Malvehy J, Puig S. Dermoscopic patterns of benign volar melanocytic lesions in patients with atypical mole syndrome. Arch Dermatol 2004; 140:538–44.

26. Miyazaki A, Saida T, Koga H et al. Anatomical and histopathological correlates of the dermoscopic patterns seen in melanocytic nevi on the sole: a retrospective study. J Am Acad Dermatol 2005; 53:230–6.
27. Bowling J, Argenziano G, Azenha A et al. Dermoscopy key points: recommendations from the International Dermoscopy Society. Dermatology 2007; 214:3–5.



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Impact of Dermoscopy on the Management of High-risk Patients From Melanoma Families: A Prospective Study

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Abstract

Few studies have investigated the impact of dermoscopy on the management of relatives from melanoma families. The objective of this study was to assess the impact of dermoscopy on clinical diagnosis and management decisions in high-risk familial melanoma patients. In a prospective study 132 consecutive patients were recruited from the pigmented lesions clinic of a tertiary reference centre for familial melanoma. Dermatologists expert in dermoscopy identified 49 suspicious pigmented lesions and recorded pre- and post-dermoscopy diagnoses and management decisions. Dermoscopy was performed in 37% of the patients. Two melanomas were identified. Dermoscopy did not influence sensitivity (1.0), but resulted in 42% fewer excisions, increasing specificity from 0.53 to 0.74 ($p = 0.031$). Dermoscopy resulted in a large reduction in the number of unnecessary excisions. These results suggest that the main effect of dermoscopy in clinical practice for this high risk population is a significant increase in specificity, rather than sensitivity.

Introduction

Incidence and, to a smaller degree, mortality rates of melanoma have increased dramatically in recent years.[1] Between 6% and 14% of all primary cutaneous melanomas occur in a familial context.[2] The melanoma risk for relatives from families with two or more melanoma patients is greatly increased. In carriers of the melanoma susceptibility gene CDKN2A, which is found in approximately 20–40% of melanoma families, the lifetime melanoma risk can be as high as 70%.[3] Surveillance of these relatives is a challenging task. Given the mostly disappointing results of treatments for metastasized melanoma, the most effective way to prevent morbidity and improve survival is the early detection and excision of tumours. Thus, additional tools that can detect early signs of melanoma are valuable.

Dermoscopy is a non-invasive technique that enables the visualization of morphological structures of the skin, from the epidermis down to the superficial papillary dermis, which are not accessible to the naked eye. Several studies have shown that dermoscopy is better than naked eye examination (NEE) at discriminating melanoma from benign pigmented lesions.[4–6] However, few studies have investigated the effect of dermoscopy in everyday clinical practice by studying how it guides management decisions.[7–10] The beneficial effect of dermoscopy ultimately depends on how it improves the ability to determine whether lesions need to undergo biopsy.[8, 11–14] Dermoscopy improves sensitivity if melanomas that would not have been excised based on NEE are excised because of their dermoscopic evaluation. Specificity improves if dermoscopy results in a decrease in the number of excisions of benign lesions. A management-based evaluation might give a different picture of dermoscopy than a diagnosis-based evaluation, because management is based on the differential diagnosis rather than the preferential diagnosis (which has been the central issue in the majority of dermoscopy studies).

The aim of this study was to investigate the impact of dermoscopy on the clinical diagnosis and management of pigmented lesions of relatives from melanoma families, who had a very high personal risk of melanoma, who visit the pigmented lesion clinic (PLC) of a tertiary reference centre for familial melanoma.

Materials and Methods

Between December 2005 and June 2007 patients from melanoma families who had a high personal melanoma risk were recruited from the PLC of a tertiary reference centre for familial melanoma (Department of Dermatology, Leiden University Medical Center) during their regular screening visits. Patients could be included if they fulfilled the following two

criteria: (i) they were a member of a melanoma family (defined as a family containing at least two first- or three second-degree relatives with melanoma); and (ii) they either had a personal history of melanoma, or were a CDKN2A mutation-carrier.

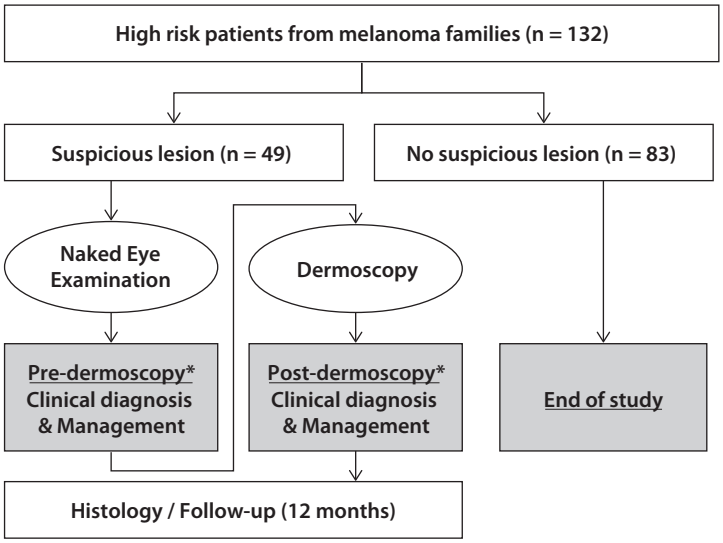
Dermatologists with extensive experience in dermoscopy (WB and NK) took a medical history and clinical examination, guided by the ABCDE criteria and the ugly duckling sign.[15] Pigmented lesions that were regarded as clinically suspicious for melanoma, and for which dermoscopy would also have been used in normal daily practice, were analysed dermoscopically. If no suspicious lesion was found the patients' next screening was scheduled, usually within one year. If more than one suspicious lesion was present, only the most suspicious was included. Prior to dermoscopic evaluation, the diagnosis and management decision based on NEE were recorded. Subsequently dermoscopy was performed and the diagnosis and management decision based on the combined NEE and dermoscopy evaluation were recorded (Fig. 1). Patients were judged in consensus by the two dermatologists (WB and NK). Dermoscopy was performed with a handheld dermatoscope on the basis of (classical) pattern analysis[16], combined with more recently described (vascular) patterns and structures.[17–19] The decision to excise a suspicious lesion was based on the combined NEE and dermoscopic evaluation in accordance with routine clinical practice. Patients with suspicious lesions that were not excised were followed for 12 months in order to detect melanomas that were missed at the examination at the time of inclusion in the study.

Data analysis

The proportion of high-risk patients in whom dermoscopy was performed because of a suspicious pigmented lesion was calculated. Pre- and post-dermoscopy preferential diagnoses were categorized as "melanoma" or "non-melanoma". Management strategies were also grouped into two categories: (i) "intervention": a diagnostic biopsy with the primary intention of histological verification and treatment of a possible melanoma; and (ii) "no intervention": follow-up according to the regular surveillance programme. For biopsied lesions histological diagnosis was used as the reference diagnosis. In an attempt to exclude that melanomas were missed in the case of lesions that were left un-excised, follow-up data was collected one year after inclusion in the study. If the patient had not developed a melanoma at that time, the initial suspicious lesion was regarded as being "non-melanoma".

True positives (TP) were defined as lesions that were classified as melanoma, and confirmed as melanoma on histological examination. True negatives (TN) were defined as lesions that were classified as "non-melanoma", with a subsequent diagnosis other than melanoma on histological examination or no melanoma after one year of follow-up. False positives (FP) were defined as lesions that were classified as melanoma, but not diagnosed as melanoma on histology. False negatives (FN) were defined as lesions that were classified

Figure 1 Study design



*Pre- and post dermoscopy clinical diagnosis and management decisions were compared.

as "non-melanoma", but were diagnosed as melanoma on histology (on inclusion or after one year of follow-up). Sensitivity was computed as TP/(TP+FN) and specificity as TN/(TN+FP). Sensitivity and specificity were also calculated from a management perspective, with the clinical diagnosis "non-melanoma" being exchanged for the management strategy "no intervention", and the clinical diagnosis "melanoma" for the management strategy "intervention". To compare sensitivity and specificity before and after dermoscopy a statistical analysis was performed, using the McNemar test (because pre- and post-dermoscopy data were not independent). Analyses were performed with SPSS 14.0, statistical significance was determined at $\alpha = 0.05$, and two-sided.

The impact of dermoscopy on management was analysed according to the two management categories, as defined above ("intervention" and "no intervention"), in two ways. The impact of dermoscopy on the detection of melanomas was calculated as the proportion of histologically confirmed melanomas that would not have been excised (management category: "intervention") without the use of dermoscopy. In addition, we calculated the proportional reduction in the number of "interventions" due to dermoscopy.

Results

Data characteristics

In total, 132 high-risk patients from melanoma families were included, consisting of: one p14ARF mutation carrier with a personal history of melanoma, four patients with a son or daughter with melanoma (obligatory gene carriers), 13 proven CDKN2A mutation carriers with a personal history of melanoma, 27 proven CDKN2A mutation carriers without a personal history of melanoma, and 87 patients with a personal history of melanoma (20 of whom had multiple primary melanomas).

Dermoscopy was performed in 37% of the patients (49/132). Data on clinical diagnosis and management was complete for all lesions. Excision with histological examination was performed in 14 cases. Two melanomas were diagnosed; one superficial spreading melanoma (SSM, Breslow-thickness 0.86 mm) and one lentigo maligna (melanoma in situ). The 35 patients with suspicious lesions that were not biopsied were followed for 12 months. During follow-up one patient was diagnosed with a melanoma in situ 11 months after inclusion in the study. This lesion had developed in a naevus that had been changing over a period of 6 months according to the patient. Management had not been changed due to dermoscopy in this patient at the time of inclusion in the study.

Clinical diagnosis (Fig. 2A)

Dermoscopy did not lead to diagnosis conversions from melanoma to non-melanoma or non-melanoma to melanoma. Before and after dermoscopy the same seven lesions were considered to be melanomas. Two of these (29%) were confirmed by histology to be melanoma and the other five lesions were dysplastic naevi (n = 4) and a common naevus. Sensitivity was 1.0 (2/2) and specificity 0.89 (42/47), both for NEE alone and for NEE combined with dermoscopy.

