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Malignant Melanoma

in Children and Adolescents

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Cover picture by Cecilia Berg, eight years old.
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“Life is what you make of it”.

To my beloved family

Ingalena, Christina, Cecilia and Claes-Henrik

Malignant Melanoma in Children and Adolescents

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ABSTRACT

The aim of the work reported in the present thesis was to investigate malignant melanoma in children and adolescents. Through epidemiological studies, we wished to investigate incidence, clinical factors and prognosis and to study the records. Also studied were etiological factors such as phototherapy in newborns and the effect of congenital nevi on the risk of malignant melanoma. Finally a genetic study of this age group was conducted using immunohistochemical analysis.

The bases for the studies were the Swedish Cancer Register, the Swedish Medical Birth Register and other related registers.

The incidence of malignant melanoma in children was constant, but in adolescents a doubling was seen for the past few decades. The most common type of malignant melanoma in adolescents is the superficial spreading type, the one most likely to be sun-related. Since the anatomical location is the same in adolescents as in adults, the same etiological factors are presumably of importance; but some other factor may reduce the induction time. For this reason, diagnosis is set at a median age of 55 in adults compared to 15 in adolescents. Median survival time was three years after diagnosis, 15.3% of patients dying as a result of the diagnosed condition, most around 20 years of age. No specific anatomical location was over-represented as fatal.

Phototherapy of newborns has probably been ruled out as a factor in the increased incidence of malignant melanoma: on the contrary phototherapy may well be a protective factor just as outdoor activities in childhood are.

Congenital nevi are often surgically removed unnecessarily: no malignant melanoma was found after review of nearly 4000 congenital nevi. It must be considered whether the reason for removal is the risk of malignant transformation or is merely cosmetic. Further, no clear association was found between congenital nevi and maternal illness/infection during pregnancy. Congenital nevi seemed to have had limited effects on social life, but had resulted in greater caution with regard to sun exposure.

Congenital nevi or their treatment seem to play no part in the increased incidence of malignant melanoma below the age of 20 years.

We found only one patient in 51 with adolescent malignant melanoma with a germline CDKN2A mutation. It is therefore likely that other genetic factors are of importance for the development of malignant melanoma in adolescents.

Knowledge of risk factors for the development of malignant melanoma, and of its early clinical characteristics, should as a matter of importance be spread to the medical profession and also to the general public.

Key words: *melanoma, epidemiology, cancer register, children, adolescents, incidence, phototherapy, hyperbilirubinaemia, etiological factors, congenital nevi, heredity, CDKN2A, germline mutation.*

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POPULÄRVETENSKAPLIG SAMMANFATTNING

Djävulsskinnet: dieffuils skindtt, dieffuels skingid.
sannolikt har namnet någon förklenande betydelse. Det kan i detta sammanhang påpekas, att såväl hud som skinn i det äldre språket användes ibland om personer, företrädesvis kvinnor, i ytterst förklenande betydelse.
(Avhandling framlagd av Erik Bruhn Lund 1931).

Malignant Melanoma in Children and Adolescents

Peter Berg

Malignt melanom (elakartad hudcancer) hos barn och ungdomar är en ovanlig sjukdom. Den fördubbling som skett av incidensen (förekomsten) efter pubertetens start hos ungdomar mellan 1972 och 1992 kan förmodas ha bakomliggande faktorer såsom livsstilsförändringar och ökad solexposition, medan ärftlighet kan vara av betydelse för maligna melanom under barndomen. Malignifieringsfrekvensen hos medfödda leverfläckar (congenitala nevi) är otillräckligt känd.

Patienter och metoder

De studier som presenteras här har kunnat genomföras tack vare Sveriges utmärkta persondata- och registersystem där intressanta uppgifter har hämtats ur cancer-, dödsfalls- och födelseregister m. fl. Målet för studierna har varit att för maligna melanom studera förekomst, utlösande faktorer, klinisk relevans av medicinska åtgärder, sociala beteenden, klinisk karakteristik och ärftliga faktorer hos barn och ungdomar. Metoderna har varit att samköra registerdata, att studera journaler samt att sammanställa samplat patientmaterial och genomföra enkäter. Vi har studerat ärftliga faktorer i utvalda grupper av patienter med maligna melanom och eftergranskat histopatologiska preparat samt utfört immunohistokemiska laboratorieanalyser. För statistiska bearbetningar har sedvanliga etablerade metoder använts.

Delarbete I: Berg P, Lindelöf B. Differences in malignant melanoma between children and adolescents. A 35-year epidemiological study. *Arch Dermatol 1997 Mar; 133: 295-97.*
Tanken med studien var att skapa en bas för huruvida maligna melanom hos ungdomar under 20 års ålder ökar i incidens precis som i den vuxna befolkningen. Vi genomförde en retrospektiv studie från Svenska cancerregistrets start 1958 till 1992. Vi fann 287 patienter under 20 års ålder med diagnosen malignt melanom. Av resultaten framgår att incidensen av malignt melanom före puberteten har varit konstant under tidsperioden, men att en fördubbling ses efter puberteten vid jämförelse mellan de två senaste decennierna i studien dvs. åren 1972 till 1992. Könsfördelningen är som hos vuxna, något mer förekommande hos kvinnor, än hos män med en snittökning av incidens ett år tidigare hos kvinnor vilket överensstämmer med pubertetsstart. Förekomsten är åldersrelaterad med högst incidens vid 19 års ålder. Utbredningen är densamma som hos vuxna dvs. vanligast på extremiteter hos kvinnor, medan män har det på bålen. 15,3 % dog av sin tumör med en median överlevnadstid på 3 år. Ingen av pa-

tienterna med genitala melanom dog och inget specifikt hudområde var överrepresenterat i dödsfallsmaterialet. Det är viktigt för kliniker att vara medvetna om förekomsten av maligna melanom efter pubertetsstarten.

Delarbete II: Berg P, Lindelöf B. Is phototherapy in neonates a risk factor for malignant melanoma development? A preliminary case-control study. *Arch Pediatr Adolesc Med 1997 Dec; 151: 1185-87.*
Målet var att bedöma om ljusbehandling på nyföddas känsliga hud vid diagnosen hyperbilirubinemi var en riskfaktor för att utveckla malignt melanom under åren 1973 till 1992. Med hjälp av samkörning av födelse-, dödsorsaks- och cancerregister utförde vi en retrospektiv studie på 30 patienter, dessa matchades med 4 kontroller med samma födelsedatum, sjukhus och kön. Vi fann ingen risk för ljusbehandling som utlösande faktor till maligna melanom, men median uppföljningstiden var bara 18 år. Ljusbehandling i större skala startade först på 70-talet, men idag ljusbehandlas cirka 5000 barn/år. Kan rentav ljusbehandling av nyfödda fungera som en förebyggande faktor?

Delarbete III: Berg P, Lindelöf B. Congenital nevocytic nevi: Follow-up of a Swedish Birth Register sample regarding etiologic factors, discomfort and removal rate. *Pediatric Dermatol 2002 Aug; 19(4): 293-97.*
Medfödda leverfläckar (congenitala nevi) både stora och små, opereras bort generöst. Av 3922 registrerade medfödda nevi i födelseregistret valde vi slumpvis var 20:e och skickade ut en enkät för att bedöma åtgärder och psykosociala effekter. Detta fungerade samtidigt som en kvalitetstest på födelseregistret. Denna gav vid handen att knappt 15 % av registrerade pigmentfläckar ej bedömdes som sannolika nevi. 40 % av leverfläckarna hade tagits bort och ingen av de resterande hade utvecklat malignt melanom. Ju större nevi desto vanligare och tidigare utfördes en operativ åtgärd. Enbart 8 % hade upplevt mobbning eller förändrat sina levnadsvanor exempelvis solning.

Delarbete IV: Berg P, Lindelöf B. Congenital melanocytic nevi and cutaneous melanoma. *Melanoma Research 2003:0 (in press).*
Som en fortsättning granskade vi incidensen av congenitala nevi och sammanställde världslitteraturen på området, samt eftergranskade histopatologiska fall av malignt melanom hos patienter med congenitala nevi. Vi fann att incidencen av congenitala nevi var 0,2 % varav 7 % utgjordes av stora nevi och 93 % små utan exakt definition av storleken. De två fall av koppling mellan congenitala nevi och maligna melanom som vi fann visade sig inte beläggas efter histopatologisk eftergranskning. Slutsatsen blev att bland nästan 4000 medfödda leverfläckar fann vi inget malignt melanom, även om detta naturligtvis kan påverkas av den 40 % operationsfrekvensen och korta uppföljningstiden (15 år).

Delarbete V: Berg P, Wennberg A-M, Touminen R, Sander B, Lundh Rozell B, Platz A, Hansson J. Germline CDKN2A mutations are rare in child and adolescent cutaneous melanoma. *Submitted to Melanoma Research.*
Ärftligheten har tolkats som en viktig faktor för utvecklandet speciellt av tidiga cancrar exempelvis i bröst, grovtarm och troligen även elakartade hudcancrar. Vår målsättning var att bedöma frekvensen av CDKN2A mutationer hos patienter med di-

agnosen malignt melanom före 20 års ålder. På 60 patienter genomförde vi en hudundersökning, tog blodprover för genetisk analys samt eftergranskade histologiska preparat. Vi fann en patient med en CDKN2A mutation med känd massiv ärftlighet. Mutationen var en “mis-sense in prolin-to-leucin substitution in codon 48”. Sökandet efter andra genetiska förändringar som bakgrund till tidiga maligna melanom får fortsätta.

LIST OF ORIGINAL PAPERS

This thesis is based on studies presented in the following papers, referred to in the text by their Roman numerals:

I. Berg P, Lindelöf B. Differences in malignant melanoma between children and adolescents. A 35-year epidemiological study.
Arch Dermatol 1997;133:295-97.

II. Berg P, Lindelöf B. Is phototherapy in neonates a risk factor for malignant melanoma development? A preliminary case-control study.
Arch Pediatr Adolesc Med 1997;Dec 151:1185-87.

III. Berg P. Lindelöf B. Congenital nevocytic nevi: Follow-up of a Swedish Birth Register sample regarding etiologic factors, discomfort and removal rate.
Pediatric Dermatology 2002 Aug;19 (4):293-97.

IV. Berg P, Lindelöf B. Congenital melanocytic nevi and cutaneous melanoma.
Melanoma Research 2003:0 (in press).

V. Berg P, Wennberg A-M, Tuominen R, Sander B, Lundh Rozell B, Platz A, Hansson J. Germline CDKN2A mutations are rare in child and adolescent cutaneous melanoma.
(Submitted to Melanoma Research).

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APPENDIX (PAPERS I-V)

1. ABBREVIATIONS

ALM	Acral lentiginous melanoma
AN	Acquired nevi
CDKN2A	Cyklin-dependent kinase inhibitor 2A (in human)
CN	Congenital nevi
DN	Dysplastic nevi
DNS	Dysplastic nevus syndrome
FMM	Familial malignant melanoma
HB	Hyperbilirubinemi
LMM	Lentigo maligna melanoma
MM	Malignant melanoma
NM	Nodular melanoma
PT	Phototherapy
SCR	Swedish Cancer Register
SMBR	Swedish Medical Birth Register
SSM	Superficial spreading melanoma
UV	Ultraviolet radiation

Omnium rerum primordia sunt dura (petr,chrysol,migne 56,656)
All vår början bliver svår, bättre går det år från år.
(Prinsens ABC-bok 1883, sid.62)

2. INTRODUCTION

The cause of malignant melanoma (MM) is not very well understood. We do not know the induction time, but young people developing a MM before the age of 20 present a specially interesting induction reason: hence the selection of this population for study in the present thesis.