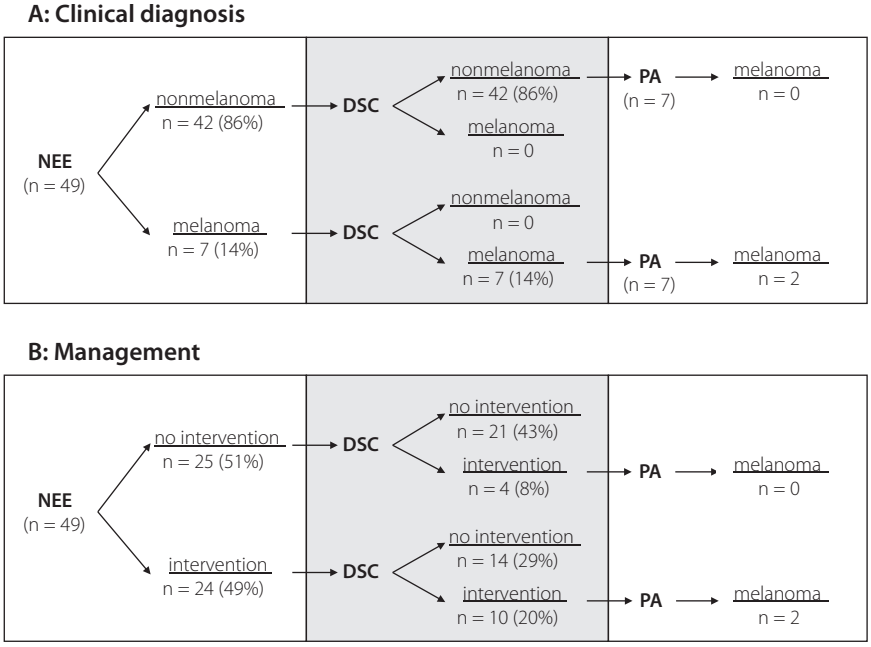
Management (Fig. 2B)

After dermoscopy the management decisions changed in 37% of lesions (n = 18). In 14 cases (29%) an excision was abandoned and in four cases (8%) an excision was decided on. These four lesions were histologically diagnosed as two dysplastic naevi and two common naevi.

Before dermoscopy an excision was intended for 49% of lesions (n = 24), compared with 29% (n = 14) after dermoscopy, resulting in a reduction in the total number of excisions by 42%. The malignant/benign ratio of excised lesions decreased from 1:12 (2/24) to 1:7 lesions (2/14).

Dermoscopy had no impact on the management of the two proven melanomas, as these were already intended to be excised before dermoscopy was performed. Calculations based on management decisions therefore did not show an increase in sensitivity. Specificity, however, increased significantly ($p = 0.031$) from 0.53 (25/47) to 0.74 (35/47).

Figure 2 The effect of dermoscopy on (**A**) the clinical diagnosis and (**B**) management decisions



NEE, naked eye examination; DSC, dermoscopy; PA, pathology

Discussion

In a prospective study we investigated the impact of dermoscopy on the management of patients with a high *a priori* melanoma risk. For this purpose 132 relatives from melanoma families, who had a high personal melanoma risk, were included in a consecutive order. We recorded the proportion of patients in whom dermoscopy was performed and the impact of dermoscopy on clinical diagnoses and management decisions by comparing the evaluation of lesions by NEE with NEE followed by dermoscopy. Patients with suspicious lesions that were not biopsied were followed for one year after inclusion in order to detect false negatives.

In accordance with Carli et al.[7] (49%) we found that, in a large proportion of patients (63%), dermoscopy was not performed. Familial melanoma patients are known to have increased numbers of (dysplastic) naevi, but the phenotype is very variable. Some of our patients

had hardly any naevi and many had only a few. Moreover, patients were under long-term surveillance, and many (suspicious) lesions had already been removed in the past.

Dermoscopy reduced the number of excisions considerably (42%), which is in agreement with other studies.[7–9] In a randomized study Carli et al.[7] found that 38% less excisions were performed in the dermoscopy study-arm compared with the NEE arm. Two prospective studies that investigated the influence of dermoscopy on the management of lesions pre-selected for excision by NEE, found a reduction in the number of excisions of 40% and 70%. [8, 9]

Dermoscopy had no impact on the clinical diagnosis or management of the two histologically proven melanomas and, as a consequence, did not improve sensitivity. Although specificity was not improved by dermoscopy from a clinical diagnosis perspective, it was significantly improved from a management perspective (0.53 before, 0.74 after dermoscopy), without a decrease in sensitivity, as no melanomas were missed due to the reduction in the number of excisions. This can be explained by the fact that, in accordance with other studies[20, 21], a considerable proportion of the lesions that were clinically judged to be benign (preferential diagnosis), were nevertheless regarded as suspicious enough to be excised (based on their differential diagnosis). For such lesions dermoscopy did not affect the clinical diagnosis, but had great influence on the selected management strategy; hence the improvement in specificity.

In a meta-analysis of studies comparing dermoscopy and NEE of suspicious pigmented lesions in a clinical setting, Vestergaard et al.[6] found that dermoscopy improved sensitivity significantly, but had no significant effect on specificity. Our results suggest that, from a management perspective, dermoscopy rather does the opposite: improving specificity rather than sensitivity. Of course, our study was limited by the fact that only two melanomas were diagnosed, but we recently reported similar trends in a larger study in the setting of general dermatologists working in general dermatology clinics.[22]

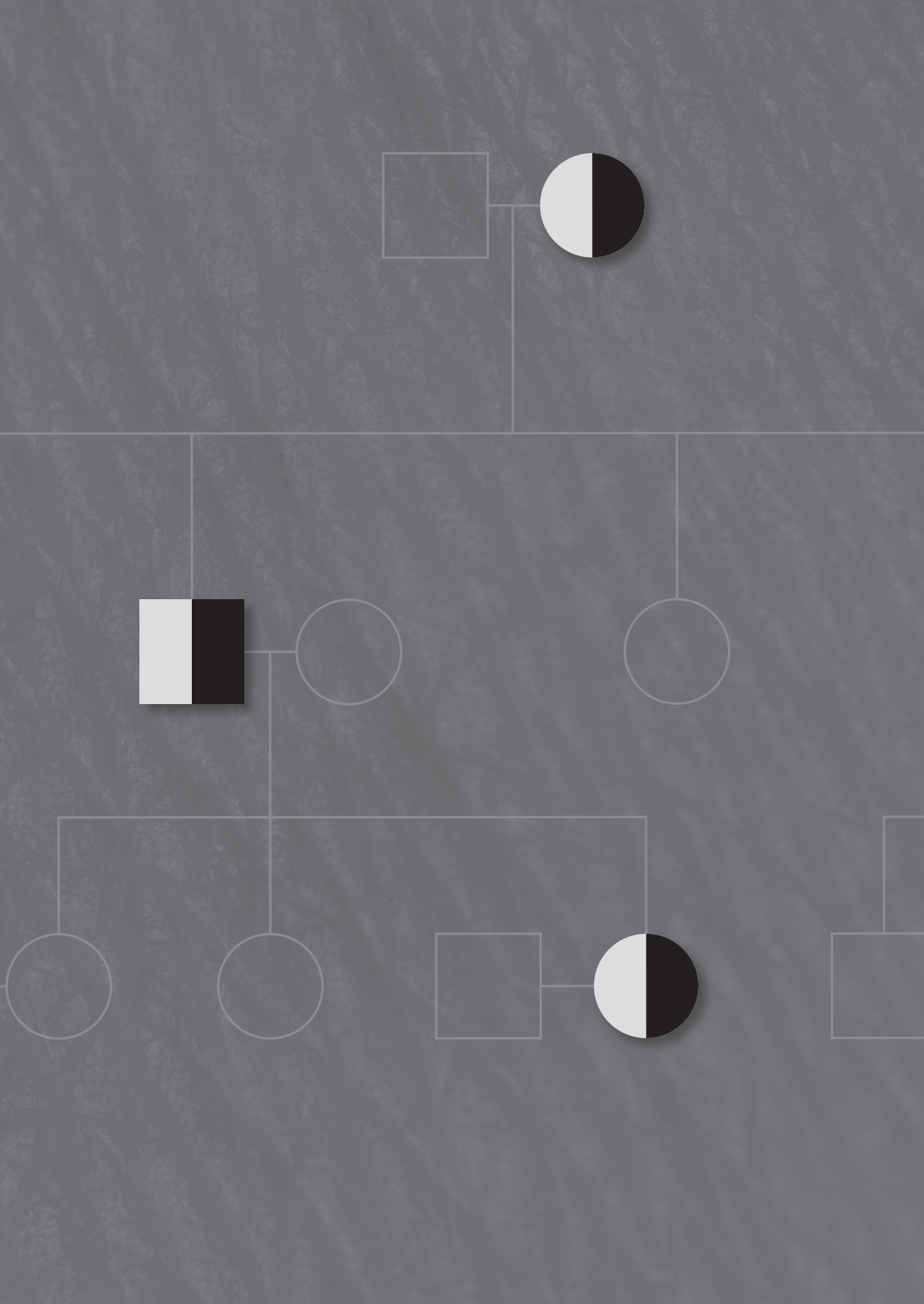
Dermatologists (specialized in) judging pigmented lesions seem to have developed considerable skills in making a final decision from patient history, clinical picture, differential-, comparative-, pattern-recognition and "gut"-feeling[23], which may limit the extent to which dermoscopy contributes to identification of lesions suspicious for melanoma by this group of specialists. However, the use of dermoscopy over the past 20 years may have sharpened the NEE of pigmented lesions, and taught dermatologists to look at a pigmented lesion in a more detailed fashion.

In conclusion, dermoscopy was not performed in the majority of patients from a regularly screened, high melanoma risk patient population. Dermoscopy reduced the number of excisions considerably, and (from a management perspective) increased specificity significantly, without compromising sensitivity. However, dermoscopy did not improve

the detection of melanomas. Studies based on clinical diagnosis may overestimate the impact of dermoscopy on the ability to detect melanomas, while underestimating its ability to reduce the number of unnecessary excisions. Future studies with higher numbers of patients are needed to determine the impact of dermoscopy in daily practice, by investigating the impact of dermoscopy on management decisions.

References

1. Lens MB, Dawes M. Global perspectives of contemporary epidemiological trends of cutaneous malignant melanoma. Br J Dermatol 2004; 150: 179–185.
2. Ang CG, Kelly JW, Fritschi L, Dowling JP. Characteristics of familial and non-familial melanoma in Australia. Melanoma Res 1998; 8: 459–464.
3. Bishop DT, Demenais F, Goldstein AM, Bergman W, Bishop JN, Bressac-de Paillerets B, et al. Geographical variation in the penetrance of CDKN2A mutations for melanoma. J Natl Cancer Inst 2002; 94: 894–903.
4. Bafounta ML, Beauchet A, Aegerter P, Saiag P. Is dermoscopy (epiluminescence microscopy) useful for the diagnosis of melanoma? Results of a meta-analysis using techniques adapted to the evaluation of diagnostic tests. Arch Dermatol 2001; 137: 1343-1350
5. Kittler H, Pehamberger H, Wolff K, Binder M. Diagnostic accuracy of dermoscopy. Lancet Oncol 2002; 3: 159–165.
6. Vestergaard ME, Macaskill P, Holt PE, Menzies SW. Dermoscopy compared with naked eye examination for the diagnosis of primary melanoma: a meta-analysis of studies performed in a clinical setting. Br J Dermatol 2008; 159: 331–336.
7. Carli P, de G, V, Chiarugi A, Nardini P, Weinstock MA, Crocetti E, et al. Addition of dermoscopy to conventional naked-eye examination in melanoma screening: a randomi•zed study. J Am Acad Dermatol 2004; 50: 683–689.
8. Argenziano G, Soyer HP, Chimenti S, Argenziano G, Ruocco V. Impact of dermoscopy on the clinical management of pigmented skin lesions. Clin Dermatol 2002; 20: 200–202.
9. Carli P, Mannone F, de G, V, Nardini P, Chiarugi A, Giannotti B. The problem of false-positive diagnosis in melanoma screening: the impact of dermoscopy. Melanoma Res 2003; 13: 179–182.
10. Argenziano G, Puig S, Zalaudek I, Sera F, Corona R, Alsina M, et al. Dermoscopy improves accuracy of primary care physicians to triage lesions suggestive of skin cancer. J Clin Oncol 2006; 24: 1877–1882.
11. Carli P, de G, V, Giannotti B. Dermoscopy and early diagnosis of melanoma: the light and the dark. Arch Dermatol 2001; 137: 1641–1644.
12. Soyer HP, Argenziano G, Talamini R, Chimenti S. Is dermoscopy useful for the diagnosis of melanoma? Arch Dermatol 2001; 137: 1361–1363.
13. Bystryn JC. Epiluminescence microscopy: a reevaluation of its purpose. Arch Dermatol 2001; 137: 377–379.
14. de G, V, Crocetti E, Carli P. Cutaneous melanoma. Lancet 2005; 365: 2003–2005.
15. Bowling J, Argenziano G, Azenha A, Bandic J, Bergman R, Blum A, et al. Dermoscopy key points: recommendations from the international dermoscopy society. Dermatology 2007; 214: 3–5.
16. Pehamberger H, Steiner A, Wolff K. In vivo epiluminescence microscopy of pigmented skin lesions. I. Pattern analysis of pigmented skin lesions. J Am Acad Dermatol 1987; 17: 571–583.
17. Argenziano G, Zalaudek I, Corona R, Sera F, Cicale L, Petrillo G, et al. Vascular structures in skin tumors: a dermoscopy study. Arch Dermatol 2004; 140: 1485–1489.
18. Malvehy J, Puig S. Dermoscopic patterns of benign volar melanocytic lesions in patients with atypical mole syndrome. Arch Dermatol 2004; 140: 538–544.
19. Miyazaki A, Saida T, Koga H, Oguchi S, Suzuki T, Tsuchida T. Anatomical and histopathological correlates of the dermoscopic patterns seen in melanocytic nevi on the sole: a retrospective study. J Am Acad Dermatol 2005; 53: 230–236.
20. Heal CF, Raasch BA, Buettner PG, Weedon D. Accuracy of clinical diagnosis of skin lesions. Br J Dermatol 2008; 159: 661–668.
21. Stanganelli I, Serafini M, Bucch L. A cancer-registry-assisted evaluation of the accuracy of digital epiluminescence microscopy associated with clinical examination of pigmented skin lesions. Dermatology 2000; 200: 11–16.
22. van der Rhee JI, Bergman W, Kukutsch NA. The impact of dermoscopy on the management of pigmented lesions in everyday clinical practice of general dermatologists: a prospective study. Br J Dermatol 2010; 162: 563–567.
23. Marghoob AA, Scope A. The complexity of diagnosing melanoma. J Invest Dermatol 2009; 129: 11–13.



9

Summary and Discussion

Nederlandse Samenvatting

List of publications

List of abbreviations

Curriculum Vitae

Dankwoord – Acknowledgements

Summary and Discussion

Introduction

Familial susceptibility is the strongest risk factor (in terms of effect size) for melanoma, and is associated with a very high life-time melanoma risk, especially in the context of the high-penetrance melanoma susceptibility genes CDKN2A, and CDK4. Early melanomas can be cured by an excision with proper resection margins, but prognosis is grim for advanced disease. For these reasons, surveillance of melanoma families has been widely advocated. The studies presented in this thesis focused on 3 related issues: 1. the clinical and histological characteristics of melanoma (patients) from melanoma families with a pathogenic germline mutation in CDKN2A; 2. the effectiveness and yield of surveillance of melanoma families with different CDKN2A mutation status and family characteristics, and 3. the impact of dermoscopy on the management of suspicious lesions in the context of melanoma surveillance. In this concluding chapter, our results will be discussed jointly and in the context of (recent) medical literature.

Part I: Clinical and histological characteristics of melanoma in CDKN2A mutated families.

Several studies have demonstrated that melanoma patients from melanoma families have a different clinical presentation than melanoma patients from the general population. In **chapter 2** we verified and substantiated these differences by comparing the clinical and histological characteristics of 182 patients with 429 CMM from families with a germline mutation in CDKN2A, to a large control population from the Leiden population based cancer registry (7512 patients with 7842 CMM).

We confirmed that melanoma patients from CDKN2A mutated families have a young age of onset and a very high risk of multiple primary melanomas (MPM). We found a mean age of diagnosis of 39.0 years, which is 15 years younger than the general population. Concerning MPM we reported for the first time a difference in concordance with regard to the body site of the first and second melanoma. Where a concordance was seen in MPM patients from the general population, concordance was absent in MPM patients from CDKN2A mutated families. These findings are all in accordance with the high melanoma susceptibility in CDKN2A mutated families, and emphasize the importance of regular skin examinations, starting at a young age and involving the whole skin.

In accordance with earlier studies we found that a relatively high proportion of superficial spreading (≈ 90%) and relatively few lentigo maligna type melanomas (2%) and nodular melanomas (8%) are diagnosed in these families. Contrary to other studies that reported an absence of acrolentiginous melanomas (ALM) in melanoma families, the proportion of ALM in our dataset was similar to the general population.[1,2]

We found that relatively few melanomas from CDKN2A mutated families were located in the head and neck region, but the difference was not statistically significant in a multivariate analysis. However, melanomas that were located in the head and neck region, were diagnosed at a relatively young age compared to melanomas localized elsewhere on the body, whereas, in the general population, melanomas in the head and neck region were diagnosed at a relatively older age compared to melanomas elsewhere on the body. This difference persisted if lentigo maligna melanomas were excluded from the analysis.

Both epidemiological and genetic data suggest that melanoma is a (clinical, histological, epidemiological and molecular) heterogeneous disease, and different pathways to melanoma genesis have been proposed.[3] As described in **chapter 2** our results with regards to the clinical and histological characteristics of melanoma (patients) from families with a germline mutation in CDKN2A suggest that their distribution along these different pathways differs from the general population. From the perspective of the divergent pathway model by Whiteman at all, CDKN2A mutated families tend to follow the nevus pathway, which is associated with a phenotype of multiple nevi and intermittent sun exposure.[4] This supports the notion that prevention of high intermittent sun exposure and sunburn is of great significance within this population. Future studies have to clarify to what extent the natural development and prognosis of melanomas in families with CDKN2A germline mutations differ from sporadic melanomas, as well as the potential implications for preventive and treatment strategies.

Part II: Management of melanoma families

Effectiveness of surveillance

Only few studies have investigated the effectiveness of surveillance of melanoma families.[5-8] We estimated the effect of surveillance on tumour Breslow thickness in: 1. a retrospective data-set of 226 melanomas from relatives of 37 CDKN2A mutated families that were under surveillance at the LUMC pigmented lesion clinic (**chapter 3**), and 2. Fifty-one prospectively detected melanomas in 37 patients among 450 members of 72 melanoma families that were registered at the Netherlands Foundation for the Detection of Hereditary Tumors (NFDHT) and under surveillance throughout the Netherlands (**chapter 4**).

In accordance with previous studies we found that melanomas detected in patients under surveillance (surveillance melanomas) had a significantly thinner Breslow thickness than index melanomas (i.e. the first melanoma of the first two family members diagnosed with melanomas). In both studies the median thickness of surveillance melanomas was 0.50 mm, which is comparable to earlier studies.[5,7,8] On average, index melanomas were approximately 1.5 times as thick as surveillance melanomas. The proportion of surveillance

melanomas with a Breslow thickness > 1.0 mm, was 11% and 22% in **chapter 3 and 4** respectively. In the LUMC PLC setting surveillance melanomas were estimated to be 3 times less likely to have a Breslow thickness > 1.0 mm compared to index melanomas. In **chapter 4** no statistically significant difference was found with respect to the proportion of melanomas with a Breslow thickness > 1.0 mm in the surveillance and index group, but significance may not have been reached due to lack of power (estimate of the odds ratio was 2.5, which is comparable to chapter 3). We did report in **chapter 4** that none of the 37 patients, who were diagnosed with their first melanoma during surveillance, had died after a median follow-up of 4.2 years since diagnosis.

Our findings support the notion from previous studies that surveillance results in earlier detection of melanomas and as such, is likely to improve survival of patients from melanoma families. Acknowledging the limitations of a direct comparison of the two datasets, our studies suggest that surveillance in the setting of a tertiary familial melanoma PLC clinic (chapter 4) was more effective in reducing the proportion of thick melanomas (11% of surveillance melanomas with Breslow thickness > 1.0 mm) than the setting of general dermatology departments (22% of surveillance melanomas with Breslow thickness > 1.0 mm, chapter 5). Effectiveness of surveillance of the highest risk populations may benefit from centralization to highly specialized clinics.