Despite our very imperfect knowledge, various risk factors contributing to the development of the disease have been identified. One factor of importance is family history[1-3].

Approximately 5-10% of patients with MM report having a family member who is affected[4, 5]. Some of these cases occur in the absence of an identifiable predisposing dermatological condition, but others occur in the setting of the dysplastic nevus syndrome (DNS)[6]. DNS is a condition characterized by the early development of a large number of atypical moles[7]. It serves as a marker of melanoma predisposition as well as being a precursor of SSM[1, 8]. The likelihood of overt melanoma developing in these patients is quite high. Other factors associated with this familial MM are lower age at diagnosis, sporadic MM and the development of multiple primary MM[4, 9, 10].

As environmental factors increase in importance, the significance of genetic factors decreases. At the same time, many different genetic locus may possibly be of importance[11].

Malignant melanoma morbidity and mortality rates have increased over the past several decades worldwide[12-14]. No changes have been seen in the incidence during childhood and there are probably special reasons for the development of MM up to puberty[15-19]. In adolescence, first girls and, a year later, boys show a steadily increasing prevalence of MM up to the age of 20 years.The incidence of MM in adolescents has nearly doubled during the past few decades[14, 20].

A further risk factor addressed in different works are sun exposure and habits[21-24].

Of interest are changes in the ways of exposing the skin to ultra-violet (UV) radiation[25-29]. These involve vacation outdoor activities[30], sun-beds[21, 31], travel to sun intensive areas[32]; and the ‘cosmetic’ attitude still reflected in and strengthened by the media, that “black is beautiful”, although changes in attitudes are visible in countries such as Australia, New Zealand and the USA[33]. Most people get half their total UV dose in the first 18 years of life[34]. The skin ‘remembers’ all the UV radiation it has

received since the beginning of life. This may be recognised in a continually increasing incidence of malignant melanoma and also of other sun-induced skin tumours.[35-37]

Studies are urgently needed on the factors that trigger the malignant transformation of melanocytes into melanomas. It is commonly thought that MM of the skin develops by the transformation of melanocytes[38].

There is controversy over how to deal with sun protection[39]. Recent articles have shown that normal outdoor activities in childhood even have a protective effect against MM[30, 40]. The face is the area with the highest density of epidermal melanocytes, and UV radiation stimulates their frequency to a certain extent[41-43]. The face also has the highest density of MM. Different types of clothing offer varying degrees of sun protection. Jeans, for example, may have a sun block factor of 1500. This has led to the development of special bathing suits for children[44-46]. Used largely in Australia and New Zealand, they are now also available in Sweden. So far there is no consensus concerning different topical sun blocks[47]. Some studies have shown that the users of topical sunscreen preparations have more MM, but sometimes they lack control for selection biases such as the more frequent sunbathing attitudes in this group of sunscreen preparation users[48-50].

In early summer our daily newspapers overflow with advice on how to protect oneself from the dangers of sun exposure. A frequent discussion is whether some ingredients in the topical sunscreens could be carcinogens, though this has not so far been proved. A meta-analysis of 11 case-control studies found no association between sunscreen use and the development of MM[51].

It will clearly take time before we return to the general attitude of the 16th century that "white is beautiful".

Another route of exposure is sun beds, where Sweden is the country with the highest frequency of use. It is easy to understand why: the psychosocial impact of light in this country where the sun is absent for long periods. In psychiatry, white daylight rooms are used with very good effect in the treatment of seasonal depression[52, 53]. In dermatology many light therapies are used for skin diseases, no other speciality possessing such good knowledge of the benefits of UV radiation rightly used. However, the latest recommendation from the Karolinska Hospital Cancer Prevention Unit is not to use UVA sun beds more then ten times a year, even though the role of UVA (ultraviolet A radiation) in the pathogenesis of human MM in humans is not clear.

Phototherapy is used for 5-10 % of all newborn children in Sweden, especially the premature, who develop physiological jaundice, a transient, unconjugated HB. The treatment of choice for marked neonatal HB is daylight fluorescent light with a continuous emission spectrum from 320 to 700 nm and including small amounts of UVA radiation in addition to the therapeutic blue-light wavelengths[54]. This is believed to give few side effects, although some years ago two newborn babies in a Stockholm hospital were burned during treatment[55].

Soft newborn skin could conceivably be vulnerable to intensive light, and we know that skin penetrance is easier in newborns – probably even more so after premature birth – and studies show that visible light may cause immunomodulation in the skin[56].

Different histopathological types of MM may possibly have different trigger factors. Superficial spreading melanomas (SSM) are the ones that increase in incidence and are the most common in young people,[57-59] while nodular melanoma (NM) frequency is stable over time. In Asians and blacks one finds acral lentiginous melanoma (ALM) and in the older age groups lentigo malignant melanoma (LMN), which have even decreased in incidence. Specific MM such as the genital, ocular and internal varieties have been stable in incidence, or even decreased over time[60-63].

There are no clear studies of reduced incidence of congenital nevi (CN) during the past 30 years[64]. If the genetic factor is the most important, no change could be expected; but if infective agents are the triggers, decreased incidence – especially in countries with high standards of living – could be expected. World-wide attitudes concerning treatment of CN tend to favour surgical removal for cosmetic and anxiety reasons[65-67]. These may involve major surgical excision, and many clinics are dependent on this treatment. For these reasons it has been difficult to reach consensus concerning the treatment of CN[68, 69].

Epidemiology is the science that produces most hypotheses, and Sweden is one of the world leaders in this respect[70]. The country's reliable and long-standing health registers offer excellent bases for studies that may be of benefit to mankind. However, many of the new regulations and laws – the biobank law, for example – are out of touch with reality, and unfortunately the best hope for the patient is that their intentions are not being realised.

The purpose of this all-round introduction has been to outline MM-related topics that are of most relevance for the general population.

Docendo discimus
(Seneca D.Y)
Lära sig själv genom att lära andra.

3. BACKGROUND

History of malignant melanoma

In the fifth century BC Hippocrates, the father of the medical art, described 'black cancer' and may be regarded as the first dermatologist.

Modern European dermatology started at the end of the eighteenth century with the publication of Joseph Plenck's *Doctrina de Morbis cutaneis* in Vienna in 1776[71](Picture 1).

Picture 1. *Joseph Plenck De morbis cutaneis Vienna 1776.* *Private collection*	Picture

Dermatology was initially a surgical speciality and it is of interest to see that in the case of malignant melanoma (MM), we are returning to this frontier. The first obvious

presentation of a patient with a melanoma was that by Sir John Hunter in 1787. Hunter described the tumour as a *cancerous fungous excrescence*. The tumour is preserved as a specimen in the Huntarian Museum of the Royal College of Surgeons of England. René Laennec (1781-1826) at the Faculté de Médicine de Paris was the first to describe the melanoma as a tumour disease. He also introduced the term "melanosis", from the Greek for 'black'. The term "melanoma" (black tumour), was used by Sir R.Carswell (1793-1857) but "melanosis" was still widely accepted. Jean Darier (1856-1938), the French cutaneous histopathologist who found, upon microscopic examination, that some clinically amelanotic melanomas include "single cells with the true melanotic characters" containing pigment.

Epidemiology of Malignant Melanoma

In the early twentieth century, the world frequency of MM was around one case in 2000 people. By the 1930s the frequency in the USA had risen to 1/1500. The changed trends in sun bathing started in the late nineteenth century when European royalty travelled to the French Riviera on vacation. The Swedish king Gustav V was very fond of visiting Nice, but fashion still prescribed clothing that offered little risk of sun tans. In Sweden wealthy people started the travel trends in the 1950s and by the 1960s the travel agent Spies had full aircraft flying wealthy Swedish sunbathers to the Canary Islands. World incidence rose quickly; by 1990 there was 105 000 new cases of MM, 15% more than in 1985[72]. In the USA in 2000, one person in seventy-five developed MM[73-76].

Incidence in adults

MM incidence has been increasing in most populations of Caucasian origin for several decades[14, 77-79]. The highest incidence rates have been in New Zealand, Australia, North America and northern Europe, while cancer registers in East, South and South-West Asia have shown the lowest incidence rates[80-82]. The highest incidence rates in the world have been reported from Auckland, New Zealand, with age-standardised incidence rates exceeding 50 per 100,000 person-years[83]. However, some developed countries, including Sweden, have recently experienced declines in MM morbidity but not mortality[84-87]. The mean age of diagnosis in Sweden is 55 years. The increased incidence is seen mainly in patients with thin melanomas, but an increase in patients with thicker lesions has also been reported[88].

One factor suggested as contributing to the increases may be selection biases due to a more efficient health service in developed countries. On a 'national melanoma day' in Sweden in 1990 many MM were harvested, with a decline in incidence the following year. However, the overall incidence has increased in all Scandinavian countries, and in Sweden the average annual increase between 1960 and 1982 was 5.4% for women and 5.8% for men[87]. The largest increase has been observed for melanoma on the trunk among males. Some believe that the habitual status of men concerning the observation of changes in the skin is lower than in women. Some studies have shown that

it is often a partner who has recognized a tumorous change in a man's skin. Among females, the dominating increase has been for leg MM[89].

In blacks, MM are rare, predominantly situated on the sole of the foot and the palm of the hand[90, 91], but often large (sometimes more than 100 mm in diameter)[92]. Most studies conclude that, among whites, the trunk in males and the lower limbs in females have been the most susceptible sites[93]. Density is highest in the face and the hypothesis is that the intermittent and intense sun exposure of untanned individuals is more important for MM development than total lifetime exposure[94-98]. To explain this site-dependent susceptibility of melanocytes to malignant transformation Green proposed the following theory. Most MM with a neval component tends to occur on sites such as the trunk that are not exposed to sunlight constantly. Hence in less stable melanocytes, mutagenesis could occur more easily after intermittent or small total doses of sun radiation[99, 100]. On the other hand, MM at the sites of more stable melanocytes, such as the face, could be associated with high total doses of UV radiation[34]. SSM is associated with recreational patterns of sun exposure, and is highly associated with atypical nevi and acquired nevi(AN). In older people LMM occurs on sun-exposed skin areas on a total-dose basis without connection with nevi[101]. NM is intermediate and stable in incidence, while ALM occurs on body sites that are protected by thick keratin, and does not seem to be associated with nor nevi or sunlight and has the same frequency in all races[102].

Incidence in children/adolescents

Sun exposure during childhood/adolescence seems to have a substantial influence on the risk of MM[103-106]. An Australian study in 1984 showed that immigrants arriving at the age of 10 had a fourfold higher risk of developing MM than those arriving after their 15th birthday, provided that they were born in countries with less sun; but of course they also had a lower total sun radiation dose[82, 99]. Analysis indicated that it was exposure during childhood/ adolescence that was of importance[107-110]. However in more recent studies a lifelong history of sunburns was associated with a lower relative risk of MM[98, 111-113]. After controlling for cofounders some authors have not found any relationship between childhood/adolescence sunburns and MM[114, 115]. In the Kaskel 2001 study, outdoor activities during childhood have even been proved to be a protective factor for MM, and the reason could be the development of a photoprotective tan[30]. Many authors attribute the association of increased childhood sun exposure with MM to higher numbers of AN among children with high levels of sun exposure[57, 116]. The number of acquired melanocytic nevi – more than 50 – appears in many studies to be a very strong risk factor with relative risks of 4.8 to 54 times, and the latter in a Scottish population[117].

MM is six times more common in large CN , than in AN. Of patients with MM 35% had associations with melanocytic nevi[118]. Several studies have also shown that the presence of freckles in adolescence is a strong risk factor for MM[119].