Surveillance interval

There is very little data on the optimal length of the interval between two skin examinations for melanoma. In **Chapter 3** we report that the majority of surveillance melanomas were detected within 6 months since the previous surveillance skin examination. The current 2012 Dutch melanoma guideline recommends patients should be seen at least annually.[9] In order to determine the appropriate surveillance interval it is important to know the natural history of melanoma. Several studies have attempted to approximate the melanoma growth rate based on patients recall of the moment the lesion was first noticed or started changing. Even though this approach has considerable theoretical as well as practical limitations, it was demonstrated that (this measurement of) growth rate is prognostic of survival, independent of Breslow thickness.[10-12] Consequently, three types of melanomas have been defined: slow growth melanoma (< 0.1 mm/month), intermediate growth (0.1 – 0.49 mm / month) and fast-growing melanoma (≥ 0.5 mm/month). It was reported that all three types represent approximately one-third of CMM.[13] This is in line with the fact that most melanomas in **chapter 3** were detected within 6 months since the previous skin examination, and suggest that, if one is to advance the detection of those melanomas that are most likely to have an unfavourable outcome, surveillance may need to be performed at least biannually.

However, we found no correlation between tumour thickness and the length of the surveillance interval for melanomas detected within 24 months since the previous skin examination. A possible explanation for this contra-intuitive finding may be that it is more

likely that patients will present intermediate and fast growing melanomas to their physician in between their regular surveillance appointments, as these melanomas are more noticeable than slow growing melanomas. In our study (**chapter 3**) 20% of surveillance melanomas were so-called interval tumors, i.e. detected in between regular surveillance visits. The majority of these were identified by patients themselves, in an early stage. Patients that participate in our surveillance program, are instructed to perform monthly skin self-examinations (SSE). The performance of SSE has been demonstrated to be associated with detection of melanomas in an early stage, and improved melanoma survival, and is therefore a valuable addition to surveillance by physicians.[14]

Compliance

In **chapter 3** we studied adherence to surveillance recommendations at the time of melanoma diagnosis. Noncompliance was defined as a diagnosis of melanoma later than 2 months after the recommended surveillance date. We found that 20% of surveillance melanomas were detected in noncompliant patients. The median surveillance interval at the time of diagnosis was 24 months. When we limited the analysis to individuals who were diagnosed with their first melanoma the proportion of noncompliant patients was 46%. These results are in accordance with two earlier studies, that reported that adherence to surveillance recommendations was approximately 50% among members of CDKN2A mutated families, and higher for individuals with a history of melanoma compared to individuals without a history of melanoma.[15,16]

We found that melanomas of patients that were noncompliant, were 5-times more likely to have a tumor thickness > 0.75 mm compared to melanomas of compliant patients. These findings are of great concern because they suggest that current health-education to high-risk patients is ineffective in communicating the importance of regular surveillance, particularly for individuals without a personal history of melanoma. Several interventions to improve compliance have been described; these include: 1. clear recommendations on surveillance, 2. psycho-education, 3. skill-based training (related to SSE), 4. time and space for individuals to explore and express their feelings and concerns about melanoma with a caring and attentive professional, 5. genetic testing, 6. shortening surveillance intervals.[15] A more detailed description of the management of health behavioral change processes is beyond the scope of this chapter.

Yield of surveillance

Members of melanoma families are invited to participate in surveillance based on the assessment of their melanoma risk. Few studies have evaluated the yield of surveillance in relation to patient and family characteristics on which risk assessment is based. In **chapters 4 and 5** we assessed the yield of surveillance in family members with different personal and family characteristics. Additional data on melanoma detection rates came from **chapters 2 and 8**.

History of melanoma

In accordance with an earlier study, we found that family members with a history of melanoma have a considerable higher melanoma detection rate then their first degree relatives (HR 3.9, **chapter 4**). [17] As melanoma patients are probable carriers of the genetic risk factor in their family, it was anticipated that this subpopulation is at a particularly high risk of developing (additional) melanomas. In **chapter 2** we performed a quantitative analysis of the risk to be diagnosed with a second melanoma in (single) melanoma patients from families with the p16-Leiden mutation, and found the 5 and 10 year cumulative melanoma incidence to be 23.4% and 34.8% respectively. This is considerably higher than the risk of second melanoma in the general population. We found that the increased risk was age dependent, with a HR of 15.8 for patients with melanoma diagnosed before the age of 40 years and 7.5 for patients diagnosed above age of 40 years respectively.

Both in **chapter 2 and 4** we computed the occurrence of only the first subsequent melanoma for each patient. As melanoma counts as high as 19 (**chapter 2**) have been reported, the overall melanoma detection rate in melanoma patients is expected to be considerable higher.

CDKN2A mutation status

In **chapter 4** we compared the melanoma detection rate in families with different CDKN2A mutation status. In a multivariate analysis we demonstrated that the melanoma risk in p16-Leiden mutated families was statistically significant higher than in CDKN2A wild-type families, independent of the families' number of melanoma patients and youngest age of melanoma diagnosis, with a HR of 3.6. Given the fact that CDKN2A (and CDK4) is the only known high risk melanoma susceptibility genes, it was expected that families with these mutations are at a particularly high melanoma risk. It should be noted however that CDKN2A/CDK4 wild-type families, are very heterogenic. As discussed in the introduction, melanoma risk in the majority of these families is probably attributable to (a combination of) low (e.g. MC1R) and moderate (e.g. MITF) risk modifier genes, and presumably to some extent environmental, as well as behavioral aspects.[18,19] It cannot be excluded however that rare high penetrance genes play a role in a small proportion of these families. It is likely that such families have similar characteristics as CDKN2A mutated families, i.e. larger numbers of melanoma patients, young age of diagnosis and patients with multiple melanomas. In **chapter 4** we found a borderline non-significant difference in melanoma detection rate between families with 2 melanoma patients and families with 3 or more melanoma patients (HR 2.2, 95% CI: 0.9 – 5.0), which is in accordance with the notion that number of affected relatives is positively correlated with melanoma risk.

Second degree relatives

In the Netherlands, historically all first and second degree relatives from melanoma families have been recommended to undergo regular skin examinations. The subject of

surveillance of second degree relatives has received hardly any attention in the medical literature and foreign melanoma guidelines. In **chapter 5** we performed a study to investigate the yield of surveillance of second degree relatives from P16-Leiden families. We found a melanoma incidence rate of 2.2 / 1000 person years if all available data on second degree relatives was included in the analysis. In a sub-analysis of second degree relatives under surveillance we found a melanoma detection rate of 8.5 / 1000 person years. Differences in these outcome are most likely related to selection bias, i.e. persons at high risk were more likely to participate in surveillance, difference in age distribution and possible under-reporting and under-detection in unscreened individuals. Standardized morbidity ratio for second degree relatives compared to the general population was 12.9.

Risk stratification

As was discussed in the introduction, the surveillance recommendations in the 2005 melanoma guidelines were independent of CDKN2A (and CDK4) mutation status and family characteristics. Based on **Chapter 2, 4, 5 (and 8)** figure 1 proposes a risk stratification diagram for members of melanoma families.

Our studies were restricted to families with the p16-Leiden mutation in CDKN2A. Although melanoma risk may vary to some extent, previous studies suggest that similar estimations probably apply to other pathogenic mutations in CDKN2A, as well as CDK4 mutations.[20-22]

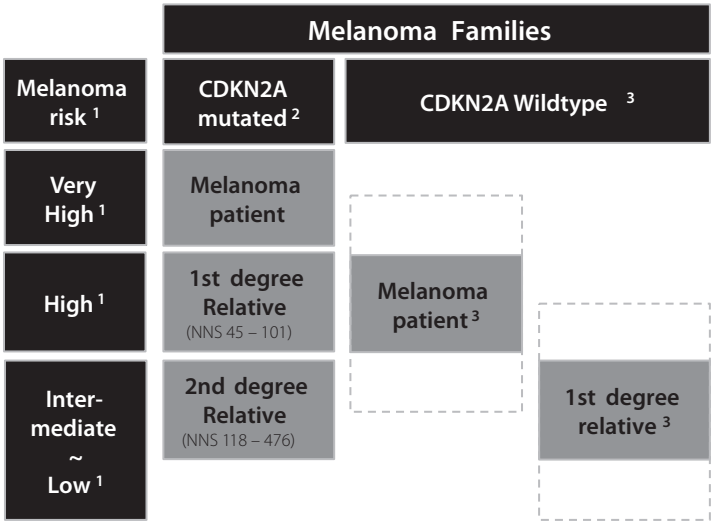
It is important to stress that the proposed risk categorization has its limitations. Families with CDKN2A (and CDK4) wild-type or unknown mutation status are very heterogeneous. The majority of these families will have a considerable lower melanoma risk than CDKN2A mutated families, but in some families yet unknown high risk susceptibility genes may be present. As described above, family characteristics can provide additional information that may justify an upgrade to a higher risk category (this issue will be addressed in more detail below, where its practical implications are discussed).

The NNS in figure 1 are based on **chapter 5** in which melanoma incidence was calculated in two ways. The lower NNS represent actual NNS in the LUMC PLC surveillance program. The higher NNS were based on calculations in which patients that were not under surveillance in this institution, were included as well. We expect actual NNS to be nearer to the lower margin.

Management recommendations

Based on this thesis a number of recommendations can be given concerning the management of melanoma families. Our data from **chapter 3 and 4** is in accordance with previous data that surveillance results in early diagnosis and a reduction in the proportion of melanomas with a Breslow thickness > 1mm. This finding supports the notion that surveillance should be recommended to family members at a significant risk of melanoma.

Figure 1 Proposed risk categories based on melanoma detection rates during surveillance of melanoma patients, first- and second degree relatives from mutated and wildtype CDKN2A families



NNS = Numbers Needed to Screen to detect one melanoma (numbers based on chapter 5)

[1] Risk categories are approximations based on hazard ratio's calculated in chapter 4 and 5: HR (melanoma patients versus first degree relatives) ≈ 4, HR (CDKN2A mutated versus CDKN2A wild-type families) ≈ 3.5, and HR (CDKN2A mutated first degree relatives versus second degree relatives) ≈ 2.5 – 5.0. NNS give an indication of the quantitative risk.

[2] Our studies were only conducted in families with the P16-Leiden mutation, the predominant CDKN2A mutation in the Netherlands.

[3] CDKN2A/CDK4 wild-type families are heterogeneous; dotted lines are used to emphasize the variance in melanoma risk. Based on chapter 4, risk of melanoma (recurrence) in families with ≥ 3 melanoma patients is expected to be higher than risk in families with only 2 affected relatives.