International Registers

- NYU(New York University)-register of Large Congenital Melanocytic Nevi, prospective follow-up from 26 different countries.
- Nevus outreach registry. www.nevus.org

Malignant melanoma

In 2000, a total of 1,616 new cases of malignant melanoma (MM) were reported to the Swedish Cancer Register (SCR). MM constituted 3.6% of all cancers in Sweden, and was the eight most common malignant tumour in men and the ninth in females [70]

In the 1950s when we started to see rising incidence of MM, Lancaster publicised the effects of sun exposure especially in people with Caucasian ethnicity. Since then many reports have concerned the relation between UV radiation and the development of MM[120-123]. Relation to skin-type, intermittent and severe burns especially during childhood, are generally-accepted factors for developing MM[76]. How UV radiation induces MM is unclear, but UVA (320-400nm) can cause mutations in DNA, while UVB (280-320 nm) can cause mutations in oncogenes and tumour suppressor genes[124]. Changes in the ozone layer have also been discussed as factors. Method and duration of exposure to UV radiation, and predisposition, are further factors to take into account[125-127]. The International Agency for Research on Cancer (IARC) stated in its 1992 declaration that there was sufficient evidence that UV radiation causes MM and also non-melanocytic skin cancer.

In Sweden, MM incidence is highest in the highest socioeconomic groups, whose members for over thirty years have had the economic possibilities to spend vacations in sunnier climes. This is a factor for more common intermittent UV-radiation in unprepared skin[128]. The prognosis is also better for this group than for the general population[86].

Congenital Nevi

Melanocytic nevi are defects of development. Melanocytes develop from stem cells in the neural crest, splitting into leptomeninges and embryonic dermis. A congenital nevus (CN) is present at birth and composed of nevus cells[129]. Today many babies see the light of day in week 28 or even earlier and there is a more frequent use of phototherapy. Defining nevi that arise after birth, tardive nevi, is very difficult and no studies have shown the importance of these in relation to CN or MM. There is no consensus concerning the histopathological criteria for CN and no studies have been undertaken to differentiate these from tardive nevi.

CN can differ greatly in size, but the total melanocytic cell burden could be of importance for the risk of malignant transformation[130]. The clinical definition of CN varies greatly, clinical characteristics including small, mid-size, large, flat, homogeneous light-brown, speckled or dark, and 60 % have hair, which is a good prognostic factor[131-133]. Many different methods are in use today to remove nevi from a child's

skin. They include surgery, chemical peeling, dermabrasion, and curettage; but many of them only reduce the number of melanocytic cells.

In large CN, satellites can continue to develop for years after birth[134]. With present-day treatment including different types of surgery, MM are rarely seen in large congenital nevi[135, 136] (Table I)[69, 130, 137, 138].

Malignant melanoma (MM) usually develops after puberty. Large CN, larger than 50 cm^2 carry the greatest risk of malignant transformation and the MM develops at the edge of the CN at the DE-junction[139]. MM is more common in patients with large CN many satellites, axially distribution, but also skin fragility and erosions. More than half the patients in whom MM develop into large CN are under the age of ten. In this age group we have not seen any increased incidence of MM[138].

Table I. Risk of malignant melanoma in large congenital melanocytic nevi as shown by literature review.

Authors, Year	No of patients with nevi	No of MM	Estimated risk Raw %
Pers, 1963	110	2	2.0
Greenley, 1965	56	6	10.7
Lorentzen, 1977	151	3	2.0
Osumi, 1983	87	4	4.6
Quaba, 1986	39	2	4.5
Gari, 1986	47	1	2.0
Hori, 1989	154	7	4.5
Swerdlow, 1995	26	2	8.0
Marghoob, 1996	92	3	3.3
Dawson, 1996	133	0	0.0

No prospective studies have shown any relation between large CN and MM. Apart from histopathology, dermascopy is a diagnostic option where the clinical characteristics are seen in CN[140]. Why CN develops during fetal life is unclear, but genetic factors may be of importance[69, 141, 142].

Another possible cause of CN could be the presence of c-met proto-oncogene products in children with neurocutaneous melanosis. Sixty percent of these children have more than 50 CN satellites[143, 144].

Future molecular and genomic research may be able to differentiate between acquired and congenital nevi.There is a problem to make adequate prospective studies due to

the worries about the pigmented lesion and the wish of the patients to do surgical cosmetic treatment[68].

The incidence of CN varies greatly in different studies, between 0.025%, in the large studies to 5.9% in the more selective ones (Table II)[64, 67, 145]. There are no reports of increased incidence of CN.

Table II. Incidence of Congenital nevi as shown by literature reviews.

Authors, Year, (Ref.)	No of patients	Size of CN	No of patients with CN	Incidence (%)
Walton, 1976	1058	small	41	3.9
Alper, 1979	2645	all	28	1.1
Castilla, 1981	531831	all	1083	0.2
Kopf, 1985	601	mid-size	15	2,5
Sigg, 1990	939	small and mid-size	55	5.9
Ruiz–Maldonado, 1992	not given	large	not given	0.025

Today we believe that nodular MM in children develops from the deeper parts of the skin and is not related to UV radiation. Reduction therapies such as dermabrasion could be efficient prophylactically to reduce the total amount of melanocytic cells at risk for malignant transformation[146].

Table III. Differential diagnosis for congenital nevi.

Café-au-lait-spots
Common acquried Nevi
Large atypical nevi, dysplastic nevi
Sunburn-freckles
Spitz nevus
Spindle cell nevus of Reed
Suttons nevus, halo nevus
Becker nevus
Vascular nevus
Nevus Spilus
Urticaria pigmentosa
Extramammary nipple

Acquired nevi

The term nevus has become firmly entrenched in dermatological usage. Its Latin origin is the word for " maternal impression". Nevus cells are the melanocytes of which these lesions are comprised, and any factor operative during fetal life is capable of inducing developmental defects. Previously all congenital abnormalities were thought to be caused by hereditary factors, but when in 1941 paediatricians revealed that maternal rubella infection during pregnancy may jeopardise the developing embryo it was realized that environmental factors could be of importance[27, 147].

Though in many development defects the etiology remains obscure, virus infections, drugs, alcohol, metal excess or deficiency or systemic disease in combination with genetics may be of importance[61, 148]. Frequent use of immunosuppressive agents in transplant patients increases the amount of nevi: these patients are given information concerning sun protection[149].

Many studies have indicated a relation between sun exposure and nevi distribution[100, 117]. Common body sites for nevi are those that receive high amounts of UV radiation. The prevalence of nevi shows large geographical difference and is related to skin type.

Few people have a melanocytic nevus at birth and the maximum number of nevi on any one individual is found in early adult life[150-153]. This is of interest when one realises that most people have acquired half of their lifetime UV radiation dose before the age of 18[154]. After early adult life there is a slow decline in numbers of nevi until in old age few or even no nevi are found.

Small AN are less than 5mm and those over 6 mm are reckoned as large[148]. Large AN have often autosomal dominant heredity. In the general population dysplastic nevi are quite common but the presence of many DN may be a potential precursor of MM[155, 156].

Histopathology is of importance in many unclear cases, but studies have confirmed that the dermatologist's clinical diagnosis is the one most likely to be correct. What is seen clinically is what appears in the microscope (Table IV).

Table IV. Different clinical types of melanocytic cells presentation in the skin.

Acquired nevi
Lentigo
Congenital nevi
Café-au-lait macule
Nevus spilus
Hypochromic nevus

Hyperbilirubinaemia and phototherapy

Born in Arras in 1758, Noël Retz described in 1790, in the third edition of his “Des maladies de la peau et de celles de l´esprit (telles que les vapeurs, la mélancolie, la manie) qui procèdent des affections du foie” (Diseases of the skin and of the spirit such as the vapours, melancholy, mania, proceeding from disorders of the liver), the disastrous consequences of neonatal icterus (Picture 2).

Picture 2 *Noël Retz. Des maladies de la peau. 1785, Private Collection.*	Picture

In 1845 photodermatology came into being when Foucault attributed the disorders caused by light on the skin to ultraviolet rays. Later on, attention was drawn to the role of “chemical rays” in the sunburn of pellagra and the possibility of protecting oneself with ‘uranium glass’ which absorbs these rays.

Finsen’s work *La photothérapie* published in Paris in 1899 describes the benefit of using UV light for dermatological diseases. Half a century later, in 1956 in the USA a nurse made the clinical observation that newborn babies in her nursery that were positioned closest to the windows were less jaundiced. Since the beginning of the 1970s phototherapy has been used increasingly for the treatment of physiological jaundice, a transient, unconjugated hyperbilirubinaemia (HB) common in a large percentage of premature infants and some other neonates. Yet today HB (65%) remains one of the

most common morbidities in premature infants[157]. Immaturity was the second most important cause, accounting for 24% of these deaths.

In newborn babies, the skin is not mature. All the anatomical structures are there but enzymatic, hormonal and metabolic function is not yet fully developed. One function of the skin is to protect the organism from radiation and to give an immunological response mediated partly by the Langerhans cells, and newborns have an incompletely activated immunological defence. We also know that visible light causes immunomodulation and we believe UV light is a risk factor for the development of MM[56]. Often the newborn's skin type is not known, nor is its ability to protect itself through pigmentation. The bronze-baby syndrome, the most common side effect of PT, is a transient, dark discoloration; but some cases of skin burning have also been seen lately. The most common phototherapeutic measure for hyperbilirubinaemia is white, blue or green light, though control of the emission spectrum is often insufficient and difficult. The untanned epidermis transmits most light in the visible wavebands[158, 159].

Since the 1950s in the USA and the 1970s in Sweden phototherapy (PT) has been used to treat newborns with hyperbilirubinaemia. The use of PT has increased every year and today around 5000 children in Sweden receive this treatment for a longer (one week) or shorter period.

Bilirubinaemia neonatorum

In foetal life un-conjugated bilirubin passes through the placenta to the mother. From birth the concentration of bilirubin increases during the first two to five days from a low level up to 200-350 mikromol/l and the icteric child becomes yellow, first in the face and then the rest of the body and nearly 20% get HB. If the child is premature the risk for icterus is increased. The reasons for the HB are a higher turnover of haemoglobulin (70 days), non-developed hepatocellular enzyme system, persistent ductus venosus flow and high beta-glukuronidase activity in combination with a sterile intestine.

Other risk factors for HB, include incompatibility, polycythaemia and haemorrhagia. There is a risk of kernicterus, as well as encephalopathy with cerebral palsy or even death. Phototherapy is the treatment of choice for marked neonatal HB[160].

The genetics of malignant melanoma

Genetics research is a very young science but one that is developing very fast worldwide.

Familial malignant melanoma (FMM) is an autosomal dominant disease with incomplete penetrance, and develops mostly in patients with the heredity of DNS[1]. Germline mutations in the tumour suppressor CDKN2A gene in a locus on chromosome 9p21 have been identified in FMM kindreds and mutations in the CDK4 gene in a few families[3, 161-163]. There are other genetic factors of importance such as numerous

melanocortin 1 receptor (MC1R) gene variants, specifically Asp48Glu which has the highest odds ratio[11]. This is also associated with an increased risk of MM. FMM is therefore believed to be a genetically heterogeneous condition.

One important dermatological child disease is the xeroderma pigmentosum, which is a genetic disorder where the incidence of MM is extremely high[164, 165]. Xeroderma pigmentosum patients are photosensitive and they develop actinic keratosis and tumours such as squamous cell carcinoma as well as basal cell carcinoma during childhood/adolescence. Compared with the general population, a patient with xeroderma pigmentosum has a 2000-fold greater risk of developing MM before the age of 20[166]. In xeroderma pigmentosum patients there is a defect in the melanocytes for DNA repair after UVB exposure[167]. Several different chromosomal loci have been of interest to study. Differential analysis has been of importance in genetic studies of MM such as molecular genetics, cytogenetics and linkage analysis.

Histopathology

The possibility of staining slides for microscopy was the beginning of histopathological research. Histopathology has seen tremendous evolution during the past century. In 1948 Spitz presented the juvenile malignant melanoma or Spitz nevus, which is today considered to be benign. Allen introduced distinct criteria for differentiating between primary melanomas and metastases, but it was not until Clark in 1969 presented the micro-staging system with different levels of lesion that good prognostic possibilities were created[168]. The year after, Breslow introduced measurement of maximum lesion thickness, with which he indirectly estimated the volume of the tumour[169]. The histopathological types are presented in Table V.

The histopathological diagnosis of melanoma in children is difficult because of the occurrence of lesions with indeterminate diagnosis, such as atypical Spitz tumours[170]. The distinction between MM and Spitz nevus or pigmented spindle cell nevus is one of the most difficult problems in pathology and may lead to overdiagnosis of melanoma[171-173]. In some studies reviewing MM, 33% have not been diagnosed as MM[172]. For many years there have been great difficulties in achieving consensus concerning histopathological criteria for DN as a possible precursor to MM with hereditary impact[174]. Many studies report remnants of melanocytic nevi in 20 to 30% of melanoma cases, topographically below or beside the MM. The reported frequency of remnants of small CN in association with MM is between 1-9%[15, 129]. MacKie demonstrated that 44% of MM arising in patients under 30 years of age developed in small nevi present either from birth or from early childhood[155]. At present, however, one problem related to the diagnosis of small CN is the lack of specificity of some of the histopathological criteria considered to be characteristic of congenital onset[175].

Table V. Histopathological criteria for the differentiation of benign nevus cells from small melanoma cells, small superficial congenital nevi and dysplastic nevi.

Naevus cells	**Small melanoma cells**
No continuity with adjacent melanoma cells	Continuity with adjacent melanoma cells
Nests do not vary in size and shape	Nests vary in size and shape
Nests do not tend to confluence	Nests tend confluence
Fibrillary collagen between indivudual cells	Fibrillary collagen around each nest
Melanocytes show maturation	Melanocytes do not show maturation
Absence of cytological atypia	Presence of cytological atypia
Absence of mitotic figures	Presence of mitotic figures
Monomorphous appearance of nuclei	Pleomorphic appearance of nuclei
Inconspicuous necleoli	Prominent nucleoli
Melanin pigment generally absent	Melanin pigment commonly present

Small superficial congenital nevi	**Dysplastic nevi**
Melanocytics cells spread between collagen boundless in the reticular dermis in single units or in an Indian file pattern	Persistent lentiginous melanocytic hyperplasia
Adnexocentricity and angiocentricity	Atypical melanocytic hyperplasia
No involvement of the lower third of the reticular dermis	Lammelar fibroplasia
	Concentric eosinophilic fibroplasia
	Sparce patchy lymphcytic infiltrate

The histopathology of CN is not fully conclusive or reliable, since differences may depend on the density of the sections and other factors[176]. MM de-novo has an even worse prognostic situation than MM associated to DN, because neourovasculation is an important mechanism for spreading MM to lymphatics and generating metastasis.

Table VI. Histopathological characterisation of malignant melanoma.

Superficial spreading melanoma
Amelanotic malignant melanoma
Nodular malignant melanoma
Lentigo maligna melanoma
Acral lentiginous melanoma
Definition of Breslow index and Clark levels I-IV and vertical growth fase

Melanogenesis

Historically, two of the colours of the medical ribbon – yellow and black – represented bile, which was commonly believed to cause skin pigmentation. Abnormal skin colours, which are very notable in black people, have been the subject of a number of interesting works. *Dissertation physique à l`occasion du nègre blanc,* by Louis Moreau de Maupertius (1698-1759) put forward a peculiar theory according to which black people´s skin colour was caused by 'black jaundice'.

Picture 3 *Pierre Louis Moreau de Maupertius. Dissertation physisque à l'occasion du nègre blanc, Paris 1744. Private Collection.*	***Picture 4*** *Claude Nicolas Le Cat. Traité de la couleur de la peau humaine, 1765. Private Collection.*

Many of the greatest dermatological findings were made by French dermatologists in the late nineteenth century. In 1847 T. Schwann first described of the microscopic pigment cell, and its star-like shape, dendrites and granules prompted him to name it the "stellate cell". The histopathologist Unna found in 1893 that the pigment cell was located in the deep layers of epidermis. Masson wrote in 1926 that pigment cells migrated from the neural-tube region to their final destination and that this explained the origin of human melanoblasts. In collaboration with Bourquelot in 1895, it was concluded that oxidation of colourless substances in mushrooms could cause changes in pigmentation. An interesting work by Bloch describing the conversion of tyrosine into melanin in basal cells of the epidermis, he named these cells melanoblasts. Melanocytes originate from the neural crest and migrate during embryogenesis to different sites in the human body. We know from the colour of skin and hair that these are im-

portant targets. Other migration sites are the leptomeninges of the brain (neurocutaneous melanosis), the mucosal membranes and the choroids of the eyes. To start melanogenesis, the skin needs to be exposed to UV light and the increased melanin production causes a darkening of the skin. Prota demonstrated clearly in 1998 the different melanins produced by melanocytes: the eumelanins and the pheomelanins[177].

Melanocytes are located in the basal layer of epidermis where together with the surrounding keratinocytes they form the epidermal melanin unit[178]. Melanocytes produce melanin, which varies with age, ethnicity, anatomical location, hormones and UV radiation stimulation. The major function of melanin in man is assumed to be protection of the lower layers of the skin from UV radiation, which activates the melanocytes to start melanogenesis by transferring melanin to the surrounding keratinocytes. Melanin is one biological weapon for protection against UV radiation; others are urocanic acid in sweat, hyperkeratinization and beta karotinoids.

Les femmes et les médecins savent seuls combien le mensonge est nécessaire aux hommes. (Anatole France, histoire comique)
Kvinnor och läkare är de enda som vet hur nödvändig och välgörande lögnen är för människorna

4. AIMS OF THE INVESTIGATION

The decision to investigate MM in children/adolescents was made in 1996. The planned epidemiological study of incidence, clinical factors and prognosis, and a study of the records, resulted in the research reported in *paper I.*

The great difficulties in obtaining complete material for a study of the relevant clinical records has meant that this aspect of the investigation remains a future possibility. We had also planned a case control study of hyperbilirubinaemia in newborns and phototherapy treatment. *Paper II* thus became a preliminary case-control study to elicit whether phototherapy in neonates may constitute a risk factor for malignant melanoma.

We further wished to investigate clinical and etiological aspects of CN, evaluate its incidence and removal rate and to conduct a histopathological review in relation to MM. This is discussed in *papers III and IV*.

In 2000 Professor Ulrik Ringborg, recommended that a genetic study would be of interest in this age group of patients, and the relevance of genetic studies to the general aim of the present investigation is realised in *paper V.*

Plans to enlarge the phototherapy study to other Scandinavian Countries were abandoned after contact with the heads of the leading universities because of scarcity of doctors willing to work in this field.

p + 5e + 3h =happiness
p= personal qualities, like adaptivness, attitude of life
e= excistence like health, friends and financial stability
h= higher values like ambitions and expectancies of life
(British researchers after 1000 interwievs with selected people published 2003)

5. METHODS

The Swedish Cancer Register (I-V)

The Swedish Cancer Register (SCR) in Stockholm has collected information on cancer incidence in Sweden since 1958, when compulsory registration began. Reports of diagnosed cancer come from both clinicians and pathologists. Thus, most cases are reported from two sources. The SCR contains information about all malignant neoplasms, certain precancerous tumours and some histologically benign tumours (from the central nervous system and the urinary tract). The registration follows guidelines from the World Health Organisation (WHO). Information from death certificates is available to the Register, supplying date and cause of death. This information is updated annually. Since 1982 all registration and validation of incoming data has been done at six local cancer registries and the data has then been accumulated at the SCR. The local registries are associated with the oncological centres in each medical region in Sweden.

The following diseases are reported to the Cancer Register

- All definitely malignant neoplasms (e.g. carcinoma, sarcoma, malignant lymphoma, leukaemia and malignant teratoma).

- Carcinoid tumours of the digestive organs, granulosa-theca cell tumours of the ovary, thymoma, adamantinoma and chordoma.

- Histologically benign tumours of the central nervous system and meninges, transitional cell papillomas of the urinary tract, all tumours of the endocrine glands (except the thyroid) and the chromaffin system.

- Precancerous lesions of the lip, mouth, larynx, bronchus, trachea, cervix uteri, skin, vulva and vagina, gastrointestinal polyps with suspected malignancy, bronchial adenomas, atypical intraductal proliferations of the breast (carcinoma *in situ* type) and adenoma phyllodes, precancerous endometrial lesions, hydatidiform moles of placental tissue and ovarian cystadenomas with suspected malignancy.

The quality of the SCR has been subject to many studies. When the diagnosis had been histologically confirmed, the registration deficit was 2%. The Register records 96-97% of cases of all cancer. For skin cancer the reporting is close to 100%[179].

Basal cell carcinoma is now included in SCR (Table VII).

Table VII. Data registered in the Swedish Cancer Register

From Cancer Reports
Unique personal identification number
Sex
Name
Domicile
Hospital
Hospital department
Hospital-record, number and year
Pathology/cytology department
Specimen number, and year *(when specimen was taken)*
Site of tumour . Code based on 7th WHO revision
Tumour serial number *(when more than one primary was diagnosed)*
Malignancy *(yes/no)*
Histological type (*WHO/HS/CAC/24.1 histology code)*
Basis of diagnosis
Clinical only
X-ray
Histological examination
Autopsy with histopathological examination
Cytological
Gross examination at surgery
Autopsy without histopathological examination
Date of diagnosis
Died of cancer *(yes/no)*
Diagnosis made incidentally at autopsy *(yes/no)*
From death certificates
Date of death
Causes of death

The Swedish Medical Birth Register (II,III,IV)

The Swedish Medical Birth Register (SMBR), covering all Sweden, started in 1973. Registration was initially based on a medical birth registration form as a part of a standardised record system for maternal health, obstetrics, child health and neonatal health

care. A copy of the medical birth registration form was sent to the National Board of Health and Welfare for all newborns, live as well as stillborn. In 1982 the medical registration form was replaced with copies of the medical records. The aim of the SMBR is to permit analysis of risks to mother and child during pregnancy, delivery and the neonatal period as well as to provide statistics for general use. Other reasons are risk analysis regarding foetal trauma or disease, illnesses, mortality and social information such as smoking or other abuse (Table VIII). For births in e.g. 1993, the dropout rate was about 1.1 % of the total and about 1.6% for perinatal deaths. About 80 % of all births take place at specialised hospitals. Only 4 % are born at hospitals with neither obstetric nor paediatric specialist departments[180]. The annual count of newborns is from 86,000 to over 120,000. The level of registration of Swedish newborns in the SMBR is 98%.

Table VIII. Data registered in the Swedish Medical Birth Register. Patient-related information concerning.