The data from **chapter 4** indicate that families with pathogenic mutations in CDKN2A have a considerable higher melanoma risk than CDKN2A wild-type families. Melanoma patients from these families have a very high risk of multiple melanoma, the majority of which (≈75%) were diagnosed in the first 10 years after diagnosis. Intensified surveillance of these relatives, at least during the first 10 to 15 years after melanoma diagnosis, is therefore recommendable. As we found (**chapter 2**) that the majority of melanomas were detectable within 6 months since the previous skin examination, at least biannual surveillance of this highest risk population should be considered. Given the fact that we did not find a correlation between Breslow thickness and surveillance interval, a switch from annual (2005 melanoma guideline) to biannual surveillance in this risk population, may be introduced in the context of a randomized prospective study. We estimated (**chapter 5**)

that the melanoma risk of second degree relatives from CDKN2A mutated families is considerable higher than the risk of the general population (Standerdized Morbidity Ratio ≈ 13). Based on results from **chapter 4 and 5**, we estimate that melanoma risk of second degree relatives from CDKN2A mutated families is comparable to melanoma risk of first degree relatives from CDKN2A wild-type families. These findings support the continuation of offering surveillance to second degree relatives from these very high melanoma risk families.

Families with an unknown CDKN2A mutation status took an intermediate melanoma risk position, which is likely attributable to a proportion of these families having a mutation in CDKN2A or CDK4. Genetic testing of these families is desirable in order to estimate melanoma risks, and facilitation of adequate surveillance recommendations. Because CDKN2A wild-type families are heterogeneous, melanoma risk estimation in these families should be accompanied by a critical appraisal of family characteristics. **Chapter 4** gives support to the notion that the number of melanoma patients in a family is positively correlated with melanoma risk in these families. With regards to the classification in figure 1, it should be considered to "upgrade" CDKN2A wild-type families with many affected relatives to a higher risk category. Melanoma risk of first degree relatives from families with only two affected relatives, and with a single melanoma, diagnosed at an older age (e.g. > 50 years) is expected to be relatively low, but still higher than the general population. The yield of surveillance of second degree relatives from (the majority of) CDKN2A wild-type families is expected to be very small, and cessation of the offering of surveillance to this population should be considered.

In **chapter 3** we reported that interval melanomas detected by patients were diagnosed after a median surveillance interval of 5 months and had a favorable tumor thickness. These findings supports the growing evidence that SSE is a key factor in effective melanoma surveillance. Instruction to perform monthly SSE should therefore be given to all patients. Formalization of these instructions may enhance patients adherence to SSE recommendations.

We found that noncompliance with the surveillance program was considerable, being close to 50% at the time of melanoma diagnosis for patients without a history of melanoma. Noncompliance was associated with thicker melanomas and is therefore a considerable threat to the effectiveness of surveillance. Compliance should therefore be one of the targets of surveillance and strategies to improve compliance should be developed.

In table 1, age at diagnosis (in percentiles) of the first melanoma in members of melanoma families with different CDKN2A mutation status as were found in **chapter 2, 4 and 5** are presented.

Table 1 shows that approximately 80% of melanomas were diagnosed before age 50 years, and 90% of melanomas were diagnosed before age 60 years. These findings suggest that

Table 1 Age of diagnosis of first melanoma in members of melanoma families (based on **studies 2, 4 and 5**)

Percentiles[1]	**Age of diagnosis (first melanoma)**				
	Chapter 2[2]	**Chapter 5**[2]	**Chapter 4**		
	P16-Leiden (n = 182)	P16-Leiden (n = 56)[3]	P16-Leiden (n = 44)	Wild-type (n = 57)	Unknown (n = 102)
Minimum	11.7	15.6	16.5	16.4	19.7
10	21,7	20,5	21,6	24,3	25,7
20	27,2	23,8	26,0	29,9	29,8
30	31,7	27,6	28,5	35,9	33,7
40	35,5	35,0	32,6	38,6	38,4
50	38,6	37,8	38,1	42,5	43,3
60	41,1	40,0	41,0	46,7	46,5
70	45,1	43,4	45,1	49,1	49,6
80	50,5	51,5	49,2	54,4	54,3
90	57,5	60,4	56,4	61,0	59,7
Maximum	72.3	72.3	69.3	78.2	75.6

[1] Percentiles represent the proportion of melanoma patients that were diagnosed with melanoma at the given age in the right columns; e.g. in Chapter 2, 40% of melanoma patients were diagnosed with their melanoma at age ≤ 35.5 years.

[2] All cases in chapter 5 are included in chapter 2 as well.

[3] These are all incident cases.

upper age limits on (intensified) surveillance, especially in patients with an intermediate risk, may improve the cost-effectiveness of surveillance, without considerable loss of effectiveness.

Over-diagnosis

Over-diagnosis of melanomas has been a recurring point of discussion. It became an issue as a result of the observation that, whereas melanoma incidence has been increasing considerably over the last decades, mortality has increased relatively little. Some have argued that the increase of melanoma incidence is largely attributable to indolent melanocytic tumors, that are diagnosed as a result of increasing attention and screening for melanoma.

In **chapter 6** we report an observation related to over-diagnosis/misclassification in 10 CDKN2A wild-type members of melanoma families with the p16-Leiden mutation. In this group 73% (7 in situ and one invasive) of 11 melanocytic lesions that were initially classified as melanomas, were reclassified as benign lesions after histological revision. This implied that 7 out of 10 of these CDKN2A wild-type family members were unnecessarily burdened with a diagnosis of melanoma.

This observation illustrates that if populations with a low *a priori* melanoma risk participate in surveillance, the positive predictive value of a histological diagnosis of melanoma decreases. In order to limit misclassification of benign lesions as melanoma (false positives), it is important to make an adequate selection of risk populations for surveillance. In addition, this finding challenges the reported increased melanoma incidence among mutation negative relatives, which has caused doubt on the value of genetic testing for mutations in CDKN2A.[23]

Table 2 Recommendations

Recommendations for the development of surveillance strategies for melanoma families *
1. Intensified surveillance of melanoma patients from (CDKN2A mutated) melanoma families, at least up to 10 years after melanoma diagnosis is recommended. *(Chapter 2, 4)*
2. Genetic testing helps identifying those families and individuals at highest risk of melanoma. Melanoma family members should therefore be encouraged to be tested (i.e. melanoma patients from families with unknown CDKN2A mutation status and first degree relatives in case of families with a pathogenic CDKN2A mutation). *(Chapter 2, 4, 5 and 6)*
3. Surveillance should start at an early age (preferably before age 15 years). Discontinuation of surveillance above age 60 years may improve the cost-effectiveness of surveillance, without considerable loss of effectiveness, especially in case of family members without a history of melanoma from intermediate to low risk families. *(Chapter 2,4 and 5)*
4. Given the fact that most melanomas in our studies were detected within 6 months since the previous skin-examination, biannual skin-examinations should be considered, especially for those at highest risk of melanoma (i.e. melanoma patients and proven CDKN2A mutation carriers). *(Chapter 3)*
5. The favourable Breslow thickness of interval melanomas, underlines the importance of health education for melanoma family members concerning the signs of melanoma and skin self-examination. *(Chapter 3)*
6. Noncompliance has a considerable impact on the effectiveness of surveillance. Strategies need to be developed to improve patients' compliance. *(Chapter 3)*
7. The probability of melanoma detection in CDKN2A mutated families is higher than in (most) CDKN2A wildtype families. CDKN2A mutated families therefore require more stringent surveillance strategies. *(chapter 4)*
8. Surveillance of second degree relatives should be considered for CDKN2A mutated families, but the yield of surveillance of second degree relatives from CDKN2A/CDK4 wildtype families appears to be very limited, especially in case of few affected relatives. *(chapter 4 and 5)*
9. The use of dermoscopy in the surveillance of melanoma families reduces the number of unnecessary excisions. Dermoscopy is therefore likely to reduce the burden of surveillance, improve cost-effectiveness and decrease the risk of overdiagnosis. The use of dermoscopy is therefore recommended. *(chapter 6 and 7)*

* Except for recommendation 6, all recommendations in this table are (fully or in part) reflected in the 'new' 2012 Dutch melanoma guideline.

Part III: Impact of dermoscopy on clinical practice

Several meta-analysis have concluded that dermoscopy improves the diagnostic accuracy of the clinical diagnosis of melanoma with the unaided eye.[24-26] We investigated how this is reflected in the impact of dermoscopy on management in two different clinical settings:

1. The specialized pigmented lesion clinic of the LUMC in which two dermoscopy experts identified and evaluated in consensus 49 suspicious lesions including 2 melanomas, in 132 consecutive FAMMM patients with a history of melanoma and/or a pathogenic mutation in CDKN2A (**chapter 8**).
2. The general dermatology clinics of 17 dermatologists, that evaluated a total of 209 suspicious pigmented lesions, including 14 melanomas (**chapter 7**).

The impact of dermoscopy on clinical diagnosis

In neither of the clinical settings the addition of dermoscopy to naked eye evaluation (NEE) resulted in a statistically significant improvement of sensitivity or specificity of the clinical diagnosis of melanoma with the unaided eye. However, we did find an (not statistically significant) increase in sensitivity from 0.79 to 0.86 and in specificity from 0.96 to 0.98 for dermatologists working in general dermatology clinics (**chapter 7**), which is very similar to a meta-analysis of dermoscopy studies performed in a clinical setting, reporting that dermoscopy improves sensitivity rather than specificity.[26]

In the expert dermoscopists / familial melanoma surveillance setting, dermoscopy did not result in an improvement of sensitivity (1.0) or specificity (0.89) (**chapter 8**). We believe that this finding is related to the fact that only 49 suspicious lesions with only 2 melanomas could be included in our study, as suspicious lesions necessitating the use of dermoscopy were detected in only 37% of the 132 patients that were recruited. This could be attributed to the highly variable nevus phenotype of familial melanoma patients (gene carriers can have very few (atypical) nevi), and the fact that patients were under regular surveillance, implicating that (suspicious) lesions may have already been removed in the past.

The impact of dermoscopy on clinical practice

Dermoscopy was associated with a considerable reduction in the number of lesions that were excised, both in the expert dermoscopists / familial melanoma surveillance setting (42% reduction, statistically significant) and general dermatology clinics setting (9%, not statistically significant) **(chapter 7 & 8)**. Our findings on the impact of dermoscopy on management in the expert dermoscopists setting are very similar to the only randomized controlled trial in a (dermoscopy expert) dermatologists setting that has ever been conducted, reporting a 38% reduction in excisions.[27] The reduction of excisions was considerably lower in the general dermatologists setting. Some confirmation of this findings comes from two recent studies. First, a study investigating the impact of

dermoscopy on management of pigmented lesions by general dermatologists based on photographic images, found dermoscopy actually resulted in a slight increase in the intention to excise lesions.[28] An important limitation of this study however was the fact that it only included excised lesions. Second, an international study reported that between 1998 and 2007 the number needed to excise to detect one melanoma almost halved in specialised clinical settings, but remained unchanged in non-specialised clinical settings (including general dermatologists and general practitioners).[29] The authors of this study suggested that the difference is most likely related to differences in the uptake of dermoscopy.[25] Our results suggest that, in addition to differences in the uptake of dermoscopy, dermoscopy may be less effective in reducing the numbers needed to excise in the hands of non-expert dermoscopists and/or in the patient population setting of non-specialised clinics.

In summery chapter 7 and 8 suggest that the main effect of dermoscopy in the dermoscopy expert / familial melanoma surveillance setting, was a significant improvement of specificity (from a management, but not from a clinical diagnosis standpoint), and a decreased burden of unnecessary excision. Dermoscopy is therefore likely to improve the cost effectiveness of familial melanoma surveillance, and in addition may decrease the risk of 'overdiagnosis'. The impact of dermoscopy in the general dermatology setting was considerably smaller, and no statistically significant improvement of sensitivity or specificity were noticed. Possible explanations for the differences between the two clinical settings may be related to the degree of dermoscopy expertise as well as differences in the population under study. Differences in lesion ascertainment between the two clinical settings may have resulted in an underestimation of the reduction of the number of excisions in the general dermatology setting.

An important issue that has been suggested before, but has now been clearly demonstrated in our studies, is the fact that prior studies that evaluated dermoscopy from a clinical diagnosis perspective, give an inaccurate picture of its impact in clinical practice. The just mentioned meta-analysis by Vestergaard et al. concluded that dermoscopy rather improves sensitivity than specificity and the promotion of dermoscopy has benefitted considerably from the proclamation that it advances melanoma diagnosis.[26] Our studies confirm an earlier randomized controlled trial, that dermoscopy primarily results in the reduction of excisions. Our studies were considerably limited in their ability to detect an improvement of sensitivity due to the small number of melanomas. Still, it appears logical that dermoscopy predominantly reduces the proportion of excisions (i.e. improve specificity rather/more than sensitivity), as the majority of clinicians use dermoscopy predominantly for the evaluation of lesions that have been judged to be suspicious by naked eye examination. Dermoscopy can be applied in several ways however (table 3); first of all dermoscopic evaluation can be performed only once or sequentially in case of

dermoscopic follow-up; secondly lesion selection for dermoscopic evaluation can be limited to lesions that appear suspicious by NEE evaluation, or include lesions that are regarded inconspicuous by NEE. The impact of dermoscopy on sensitivity and specificity are expected to be highly dependent on the way in which it is used. This aspect has had limited attention in the literature so far.

Table 3 Different applications of dermoscopy

		TIME-FRAME	
		SINGLE EVALUATION	FOLLOW-UP
LESION APPRAISAL BY NAKED-EYE EVALUATION	SUSPICIOUS	**Single evaluation of suspicious lesions***	Follow-up of suspicious lesions
	INCONSPICUOUS	Single evaluation of inconspicuous lesions	Follow-up of inconspicuous lesions

* method of dermoscopy in chapter 8 & 9, as well as the majority of studies evaluating dermoscopy.

If the primary effect of dermoscopy in clinical practice is a reduction of the number of excisions, this implies that, as an effect, sensitivity could be compromised. This danger is illustrated in **chapter 7** by a histologically proven melanoma that was diagnosed as melanoma prior to dermoscopy, but as a benign lesion after dermoscopy. In this case the change in clinical diagnosis had no impact on the decision to excise this particular lesion, but it clearly illustrates the possible drawback of dermoscopy. In accordance with a few other studies, we found (**chapter 8**) that the increase of specificity does not result in a loss of sensitivity in case of dermoscopy experts.[27,30,31] This cannot be stated with certainty for the non-expert dermatologists because we had no follow-up data with respect to lesions that were not excised as a result of dermoscopy. Data on the safety of dermoscopy performed by non-expert dermoscopists is limited.[25,32] This issue, as well as the issue concerning the amount of training that is necessary to develop optimal dermoscopy skills (reflected in its impact on lesion management), may be a subject of future studies.

Summary and concluding remarks

In the first part of this thesis it was shown that clinical and histological characteristics of melanoma (patients) from CDKN2A mutated families differ from the general population in several ways. As evidence on the existence of distinct melanoma subtypes is increasing, our data suggest that there may be overrepresentation of certain subtype(s) in these families, which may impact future preventive and treatment strategies.

In the second part of this thesis we confirmed previous reports that surveillance of melanoma families results in early diagnosis. We showed that noncompliance is a major concern, especially for family members without a history of melanoma. The favourable tumour stage of self-detected (interval) melanomas and the observation that the majority of melanomas were detected within 6 months since the previous skin-examination stress the importance of monthly self-examination as a key element in successful melanoma surveillance. Based on our data we propose a risk classification system for members of melanoma families, that supports the development of a more tailored and cost-effective surveillance program. Our findings support the surveillance of second degree relatives from CDKN2A mutated families. Further studies are needed in order to optimize current surveillance strategies, including optimal surveillance intervals and to facilitate the assessment of melanoma risk in CDKN2A wild-type families.

In the third part of this thesis we demonstrated that dermoscopy in the context of an expert / high melanoma risk PLC setting, resulted in a considerable reduction of unnecessary excisions. Dermoscopy is therefore likely to reduce the burden of surveillance, improve cost-effectiveness and decreases the risk of overdiagnosis. The impact of dermoscopy on management in the expert PLC setting was considerably higher than in the general dermatology setting. These findings confirm and clarify the value of dermoscopy for the PLC setting and suggest that more studies are needed to clarify and optimize the role of dermoscopy in a non-expert dermoscopist, general dermatology settings.

References

1. Nagore E, Botella-Estrada R, Garcia-Casado Z, Requena C, Serra-Guillen C, Llombart B et al. Comparison between familial and sporadic cutaneous melanoma in Valencia, Spain. J.Eur.Acad.Dermatol.Venereol. 2008.
2. Chiarugi A, Nardini P, Crocetti E, Carli P, de G, V, Borgognoni L et al. Familial and sporadic melanoma: different clinical and histopathological features in the Italian population - a multicentre epidemiological study - by GIPMe (Italian Multidisciplinary Group on Melanoma). J Eur.Acad.Dermatol.Venereol. 2012;26:194-9.
3. Whiteman DC, Pavan WJ, Bastian BC. The melanomas: a synthesis of epidemiological, clinical, histopathological, genetic, and biological aspects, supporting distinct subtypes, causal pathways, and cells of origin. Pigment Cell Melanoma Res. 2011;24:879-97.
4. Whiteman DC, Watt P, Purdie DM, Hughes MC, Hayward NK, Green AC. Melanocytic nevi, solar keratoses, and divergent pathways to cutaneous melanoma. J.Natl.Cancer Inst. 2003;95:806-12.
5. Carey WP, Jr., Thompson CJ, Synnestvedt M, Guerry D, Halpern A, Schultz D et al. Dysplastic nevi as a melanoma risk factor in patients with familial melanoma. Cancer 1994;74:3118-25.
6. Hansson J, Bergenmar M, Hofer PA, Lundell G, Mansson-Brahme E, Ringborg U et al. Monitoring of kindreds with hereditary predisposition for cutaneous melanoma and dysplastic nevus syndrome: results of a Swedish preventive program. J.Clin.Oncol. 2007;25:2819-24.
7. Masri GD, Clark WH, Jr., Guerry D, Halpern A, Thompson CJ, Elder DE. Screening and surveillance of patients at high risk for malignant melanoma result in detection of earlier disease. J.Am.Acad.Dermatol. 1990;22:1042-8.
8. Vasen HF, Bergman W, van Haeringen A, Scheffer E, van Slooten EA. The familial dysplastic nevus syndrome. Natural history and the impact of screening on prognosis. A study of nine families in the Netherlands. Eur.J.Cancer Clin.Oncol. 1989;25:337-41.
9. Nederlandse Melanoom Werkgroep, Integraal Kankercentrum Nederland. Melanoom, Landelijke richtlijn, versie: 2.0. 2012.
10. Nagore E, Martorell-Calatayud A, Botella-Estrada R, Guillen C. Growth rate as an independent prognostic factor in localized invasive cutaneous melanoma. J Eur.Acad.Dermatol.Venereol. 2011;25:618-20.
11. Grob JJ, Richard MA, Gouvernet J, Avril MF, Delaunay M, Wolkenstein P et al. The kinetics of the visible growth of a primary melanoma reflects the tumor aggressiveness and is an independent prognostic marker: a prospective study. Int.J Cancer 2002;102:34-8.
12. Tejera-Vaquerizo A, Barrera-Vigo MV, Lopez-Navarro N, Herrera-Ceballos E. Growth rate as a prognostic factor in localized invasive cutaneous melanoma. J Eur.Acad.Dermatol.Venereol. 2010;24:147-54.
13. Martorell-Calatayud A, Nagore E, Botella-Estrada R, Scherer D, Requena C, Serra-Guillen C et al. Defining fast-growing melanomas: reappraisal of epidemiological, clinical, and histological features. Melanoma Res. 2011;21:131-8.
14. Berwick M, Armstrong BK, Ben-Porat L, Fine J, Kricker A, Eberle C et al. Sun exposure and mortality from melanoma. J Natl.Cancer Inst. 2005;97:195-9.
15. Kasparian NA, McLoone JK, Meiser B, Butow PN, Simpson JM, Mann GJ. Skin cancer screening behaviours among individuals with a strong family history of malignant melanoma. Br.J Cancer 2010;103:1502-9.
16. Aspinwall LG, Leaf SL, Dola ER, Kohlmann W, Leachman SA. CDKN2A/p16 genetic test reporting improves early detection intentions and practices in high-risk melanoma families. Cancer Epidemiol.Biomarkers Prev. 2008;17:1510-9.
17. Tucker MA, Fraser MC, Goldstein AM, Elder DE, Guerry D, Organic SM. Risk of melanoma and other cancers in melanoma-prone families. J Invest Dermatol. 1993;100:350S-5S.
18. Udayakumar D, Mahato B, Gabree M, Tsao H. Genetic determinants of cutaneous melanoma predisposition. Semin.Cutan.Med.Surg. 2010;29:190-5.
19. Yokoyama S, Woods SL, Boyle GM, Aoude LG, MacGregor S, Zismann V et al. A novel recurrent mutation in MITF predisposes to familial and sporadic melanoma. Nature 2011;480:99-103.
20. Goldstein AM, Struewing JP, Chidambaram A, Fraser MC, Tucker MA. Genotype-phenotype relationships in U.S. melanoma-prone families with CDKN2A and CDK4 mutations. J.Natl.Cancer Inst. 2000;92:1006-10.
21. Bishop DT, Demenais F, Goldstein AM, Bergman W, Bishop JN, Bressac-de Paillerets B et al. Geographical variation in the penetrance of CDKN2A mutations for melanoma. J.Natl.Cancer Inst. 2002;94:894-903.