PREGNANCY
CLINIC
duration of pregnancy
delivery
mother´s and child´s diseases
operations
anesthesia
child gender
weight
length
head-size
status at birth
social situation

The Swedish Cause-of-Death Register (I,II,V)

The Swedish Cause-of-Death Register (SCDR) prospectively collects information on date and cause for all Swedish citizens. Cause-of-death statistics have been collected in Sweden since 1749. Since 1994 the National Board of Health and Welfare has been responsible for a yearly publication on causes of death, the underlying cause being registered according to the ICD. All deaths in Sweden must be certified by a physician. Until 1980 if a malignant tumour was reported as the underlying or contributory cause of death, the tumour was recorded as the underlying cause. In 1981, the registration procedure was harmonised with WHO recommendations[181].

The Register makes no distinction between deaths caused by cutaneous melanoma or non-cutaneous melanoma: for exact information, the individual death certificate has to be checked. The main source of error in the classification is the reliability of the cause of death certified by the physician (Table IX) . The age of the patient may be of importance, since errors of reported underlying cause of death appear to be more frequent among the elderly. During the past few decades, there has been a secular trend towards a decreased autopsy frequency in Sweden, from 50% of all deaths in 1970 to 15% in 2000.

Of all cases with MM registered as the underlying cause of death, an autopsy was performed in 5.5% of the males and 6.8% of the females. These circumstances may have increased misclassification in the cause-of-death statistics.

Table IX. Variables registered in the Swedish Cause-of-Death Register

Personal identification number
Date of death
County
Community
Type of death certificate
Disease or condition directly leading to death
Number of antecedent causes
Up to six antecedent causes *(diagnoses)*
Municipality
Sex
Age
Civil status
Country of birth
Year of Swedish citizenship *(if immigrated to Sweden)*

The Swedish personal identification number (I-V)

Every person permanently living in Sweden, which has a population of nearly nine million, has a unique personal identification number, which is used in all population statistics, health services and official services. It is composed of six digits based on year, month and day of birth, with a registration number (three digits) and a check digit for validation purposes. The identification number is not affected by name changes, and allows record linkage between different registers.

Immunohistochemical analysis (V)

EXTRACTION AND SEQUENCING OF DNA FROM BLOOD SAMPLES:

Preparation of DNA templates for sequencing. Genomic DNA from whole blood from 60 subjects was isolated at the Center for Genomics and Bioinformatics, Karolinska Institutet using commercial extraction kits (Gentra Systems, Inc., MN, USA). Direct bi-directional sequencing of exons 1 and 2 of CDKN2A was performed. Exon 1 and exon 2 were first amplified using primed pairs AP161/AP162 and AP163/PP16-2RE, respectively. The exon 1 amplification was performed using 200 ng of genomic DNA in a total reaction volume of 20 µl with 1 M betaine, 200 µM dNTPs, 1.5 mM $MgCl_2$, 500 nM of each primer, 1 U AmpliTaq® DNA polymerase and 1X GeneAmp® PCR buffer. The amplification consisted of initial denaturation at 94°C for 4 min followed by 35 repeated amplification cycles at 94°C 20 s, 57°C 20 s and 72°C 30 s. Finally, samples were incubated at 72°C for 7 min and soaked at 4°C. For 11 samples the DNA concentration was low (<60 ng/µl), requiring amplification of these samples for 40 cycles. The PCR for exon 2 of CDKN2A was identical to the amplification of exon 1 except that in total 19 samples were run for 40 PCR cycles. The PCR products from CDKN2A exon 1 and exon 2 were electrophoresed in a 1.6 % agarose gel with ethidium bromide at 90 V and purified using the QIAquick Gel Extraction Kit (Qiagen, GmbH, Hilden, Germany).

Direct DNA sequencing. A fraction of the purified PCR product was used as a template in automated sequencing. We used the ABI Prism® BigDye™ 2.0 Terminator Cycle Sequencing Kit version 2.0 in an ABI Prism® 310 genetic analyser according to the manufacturer's instructions. The primers used in the forward and reverse sequencing reactions were the same as used for the PCRs except that primer PP16-2RE was replaced by primer AP164.

Statistical methods (I,II)

Paper I

To evaluate hypotheses of variables in contingency tables, the chi-square test was used or, for small expected frequencies, Fisher's Exact Test. In addition, descriptive statistics and graphical methods were used to characterise the data. All analyses were carried out with the SAS system, version 6.08. The level of significance chosen was 5%.

Paper II

To evaluate hypotheses of variables in contingency tables Fisher's Exact Test was used. In addition, descriptive statistics were used to characterise the data. All analyses were carried out with the SAS System (Statistical Analysis System; SAS Institute 1990a, 1990b, 1990c) and the level of significance was set at 5%.

Questionnaire (III, V) (Table X,XI)

Paper III.

Table X. Items included in the questionnaire for congenital nevi.

Items	**Reason for inclusion**
Age, sex	Relation to gender and age
Surgical removal	Removal frequency, medium age, technique been used, by which specialist and where, fol-low-up treatment
Characterization and localisation	Size, localisation, colour, hair frequency (V.A.S) and appearance of not-removed CN
Patient's illness	Any related disease
Social discomfort or bullying	Cosmetic or other discomfort caused by the nevi (V.A.S)
Changed social activities	Current participation in sports and activities (V.A.S)
Sun behavior	Whether scar or nevi changed sun behavior (V.A.S)
Relatives with CN	Hereditary factor in close relatives
Mother's impact with smoking	Smoking during pregnancy as a risk factor for maltransformation of nevi cells
Mother's infections/ illness	A cause of CN during pregnancy
Mother´s illness, now or earlier	CNs connection with other diseases

(V.A.S) = Visual Analogue Scale

Paper V.

Table XI. Questionnaire for familial malignant melanoma.

Identification number
Gender
Age
Year and month of diagnosis
Treatment
Follow-up time
Recurrence
Hair and eye colour
Skin-type
Counts of acquired nevi and dysplastic nevi
Freckles and location
UV-light actual use and earlier
Amount of sunburns
Other diseases
Relatives with skincancer
Location of MM
How it got diagnosed
Surgery before and after
Other skin diseases
Psychosocial effects of diagnosis

An additional questionnaire concerning use of UV-light, sun-beds, sunburns and changes in attitudes to UV-light after MM diagnosis etc. has not yet (April2003) been evaluated.

Children is a gift from the Lord
Barn äro en herrens gåva
(the Bible, Psalms 127)

6. PATIENTS

Paper I Epidemiology

The study reported in Paper I included all 287 cases of MM in patients below twenty years of age registered in the Swedish Cancer Register during the years 1958-1992.

Paper II Phototherapy

Paper II covers all 30 cases (17 females and 13 males) of childhood MM of the skin below 20 years of age born in Sweden between 1973-1992 and registered in the Swedish Cancer Register and the Swedish Medical Birth Register. The median age at diagnosis was 15 years (range 7-18 years). One hundred and twenty controls, i.e. four for each case, were selected, with the same date of birth, at the same hospital and of the same gender. The cases and controls were checked for birth diagnoses registered in the Swedish Medical Birth Register.

The records of the cases and controls diagnosed partus normalis or with non-PT-related diagnoses were not checked, while all the other records were checked for PT.

Paper III Questionnaire

Between 1973 and 1993, 3 922 infants were registered in the SMBR with congenital nevi. We made a sample of one in twenty i.e. 192 persons. For seven persons we could not trace registration in the Swedish population register (SEMA group) because they had left the country, or for other reasons. This left us with 185 cases.

To reduce the role of confounding factors especially concerning mothers smoking during pregnancy, we checked the SMBR registration of smoking mothers and all newborn children registered between 1983 and 1997 with the diagnosis nevi.

A questionnaire was posted to 185 persons registered in the SMBR as CN. We got answers from 150 subjects (81.1%). 83 women (55%) and 67 men (45%). We used the answers as additional information and as a quality test of the SMBR. The questionnaire had eleven questions with up to five part questions. Four of the questions were related to visual analogue scales.

The questionnaire sought additional information about gender and age, and when, where, how and by whom any surgical removals were made; also whether the respondent still had some CN left. If so, the location, size, colour and structure of the skin

lesion were elicited. We further asked whether the skin lesion had caused any social or physical discomfort.

We asked whether any close relatives of respondents had or had had CN; and also whether the CN had changed their behaviour with regard to sun exposure. Further, we asked whether the scars or remaining CN caused any social discomfort which reduced possibilities for normal activities.

Two questions concerning the mother were whether she had smoked or had had any infections or illnesses during the pregnancy, or had any known present illness or disease.

The CN were classified into three groups: definite, probable and not-probable, for judgement. In the questionnaire we used excision with pathohistological diagnosis as a criterion for the group ”definite CN”. “Probable CN” due to the clinical setting but without biopsy, in contrast to laser treatment, gave ”not-probable CN”. In Sweden we do not treat nevi with lasers due to the risk of a diagnosis of pseudomelanoma in the treated area when doing histopathological controls. The classification was further based on the shape, hair growth, colour, location and size of the CN. In some cases the patient reported a diagnosis ”not a CN”, i.e. haemangioma or café-au-lait spots.

Quality test

The medium age of the patients in the quality test was 14 years (range 5-27 years).

Of 150 patients 78.6% had definite CN, 6.7% had probable CN and 14.7% had probably-not CN; (haemangioma, café-au-lait spots and other lesions). Definite and probable CN thus made up 85.3% of the material, which we defined as true CN in the quality test.

Paper IV Congenital nevi

Among 2 198 619 infants born between 1973 and 1993, 3 922 were registered in the SMBR with CN. We traced the patients in the Swedish Cancer Register for the years 1973-1993, and all cancer was analysed. Most paediatricians had entered the diagnosis in the SMBR. Coding of cause of death, cancer and nevi registration in newborns was according to the revision of the International Classification of Diseases (ICD) in force at the time of registration; ICD 8 for 1968 to 1986, and ICD 9 for 1987 onwards. In ICD 8 the number 757,10 represented large CMN and 757,19 represented CMN. In ICD 9 all are registered with the number 216.

Paper V Genetics

We found 400 patients in the Swedish Cancer Register (SCR), between 1958 and 1998 with a diagnosis of MM before the age of 20 years. We selected those who were still

alive, diagnosed after 1979 and able to come to Stockholm or Göteborg for clinical examination and blood sampling.

This yielded 98 patients, who received a letter inviting them to participate in the study. The investigation was approved by the Research Ethics Committee, Karolinska Institutet. Sixty-nine patients were interested in participation, but for practical reasons such as long travel distances and lack of time, only 60 patients were enrolled. The histopathology of the diagnostic slides was reviewed by two pathologists, and nine was found to be no MM. This left us with 51 patients.

The patients filled in a questionnaire, an oral history was taken, and the skin was examined physically by two dermatologists and a venous blood sample was obtained for the genetic analysis.

In the physical examination we registered the following parameters; gender, age, skin-type, colour of hair and eyes, count of DN, location of MM, presence of freckles.

A specialist is a man, who knows more and more about less and less,
and when he has reached perfection he knows everything of nothing.
(William Mayo deceased 1911, American surgeon)

7. RESULTS

Paper I Epidemiology

In a retrospective study from 1958 to 1992 we found 287 cases of malignant melanoma in the Swedish Cancer Register in patients younger than 20 years. For further measures we used data from death certificates and cancer reports. We noted a stable incidence up to puberty where the increase started one year earlier in girls aged 15 years than in boys aged 16 years (Figure 1).