22. Goldstein AM, Chan M, Harland M, Gillanders EM, Hayward NK, Avril MF et al. High-risk melanoma susceptibility genes and pancreatic cancer, neural system tumors, and uveal melanoma across GenoMEL. Cancer Res. 2006;66:9818-28.
23. Kefford R, Bishop JN, Tucker M, Bressac-de Paillerets B, Bianchi-Scarra G, Bergman W et al. Genetic testing for melanoma. Lancet Oncol. 2002;3:653-4.
24. Bafounta ML, Beauchet A, Aegerter P, Saiag P. Is dermoscopy (epiluminescence microscopy) useful for the diagnosis of melanoma? Results of a meta-analysis using techniques adapted to the evaluation of diagnostic tests. Arch.Dermatol. 2001;137:1343-50.
25. Kittler H, Pehamberger H, Wolff K, Binder M. Diagnostic accuracy of dermoscopy. Lancet Oncol. 2002;3:159-65.
26. Vestergaard ME, Macaskill P, Holt PE, Menzies SW. Dermoscopy compared with naked eye examination for the diagnosis of primary melanoma: a meta-analysis of studies performed in a clinical setting. Br.J.Dermatol. 2008.
27. Carli P, de G, V, Chiarugi A, Nardini P, Weinstock MA, Crocetti E et al. Addition of dermoscopy to conventional naked-eye examination in melanoma screening: a randomized study. J.Am.Acad.Dermatol. 2004;50:683-9.
28. de G, V, Grazzini M, Rossari S, Gori A, Alfaioli B, Papi F et al. Adding dermatoscopy to naked eye examination of equivocal melanocytic skin lesions: effect on intention to excise by general dermatologists. Clin.Exp. Dermatol. 2011;36:255-9.
29. Argenziano G, Cerroni L, Zalaudek I, Staibano S, Hofmann-Wellenhof R, Arpaia N et al. Accuracy in melanoma detection: a 10-year multicenter survey. J Am.Acad.Dermatol. 2012;67:54-9.
30. Argenziano G, Soyer HP, Chimenti S, Argenziano G, Ruocco V. Impact of dermoscopy on the clinical management of pigmented skin lesions. Clin.Dermatol. 2002;20:200-2.
31. Carli P, Mannone F, de G, V, Nardini P, Chiarugi A, Giannotti B. The problem of false-positive diagnosis in melanoma screening: the impact of dermoscopy. Melanoma Res. 2003;13:179-82.
32. Binder M, Schwarz M, Winkler A, Steiner A, Kaider A, Wolff K et al. Epiluminescence microscopy. A useful tool for the diagnosis of pigmented skin lesions for formally trained dermatologists. Arch.Dermatol. 1995;131:286-91.

Nederlandse samenvatting

Het melanoom van de huid is een kwaadaardige tumor uitgaande van de pigmentcellen in de huid. Als er in een familie sprake is van tenminste twee eerstegraads verwanten (ouders, broers, zussen en kinderen) met een melanoom, of drie melanomen bij tweedegraads verwanten uit één familie, wordt gesproken van familiair melanoom.[1] In ongeveer 40% van deze families wordt een pathogene kiembaanmutatie in het CDKN2A gen gevonden. CDKN2A mutatiedragers hebben een sterk verhoogd risico (ca. 67%) om tijdens hun leven één of meer melanomen te ontwikkelen. De meest voorkomende pathogene CDKN2A mutatie in Nederland is de p16-Leiden mutatie (c.225-243del19), vernoemd naar de regio waar deze mutatie voor het eerst werd ontdekt. Naast CDKN2A is er één ander hoog penetrant melanoom risico gen geïdentificeerd. Mutaties in dit gen, CDK4, zijn wereldwijd in slechts enkele families gerapporteerd. Het verhoogde melanoom risico in de families zonder een mutatie in CDKN2A of CDK4 wordt grotendeels toegeschreven aan (combinaties van meerdere) medium- en laagrisico melanoom predispositie genen, waarvan er inmiddels diverse zijn geïdentificeerd.

De prognose van melanoompatiënten is sterk afhankelijk van het stadium ten tijde van de diagnose. Om vroegtijdige detectie van melanomen te bewerkstelligen, wordt geadviseerd de huid van leden van melanoom families periodiek te controleren (surveillance). Een veel gebruikt hulpmiddel bij de beoordeling van verdachte huidlaesies tijdens het huidonderzoek, is de dermatoscoop. Met dit niet-invasieve instrument is het mogelijk structuren in de huid zichtbaar te maken die niet met het blote oog waarneembaar zijn.
In dit proefschrift hebben we getracht meer inzicht te krijgen in de effectiviteit en aanknopingspunten voor optimalisatie van surveillance van families met een verhoogd melanoom risico, door onderzoek te doen naar:

1. de klinisch en histologische kenmerken van melanoom(-patiënten) in deze families.
2. de wijze waarop melanomen worden gedetecteerd in het surveillance programma
3. de opbrengst van surveillance in relatie tot de kenmerken van de deelnemers en hun familie
4. de rol van dermatoscopie in de praktijk van surveillance.

Klinisch en histologische kenmerken

In **hoofdstuk 2** werden de klinische en histologische kenmerken van 429 melanomen van 182 melanoompatiënten uit families met een p16-Leiden mutatie vergeleken met melanoom (patiënten) uit de algemene populatie.

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1 Deze definitie is afkomstig uit de Nederlandse richtlijn melanoom van de huid van 2005, die het uitgangspunt vormde van dit proefschrift. In de ge-update richtlijn uit 2012 is deze definitie verlaten en vangen door: "tenminste 3 melanomen, waarvan twee bij eerstegraads verwanten, waarbij twee tumoren mogen voorkomen bij één individu (de aangedane personen moeten dan ook eerstegraads verwanten zijn)"

Leden van p16-Leiden families hadden hun eerste melanoom op jongere leeftijd (gemiddeld 39 jaar, 15 jaar eerder dan de algemene bevolking) en een sterk verhoogd risico op multiple melanomen (5 en 10-jaars cumulatieve incidentie, 23% en 35%), vooral als het eerste melanoom op jonge leeftijd (< 40 jaar) werd gediagnostiseerd. Een associatie tussen de locatie van het eerste en tweede melanoom werd in de p16-populatie niet gevonden, in tegenstelling tot de algemene populatie, waarin dit wel het geval leek te zijn. P16-Leiden melanoom patiënten hadden relatief vaker een superficieel spreidend melanoom en minder lentigo maligna en nodulaire melanomen. Melanomen in het hoofd hals gebied werden in de p16-Leiden populatie ten opzichte van melanomen op een andere locatie op een relatief jonge leeftijd gevonden, waar deze in de algemene populatie juist op relatief hoge leeftijd werden gediagnosticeerd. Deze bevindingen onderstrepen het belang van het starten van surveillance van deze families op jonge leeftijd en geven aan dat extra waakzaamheid nodig is na de diagnose van een (eerste) melanoom. Daarnaast sluiten onze bevindingen goed aan bij recentere theorieën over de (klinisch, histologisch, epidemiologisch, moleculaire) heterogeniteit van het melanoom en suggereren deze dat het verhoogde melanoom risico in p16-Leiden families mogelijk vooral bepaalde melanoom subtypen betreft (in het bijzonder die welke ontstaan volgens het zogenaamde "nevus pathway", dat geassocieerd is met patiënten met veel moedervlekken en intermitterende zonblootstelling). Toekomstig onderzoek (zoals naar het genetische profiel van melanomen in deze families) moet uitwijzen in hoeverre dit beeld klopt en wat de consequenties hiervan zijn met betrekking tot preventie, prognose en behandeling.

Effectiviteit van surveillance

In **hoofdstuk 3, 4 en 5** werd gekeken naar de effectiviteit van surveillance van melanoom families. In **hoofdstuk 3 en 4** werd gevonden dat melanomen van verwanten uit melanoom families die deel namen aan het surveillance programma gunstigere prognostische kenmerken hadden (dunnere Breslow dikte) dan melanomen van de eerste verwanten uit de familie met een melanoom, die ten tijde van de diagnose geen deel namen aan surveillance.

In **hoofdstuk 3** werd het moment van detectie van 226 melanomen van patiënten uit p16-Leiden families, die onder controle waren op de afdeling dermatologie van het Leids Universitair Medisch Centrum, onderzocht. Er werd gevonden dat 53% van alle melanomen werden gediagnosticeerd op reguliere huidcontroles. Zeven procent werd gevonden tijdens het allereerste huid-onderzoek, 20% tussen de vaste afspraken in (interval melanomen), en 20% werd gevonden bij patiënten die therapie ontrouw waren met betrekking tot de geadviseerde termijn tussen de huid controles. Therapie ontrouw was vooral een frequent fenomeen bij patiënten bij wie voor het eerst een melanoom werd gediagnosticeerd (46%). Melanomen van therapie-ontrouwe verwanten hadden een dikkere Breslow dikte dan die van therapie-trouwe verwanten. Ook bleek dat de

meerderheid (58%) van de melanomen binnen 6 maanden na de laatste huidcontrole werden gevonden. Er werd echter geen correlatie gevonden tussen de Breslow dikte en de tijdsinterval tussen de huidcontroles, voor een interval korter dan 24 maanden.

In **hoofdstuk 4** werd de opbrengst van surveillance in melanoom families met verschillende karakteristieken vergeleken. Er werd gevonden dat er significant meer melanomen werden gediagnosticeerd in families met de p16-Leiden mutatie in CDKN2A vergeleken bij families waarin geen mutatie in CDKN2A werd gevonden (hazard ratio (HR): 3.6, 95% betrouwbaarheidsinterval (CI): 1.4 – 9.0). In een subanalyse van ongeteste en families zonder CDKN2A mutatie werd een net niet significant verschil gevonden tussen families met twee melanoom patiënten ten opzichte van families met drie of meer melanoom patiënten (HR (≥3 t.o.v. 2): 2.2, 95% CI: 0.9 – 5.0). Verder werd gevonden dat het risico op een (volgend) melanoom aanzienlijk groter was voor melanoompatiënten ten opzicht van hun eerstegraads verwanten (HR: 3.9, 95% CI: 2.0 – 7.7).