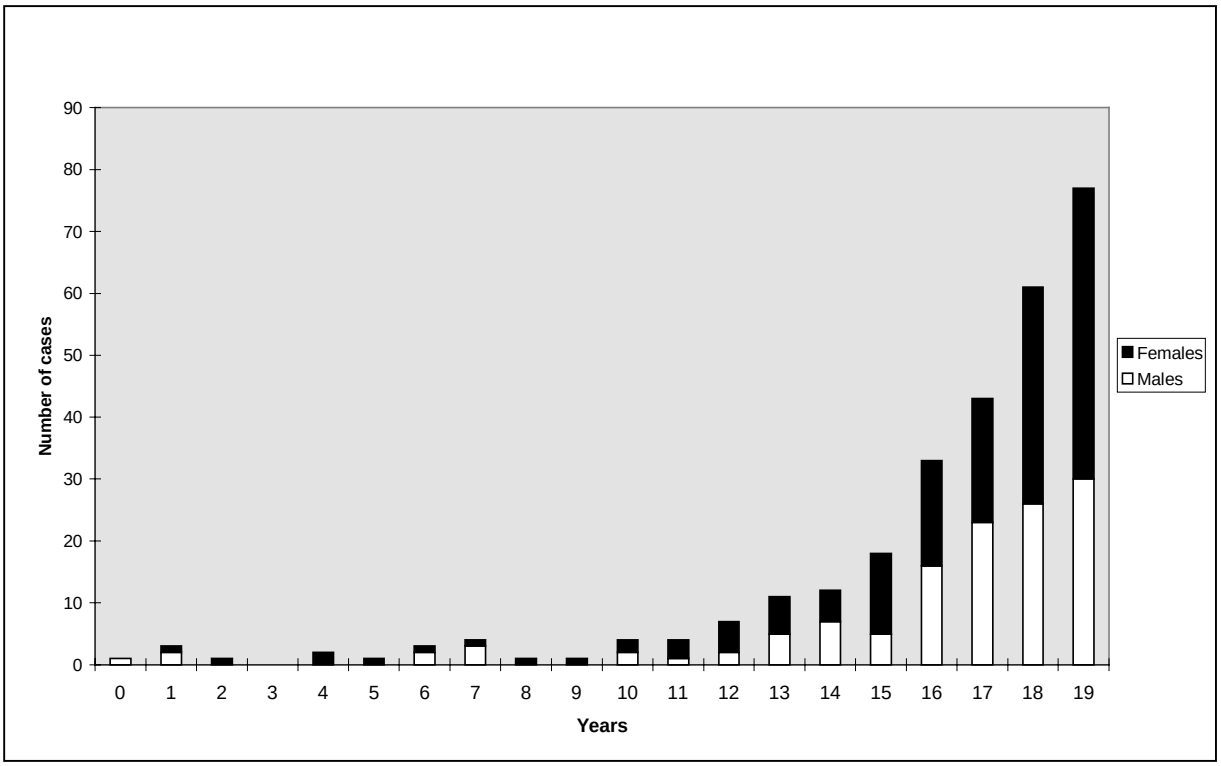

Figure 1. Childhood malignant melanomas in males and females by age (n=287)

As in adults MM is also more common in females than males (162/125). Without selection due to reviewed histopathology we found a doubling of cases during the past ten years compared with the previous ten-year period (1973-82/ 1983-92). The incidence of pre-pubertal MM was low and more or less constant during these 35 years with about 1-3 cases per year.(Figure 2).

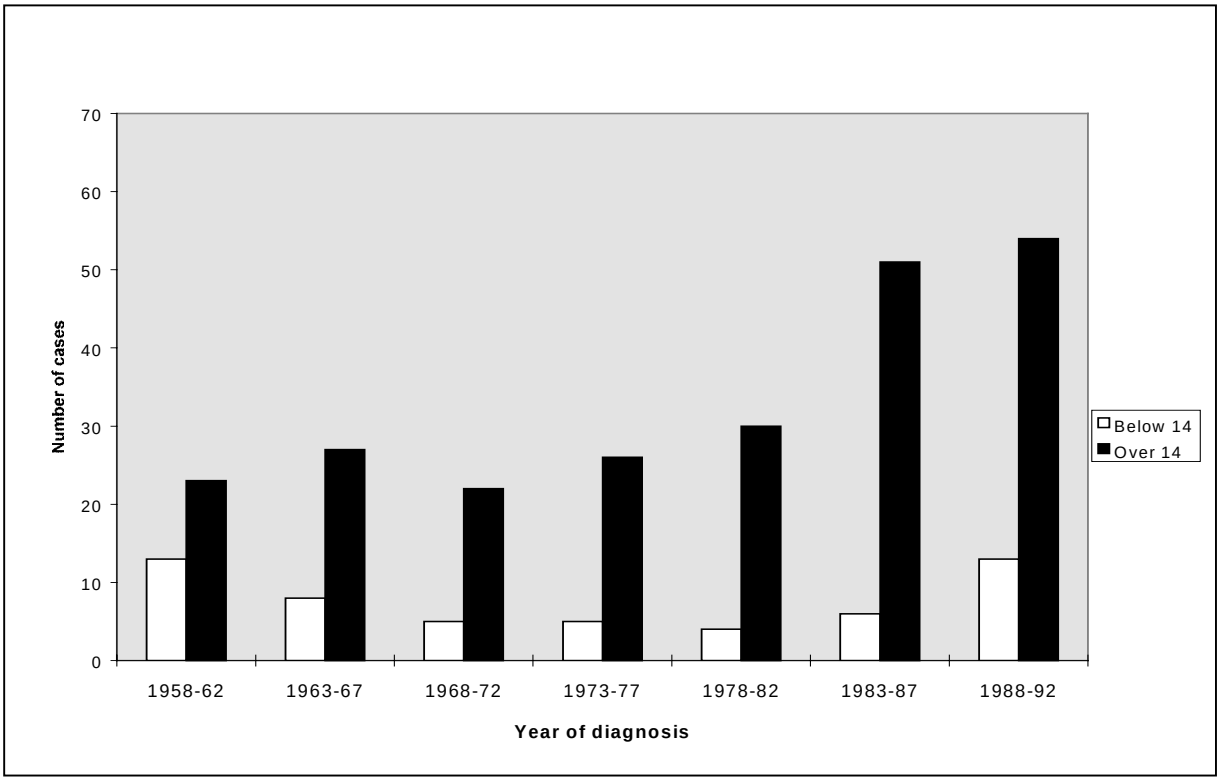

Figure 2. Numbers of childhood malignant melanomas in Sweden 1958-1992. Distribution with respect to puberty (n=287)

We found no difference concerning the anatomical distribution between early-onset MM and adult. In distribution, there were significant differences between females with more MM on upper and lower limbs, and males with more on the trunk. Median survival time was three years after diagnosis and 15.3% patients died as a result of the diagnosed condition, most around 20 years of age (Figure 3).

No specific location was over-represented as fatal compared with the total distribution and no higher risk was found for palms, genitals or plantar areas as locations. As in adults there was a slightly higher fatal course in males than in females. Of interest is that no patients younger than 10 years were reported after 1983. The material is too small for identifying any specific geographical area as over-represented in either incidence or mortality.

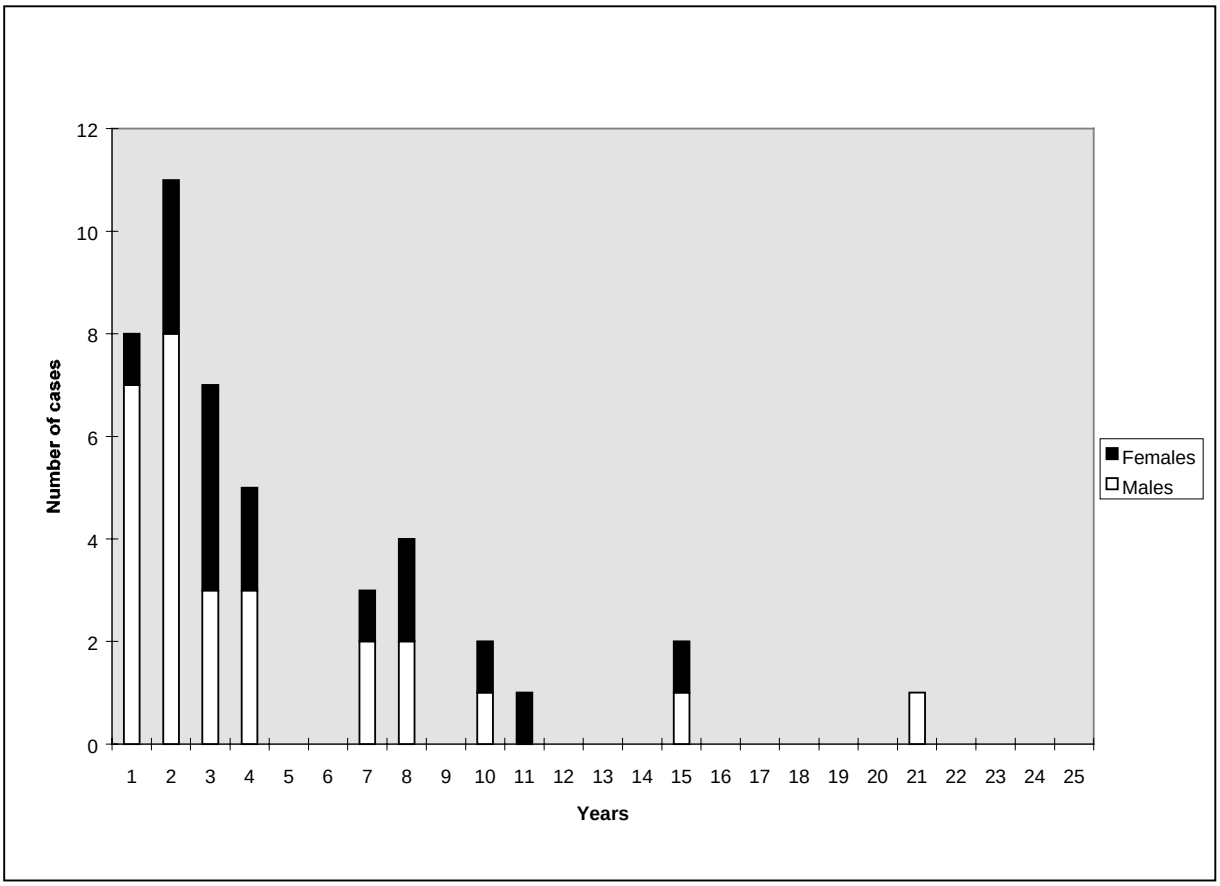

Figure 3. Survival time for fatal childhood malignant melanomas (n=44)

Paper II Phototherapy

Paper II was a retrospective study from 1973 – when the SMBR started – in which we included all 30 cases of MM of the skin in patients younger than 20 years. We found 17 females and 13 males with early-onset MM in the SCR and the SMBR, median age at diagnosis being 15 years. One hundred and twenty controls, i.e. four for each case, were selected. They had the same date of birth, were born at the same hospital and were of the same sex as the cases. The cases and controls were checked for birth diagnoses registered in the SMBR. Of the 30 cases with MM, two had been diagnosed with HB but none had undergone PT. Of the 120 controls, 11 had undergone PT. The average follow-up time was only 18 years. The difference was not statistically significant. This preliminary report showed no significant risk of developing MM after PT of the skin in neonates with HB.

Paper III Questionnaire

The medium age of the patients when filling in the questionnaire reported in paper III was 14 years (range 5-27 years). We classified the 150 patients from the information in the questionnaire as having; definite CN (78.6%), probable CN (6.7%), or probably-not CN (14.7%); (haemangioma, café-au-lait spots and other lesions that had disap-

peared). Definite and probable CN thus made up 85.3% of the material, which we defined as true CN in the quality test.

Fifty-one patients had undergone one or more operations (dermabrasion or excision). Thus the removal rate was 39.8% related to the true CN. Ten of these had large CN and 41 had small CN. In the group with definite CN the excision frequency was 51/118 i.e. 43.2%. In the group with probable CN the excision frequency was 0/10. In the group with not-probable CN the excision frequency was 0/22, but 33% of the haemangioma had been laser-treated.

The medium age at surgery was 9.7 years (0-25). This concerned the first excision since some patients underwent surgery many times. Almost 90% of the excisions were done by plastic surgeons or general surgeons, 5% by dermatologists and the rest by other doctors. The larger the CN, the more common and earlier the excision was.

Mother's influence

Of the mothers of those excised, (all defined as definite CN,) 39.2% had smoked during pregnancy, but in the total group with definite CN only 25.4% of the mothers were smokers; and in the group with probably-not CN, 18% were smokers. However, the SMBR statistical control of the sample with all living newborn children with nevi during 1983-1997 showed no significance concerning smoking mothers. Sixteen of the mothers had had infections during pregnancy and 21 had a current one, but no specific infection/illness was noted.

Characterisation and distribution

CN were most common on the trunk, the legs, the head, the arms and the neck, in that order. The head, however, was over-represented. Twenty-nine percent of the patients reported CN had hair growth on their nevi.

Social behaviour and discomfort

As many as 30% said they 'were very careful' with sun exposure and 25% said they were careful. In total 8% of all cases with true CN disliked social reactions to their skin lesions i.e. felt they were targets for bullying. This affected their social activity, which was reduced. We were unable to evaluate reported CN in relatives because it was unclear whether they were true CN.

Paper IV Congenital nevi

The results concerning the numbers of CN in the cohort are summarized in (Figure 4) and (Figure 5).

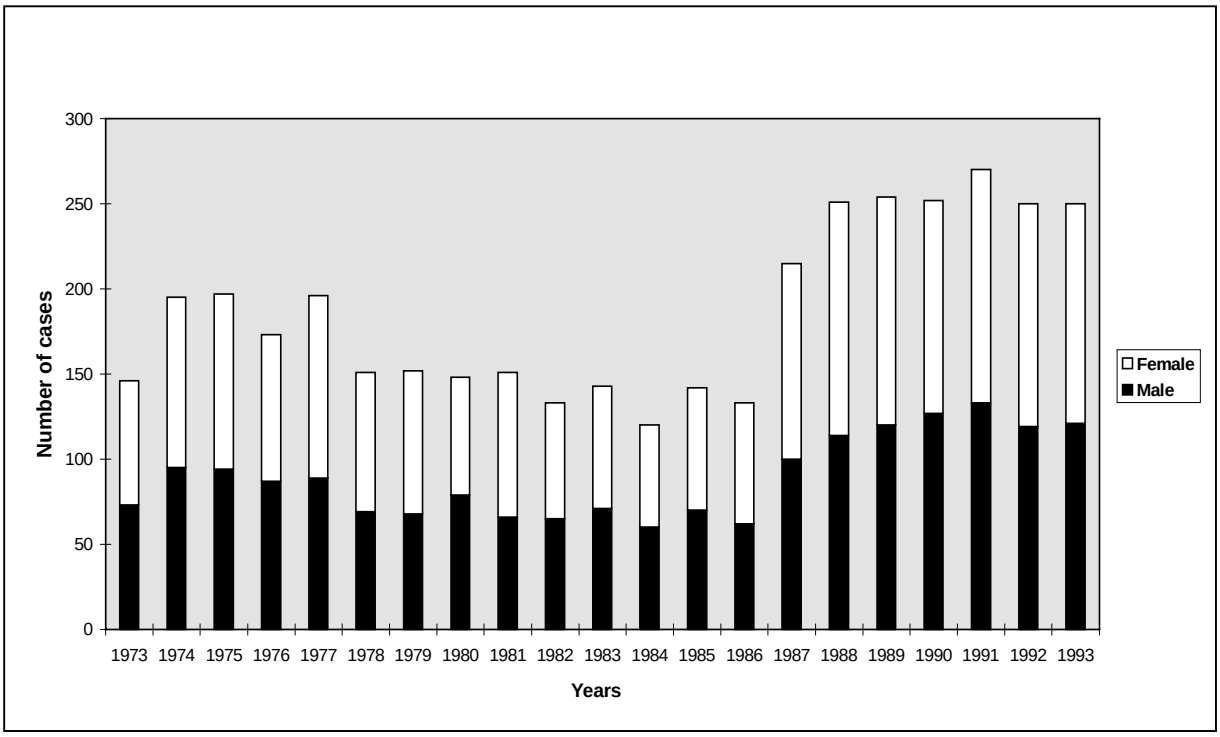

Figure 4. Congenital Melanocytic Nevi, all sizes (n=3922). Reported in the Swedish Medical Birth Register during the years 1973-1993

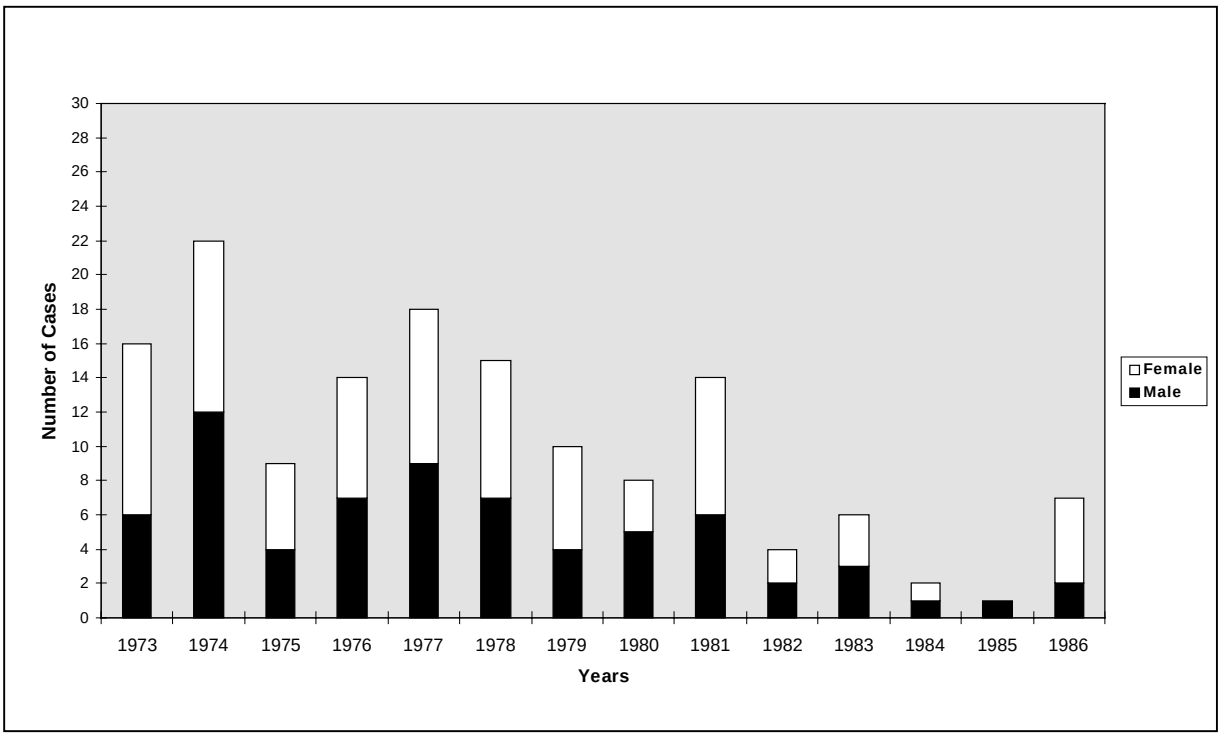

Figure 5. Large congenital melanocytic nevi (n=146) Reported in the the Swedish Medical Birth Register during the years 1973-1986

The incidence varied between 127 and 223/100 000, with a mean of 204/100 000 during the years 1973-1993. This represents a 0.2% risk for newborns to present with CN. The numbers of CN in newborns by mother's age group did not vary. Between 1973 and 1986, 7% of all CN in newborns were large (146 cases) and 93% small. Median follow-up time was 10 years (0-21), while 1 058 cases had been followed-up for 15 years or more. We found two cases regarded as MM in the cohort of 3 922 patients with CN at birth. Four other patients had non- melanoma cancer while two were associated with cancer in neurological tissue.

Case reports, case I

At birth this male child had a dark brown pea-sized lesion above the right ear. In his 16th month a 3x3 cm pigmented infiltration was found near the ear and a 1x1 cm CN in the neck. The pigmentation in the neck showed histologically an intradermal nevus. The pigmented lesion near the ear was first diagnosed histologically as a Spitz nevus, but after re-excision one month later the lesion was diagnosed as a MM with local lymph node metastasis. The MM continued to grow and at two years the boy died of malignant melanoma. In the reviewed histology, a MM with mostly intradermal nodular proliferating foci was found. There was focal ulceration and fibrosis. Clark level was III/IV. Mitotic frequency was more than 10/10 HPF, metastasing.

Case reports, case II

At birth this male child had a large CN involving the whole right arm, parts of the right upper leg and the head and left shoulder. Dermabrasion was carried out twice in the neonatal period and a small excision was made at the age of one year. At seven, three excisions were made, one of them in the right arm histologically diagnosed as a minimal-deviation melanoma. Today at the age of nineteen the boy is healthy and is quite content with the treatment results from a cosmetic point of view. With the reviewed histology, in consensus with the pathologist who made the diagnosis in 1989, no melanoma cell was found and the MM is at present regarded as a benign melanocytic, proliferative nodule within a congenital nevus.

Paper V Genetics

Histopathology

Eighteen slides were reviewed by two histopathologists as part of the present study, while twenty-four slides were previously reviewed by histopathologists and published in an earlier clinico-pathological study. For cases that could not be re-reviewed due to lost or destroyed slides, we accepted the diagnosis reported to the SCR. From the original 60 patients nine were excluded due to non-melanoma diagnosis. Five of these being diagnosed as Spitz nevus and four as dysplastic or atypical nevi.

After reviewing the histopathology we classified the patients into the groups shown in table XII.

Table XII. Clinical and histopathological features of malignant melanoma.

	NO of MM
Site	
Head-neck	2
Upper extremities	5
Trunk	22
Lower extremities	22
Histotype	
SSM	38
NM	10
ALM	1
Unclassified MM	2
Level	
I	14
II	20
III	10
IV	7

Patient characteristics

Of the 51 patients, 33 were females and 18 males. The median year of diagnosis was 1987 (range 1980-1998) and the median age at diagnosis was 17 years (range 14-19). Median age on attending the test was 31 years. Skin-type II was the most common (Table XIII). The MM were all located in sun-exposed skin surface areas.

Seventeen of the 51 patients underwent excisions for clinically suspected DN before the diagnosis of MM and 45/51 had had multiple excisions after treatment for MM. On skin examination we found DN in 31 patients (61%), some with large numbers. The clinical examination revealed 101 DN on the trunk, 14 DN on lower extremities, nine DN in the head-neck, and six nevi in upper extremities. Two patients had disease relapse following treatment for primary MM.

In the family history we found four patients with a first-degree relative and two with a second-degree relative with MM heredity. Some patients had other skin diseases; one UV-treated psoriasis, one urticaria pigmentosa, one nevus spilius, one vitiligo and three halo nevi.

Table XIII. Patients characteristics. In total 51 patients with malignant melanoma after histopathological review.