In **hoofdstuk 5** werd de opbrengst van surveillance in tweedegraads verwanten van melanoompatiënten uit families met een p16-Leiden mutatie onderzocht. Er werd een incidentie van 2.2 / 1000 persoonsjaren gevonden in de gehele groep van tweedegraads verwanten, en 8.5 / 1000 persoonsjaren voor tweedegraads verwanten die deelnamen aan surveillance. Dit verschil is mogelijk toe te schrijven aan een selectie bias van tweedegraads verwanten met een grotere kans op een melanoom bijvoorbeeld op grond van moedervlekkenpatroon. Ten opzichte van de algemene bevolking vonden we een 'standardized morbidity ratio' (SMR)voor het risico op melanoom voor tweedegraads verwanten van 12.9 (95% CI, 7.2-23.4).

Over-diagnostiek

De incidentie van het melanoom van de huid is de afgelopen decennia sterk toegenomen. Omdat de mortaliteit in dezelfde periode minder sterk is gestegen, terwijl de behandelopties tot zeer recent niet zijn verbeterd, is gesuggereerd dat de stijging in melanoom incidentie mogelijk gedeeltelijk is toe te schrijven aan over-diagnostiek: het diagnosticeren van melanocytaire tumoren met een indolent beloop als melanoom. Over-diagnostiek is een bekend fenomeen bij screening voor andere vormen van kanker (o.a. borstkanker).

In **hoofdstuk 6** rapporteerden we een observatie met betrekking tot over-diagnostiek in melanoom families. Een patholoog met speciale expertise in de beoordeling van melanocytaire laesies reviseerde melanomen van leden van families waarin de p16-Leiden mutatie was gedetecteerd, geblindeerd voor de mutatie status van de individuele patiënten. Het is een herhaaldelijk gerapporteerd fenomeen dat in families met een CDKN2A mutatie vaker dan verwacht melanomen worden gediagnostiseerd bij verwanten die de mutatie zelf niet blijken te hebben. Het bleek in onze dataset dat bij 7 van 10 melanoom-

patiënten (met 8 melanomen, waarvan 7 in situ) zonder CDKN2A mutatie, de diagnose melanoom na revisie werd verworpen. Bij bewezen mutatiedragers gebeurde dit in een veel lager percentage (11%) van de melanomen. Dit illustreert dat bij surveillance van personen met een onterecht geanticipeerd sterk verhoogd melanoomrisico, overdiagnostiek een reëel risico vormt.

Dermatoscopie

In het laatste deel van het proefschrift (hoofdstuk 7 en 8) werd gekeken naar de invloed van dermatoscopie in de klinische praktijk. Tientallen studies hebben aangetoond dat dermatoscopie de sensitiviteit en specificiteit van de diagnose melanoom met het blote oog kan verbeteren. Er is echter minder bekend over de invloed van dermatoscopie op de beslissing om een verdachte gepigmenteerde laesie te excideren. Hiertoe werden twee studies verricht, respectievelijk (**hoofdstuk 8**) in de setting van een tertiaire pigmentpoli met dermatoscopie experts die hoog risico patiënten uit melanoom families controleerden en (**hoofdstuk 7**) in de setting van algemene dermatologen in perifere ziekenhuizen met een gemengde patiënten populatie.

Waar eerdere studies suggereren dat het belangrijkste effect van dermatoscopie een verbetering van de sensitiviteit is, vonden wij in beide settings dat dermatoscopie vooral leidde tot een afname van het aantal onnodige excisies van goedaardige laesies. Dit effect was het grootst in de dermatoscopie expert / hoog-risico patiënten setting (42%) en resulteerde in een significante verbetering van de specificiteit (van 0.53 met het blote oog alleen, naar 0.74 bij toevoeging van de dermatoscoop). In de non-expert / algemene populatie setting werd een reductie van het aantal excisies van 9% gezien (zonder significante verbetering van de specificiteit (van 0.64 naar 0.68)). In onze studies had dermatoscopie geen (significant) effect op de sensitiviteit, al werd de bewijskracht van de studies om deze te detecteren beperkt door het kleine aantal melanomen in de studie.

Hoofdstuk 9 geeft een samenvatting en bediscussieert de resultaten uit de voorgaande hoofdstukken. Op grond van onze bevindingen werden adviezen gegeven ter optimalisering van het beleid ten aanzien van families met verhoogd risico op melanoom (tabel 2, Summary and Discussion).

List of publications

J.I. van der Rhee, W.J. Mooi, N.A. Kukutsch, F.A. de Snoo, W. Bergman; Iatrogenic melanoma. Comment on: Melanoma epidemic: a midsummer night's dream?
Britisch. Journal of Dermatology, 2010 Feb 1;162(2):457-8

J.I. van der Rhee, W. Bergman, N.A. Kukutsch; The impact of dermoscopy on the management of pigmented lesions in everyday clinical practice of general dermatologist: a prospective study.
Britisch. Journal of Dermatology,2010 Mar;162(3):563-7

J.I. van der Rhee, W. Bergman, N.A. Kukutsch; Melanomen in disguise.
Nederlands Tijdschrift voor Dermatologie & Venereologie, 2010 Mei; 20 (5): 288-92

J.I. van der Rhee, R. van Doorn; Necrotiserende fasciitis,
Nederlands Tijdschrift voor Dermatologie & Venereologie,2011 Mrt; 21 (3):179-81

A.J. Gelderblom, **J.I. van der Rhee**, J. Ouwerkerk: A man with blisters on hand and feet.
Nederlands Tijdschrift voor Geneeskunde, 2011;155(45):A3994

J.I. van der Rhee, W. Bergman, N.A. Kukutsch; The impact of dermoscopy on the management of high-risk patients from melanoma families: a prospective study.
Acta Dermato-Venereologica, 2011 Jun;91(4):428-31

J.I. van der Rhee, P. Krijnen, N.A. Gruis, F.A. de Snoo, H.F.A. Vasen, H. Putter, N. Kukutsch, W. Bergman; Clinical and histological characteristics of malignant melanoma in families with a germline mutation in CDKN2A.
Journal of the American Academy of Dermatology, 2011 Aug;65(2):281-8

J.I. van der Rhee, F.A. de Snoo, H.F.A. Vasen, W.J. Mooi, H. Putter, N.A. Gruis, N. Kukutsch, W. Bergman; Effectiveness and causes for failure of surveillance of CDKN2A mutated melanoma families.
Journal of the American Academy of Dermatology,2011Aug;65(2):289-96

K.D. Quint, **J.I. van der Rhee**, N.A.Gruis, J.A. ter Huurne, N. van der Stoep, W. Bergman, N.A. Kukutsch; The association of MC1R variants and dermoscopic features in atypical nevi and melanoma from CDKN2A mutation carriers.
Acta Dermato-Venereologica,2012 Nov;92(6):587-92

K.D. Quint, **J.I. van der Rhee**, R.E. Genders, A.P.M. Lavrijsen; Cutane myasis na een vakantie in Frans-Guyana.
Tijdschrift voor Infectieziekten, 2013;8(3):94-98

J.I. van der Rhee, S.E. Boonk, S.C. Cannegieter, H. Putter, F.A. de Snoo, N. Gruis, N.A. Kukutsch, W. Bergman; Management of second degree relatives from CDKN2A mutated families.
July 2013, Accepted for publication in Cancer Epidemiology, Biomarkers & Prevention

J.I. van der Rhee, H. Putter, N.A. Gruis, R. Blanken, E.B. Cohen, M.B. Crijns, C.L. Hebeda, T. van Meurs, H. Neering, F.G. Rosweide, D.G. Snels, P.C. van Voorst Vader, N.A. Kukutsch , W. Bergman, H.F.A. Vasen; The effect of screening of melanoma families according to the familial CDKN2A mutation status and number of melanoma patients: a prospective study.
Submitted

List of abbreviations

ALM	acral lentiginous melanoma
AN	atypical nevi
BCC	basal cell carcinoma
CI	confidence interval
CMM	cutaneous malignant melanoma
FAMMM	familial atypical multiple mole-melanoma syndrome
FN	false negatives
FP	false positives
GEE	generalized estimation equations
HR	hazard ratio
LCR	Leiden cancer registry
LM	lentigo maligna
LMM	lentigo maligna melanoma
LUMC	Leiden university medical center
Mis	melanoma in situ
MPM	multiple primary melanomas
NEE	naked eye examination
NFDHT	Netherlands foundation for the detection of hereditary tumours
NM	nodular melanoma
NNS	number needed to screen
OR	odds ratio
PLC	pigmented lesion clinic
RR	relative risk
SK	seborrhoeic keratosis
SMR	standardized morbidity ratio
SSM	superficial spreading melanoma
TN	true negatives
TP	true positives

Curriculum Vitae

Jasper van der Rhee werd geboren op 18 augustus 1975 te Leiden. Na het behalen van het eindexamen op het Stedelijk Gymnasium in Leiden, begon hij in 1994 met de studie medische biologie aan de Vrije Universiteit in Amsterdam. Na het behalen van het propedeuse, startte hij in 1995 met de studie geneeskunde aan de Katholieke Universiteit Leuven in België. In 1996 werd de studie geneeskunde gecontinueerd aan de Vrije Universiteit in Amsterdam. Tijdens zijn studie was hij als student-assistent betrokken bij het curriculair onderwijs medische filosofie, medische ethiek en cultuur en gezondheid. Na een wetenschappelijke stage in Biratnagar (Nepal) in samenwerking met de Nederlandse Lepra Stichting, werd het doctoraal examen behaald in 2003, in 2004 gevolgd door het artsexamen. In dit jaar was hij gedurende 5 maanden als arts werkzaam in Flebologisch Centrum Oosterwal in Alkmaar. In december 2004 werd hij aangesteld als AIOSKO (Arts In Opleiding tot Specialist en Klinisch Onderzoeker) op de afdeling Dermatologie van het Leids Universitair Medisch Centrum en werd onder leiding van mw. Prof. Dr. W. Bergman en mw. Dr. N.A. Kukutsch aangevangen met het onderzoek dat leidde tot dit proefschrift. In juli 2013 ronde hij de opleiding tot dermatoloog af (opleider Prof. Dr. R. Willemze). Sinds september 2013 is hij als dermatoloog verbonden aan centrum Oosterwal en Medisch Centrum Alkmaar.

Jasper woont samen met Carla Sneek, hun dochter Lieke en zoon Jonathan.

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