	NO
Gender F/M	33/18
Skin-type	
I	4
II	28
III	18
IV	1
Eye-colour	
Blue	24
Brown	4
Green	10
Mixed	13
Hair-colour	
Blonde	10
Medium-blonde	29
Dark	7
Red	5

DNA sequencing

One female patient with the diagnosis SSM, MM, Clark II at the age of 18 years and self-reported massive melanoma heredity (two first-degree relatives with MM and multiple DN) had a germline mutation in exon 1 of CDKN2A consisting of a mis-sense mutation resulting in a prolin-to-leucin substitution in codon 48. This mutation was known earlier, and has been presented in another study by Platz 1997. Four other patients had a previously-reported alanine-threonine polymorphism in codon 148 of exon 2, which is of no known functional significance.

-Winston, you are drunk!
-Madam, you are ugly.
-Tomorrow I will be sober.
(attr: Winston Churchill)

8. CLINICAL PHOTOGRAPHY

1. Nodular malignant melanoma in a congenital nevus.

2. Congenital nevus in a 14 years old boy .

3. Dermascopy in the same congenital nevus.

4. Histopathology of congenital nevus in the same boy.

5. **Superficial spreading melanoma.**

6. **Large congenital nevus in a child.**

7. **Large congenital nevus after. treatment.**

8. **Histopathology of congenital nevus in a newborn.**

"I have been struck by a sexually transmitted disease.
It lacks cure and always leads to death.
The name of the disease is – life!".
(Sven Delblanc)

9. GENERAL DISCUSSION

Malignant melanomas are rare in children/adolescents younger than 20 years. With up-to-date histopathology techniques, probably even fewer patients will get the false diagnosis MM, so the risk of over-diagnosis and its associated psychosocial problems will be reduced. However, lacking a specific laboratory indicator of malignant melanoma cells, we have to give the patient the benefit of the doubt - it is better to be safe than sorry. Of interest is that a study by Sander et al has shown that especially many of those given this diagnosis in the early 1960 and the 1970 were not MM, i.e. the increase is probably higher than shown in paper I [172].In the review of slides reported in paper V, 15% of the malignant melanomas registered in the Swedish Cancer Register since 1980 were given other diagnoses such as Spitz nevi or dysplastic, atypical nevi.

However, histopathology in this respect is a great international problem since neither MM nor CN have a clear consensus and no 100% specific diagnostic analysis is available. Hence it is always important to review the slides for a new study so as to get the latest histopathological opinion.

Many international studies show that the incidence of MM is still increasing in the general population. We have shown that this is also the case for adolescent MM in Sweden over the past few decades. Many countries, though, report a lower increase in incidence during the past few years and we can believe we are going the same way. So hopefully for many patient groups information on the risks with UV radiation have had an impact. Some recent studies have shown that information received in adolescence has a low effect on sun exposure behaviour[25].

It is thus understandable that we found a doubling of incidence among adolescents between 1972 and 1992.

In one of the Swedish daily newspaper "Dagens Nyheter", January 12, 2003, P.C Jersild wrote: "Sweden has an invaluable natural resource in its well-managed registers. In a few areas Swedish medical research is especially eminent. One is stem cell research. Less is written about the fact that we are even better, really world-leaders, in the research area of clinical epidemiology – disease prevalence in the Swedish population". The science of clinical epidemiology, rightly used, present facts about a health situation and is the most efficient science on which to base strategies for solving health problems. In the interests of personal integrity, permission is required to start a register in Sweden, and the people working on and with it are under an obligation to observe

secrecy. Few countries conduct better and more correct epidemiological research than Sweden.

And yet government interest is lukewarm, and new regulations are continuously hampering research. A view in some official circles is that all registers represent violations of personal integrity. Most people in Sweden have samples of their cells saved in different medical laboratories under a code. To use these for research following agreement from a Research Ethics Committee is by no means an infringement of personal integrity. Many of the new regulations appear to be designed on the "Catch 22" principle; their unnecessary restrictions are making future research in epidemiology unreasonably difficult. This is a great loss for patients and the world's people: the benefits of the research results greatly outweigh any potential risk to integrity.

The epidemiological features of MM point to a complex relationship to sunlight[182, 183].
Many changes in lifestyle may significantly increase the risk of MM. Examples of this are immunosuppressive treatment in young renal transplant patients, or changes in sunbathing behaviour with or without the use of sun-protection agents.

Many studies indicate that risk factors with a strong effect on the development of MM are; skin phototype, solar lentigo, actinic keratosis and chelitis, immediate skin reaction to UV light at the start of the outdoor season, sunburns in childhood and sun exposure during holidays in the sunny areas 20 years before MM was diagnosed. A genetic factor is the existence of MM in a first-degree relative.

However, a study presented in 2001 indicated that normal outdoor activities in childhood, with sparse UV radiation could be a protective factor[30].

Some authors believe that AN are also precursors of malignant transformation. A paper in The Lancet shows that young patients with kidney transplants have a higher frequency of AN, although they are advised not to get suntans. We know today that it is important for patients with immunosuppressive treatment to be very careful with UV exposure and in this patient group we find many more UV-induced skin malignancies such as squamous cell cancer and basaliomas so far not seen in younger patients[184].

Further, the incidence of MM is higher in the higher socioeconomic classes. Thus the latest hypothesis is that intermittent and intense sun exposure of untanned skin is a strong risk factor for MM especially the SSM type[185].

The young persons in study V with MM and those in study III were aware of the risk of exposing their nevi to the sun, which indicates that the patients or their parents were concerned about malignant transformation. Still, daily sun habits were very little changed and completely other factors than malignant transformation were important for whether the persons exposed themselves to UV radiation or sunburn.

One important factor was that questionnaire respondents reported disliking the sun due to the heat or because they had skin type I and experienced mostly irritation without any tan. This agrees many other current studies especially concerning young people's sunbathing[34].

Therefore, future topics for international conferences could be; better sun protection, sunscreens, B-caroten, retinoids. Or even less sun protection – sunbeds – rightly used.

In view of the frequent use of phototherapy (PT) in neonates with HB in immature newborn skin, with lower potential to respond immunologically, we thought it important to examine the possible effects of PT on the increased incidence of MM. During the past few years attempts have been made to re-run the study in Norway, Finland and Denmark, which also have very reliable registers. However, despite contacts with university professors in those countries, we have not managed to enlarge the study.

HB diagnosis varies considerably between hospitals, as does the use of phototherapy. Dropout rates concerning specific diagnosis should therefore be interpreted with some caution. The use of phototherapy has increased tremendously, between 5% and 10% of newborns being treated; but this more common use of phototherapy has not had a full impact on the present study.

Now, however, a new hypothesis is suggested by the lack of PT in early-onset MM; could PT be a preventive factor for MM? Could early PT stimulate the immune system? Further studies are needed to answer these questions, especially after the recent interesting reports in other studies of the protective effect of normal outdoor activities in childhood[30, 40].

Congenital nevi are defects of development present at birth. Nevi appearing later should not be considered congenital even following a premature birth.

In the absence of exact histopathological criteria for CN, nevi developing within the first months of life should be regarded as tardive nevi. Why CN arise during fetal life is not clear, but genetic or infectious factors may play a role, although we have not been able to prove this. CN may arise because of untoward events during pregnancy, such as influenza-like symptoms. Another possible cause could be the presence of a c-met proto-oncogene product in CN in children with neurocutaneous melanosis[143].

It is of interest to note that only 8% of the respondents to our questionnaire concerning CN and removal reported that their nevi or scars caused them social discomfort or reduced their activities. This seems to indicate a good acceptance of the lesions from a cosmetic point of view. At the same time the most probable reason for surgical removal of CN worldwide is cosmetology. Very often it is the relatives or parents that initiate cosmetic treatment[97, 147].

Few up-to-date studies have been made on large CN and no long-term prospective study, which makes it difficult to determine the right level of removal for reducing the

risk of MM to a reasonable level. Many large CN are also removed for cosmetic reasons, but there is indirect evidence that a certain removal rate of CN may be relevant to decreased MM development[69, 135].

In large congenital melanocytic nevi, the total cell burden probable enhances the risk of malignant transformation therefore, large CN become tumours early in life. The induction time for MM in general, or specifically in small CN, is unknown; but it is argued that the risk of tumours in small CN rises with time.

Of the two registered cases of MM in CN in our study, one was a de novo MM since birth, which metastasised and killed the patient, the other was diagnosed as a benign melanocytic proliferative nodule. Proliferative nodules were earlier commonly diagnosed histopathologically as MM. Nowadays it is possible in difficult cases to use comparative genomic hybridisation; in CN there is no chromosomal disturbance, but MM shows chromosomal fractioning[141].

The exact incidence of CN is difficult to establish due to underreporting, lack of consensus concerning clinical diagnosis and more frequent premature births. The controversy over CN strongly suggests that many studies include selection biases.

The pattern of heredity in FMM is consistent with an autosomal dominant inheritance with incomplete penetrance. Germline mutations in the CDKN2A gene on chromosome 9p21 have been identified in a proportion of FMM kindreds, and mutations in the CDK4 have been documented in a few families[186, 187].

FMM is a genetically heterogeneous condition, since a large proportion of FMM kindreds lack germline mutations in CDKN2A or CDK4. A linkage study has implicated another FMM locus on chromosome 1p36, but other investigators have not so far confirmed this finding[3].

Different errors in the five studies need to be considered here. The Swedish registers are very good, many quality tests showing a low dropout rate. For MM the registration frequency is close to 100%. One problem with the medical registers in Sweden is that they have not been in existence for long enough! The SCR started in 1958 and the SMBR in 1973. The relatively short follow-up period makes strict conclusions difficult, yet no other country, except some of our neighbours, is able to draw more reliable statistical conclusions with such a large material.

Another problem encountered in the present work was that most CN diagnoses are entered in the SMBR by paediatricians. Probably, however, the present quality testing has eliminated vascular lesions etc. The increasingly common problems associated with premature birth have prompted discussion as to whether tardive nevi should be included. The only sound way to answer this question is to surgically remove all suspected tardive nevi with the clinical appearance of a CN.

A further important problem that needs a closer look is the induction time for both MM and CN; again, young people are of the greatest interest. The etiologies of MM before puberty and after probably differ, as witness the increased incidence after puberty. The increased rate of removal of CN does not seem to have reduced MM incidence either before puberty or after; in the latter case, rather the reverse.

Lastly, MM in particular has long been over-diagnosed – understandably since missing a MM is a catastrophe for the patient. Surprisingly few patients in the USA have sued their doctors for surgically disabling treatment for diagnoses of suspected MM, later proved to be non MM. Especially important is how to deal with adolescents' attitudes to UV radiation, with the obvious increase we still find in this group. Another need is further genetic studies, since in the large majority of MM in childhood/ adolescence underlying genetic alterations remain to be identified.

"This is not the end.
It is not even the beginning of the end.
But it is perhaps the end of the beginning".
(Winston Churchill)

10. SUMMARY AND CONCLUSIONS

In the light of the rapidly increasing incidence of malignant melanoma, even in adolescence, it is important that knowledge of risk factors for its development, and its early clinical characteristics, be spread to not only the medical profession but also to the general public.

We found no signs of increased incidence of MM in childhood, and this could probably constitute a subgroup not induced by UV radiation among other MM subgroups. In adolescents most of the MM were of SSM type, which is suspected to be sun-induced. Here we also reported a doubling during the past few decades. We found no difference concerning the anatomical distribution between early onset MM and adult. Median survival time was three years after diagnosis and 15.3% patients died as a result of the diagnosed condition, most around 20 years of age.

No specific anatomical location was over-represented as fatal.

Phototherapy of newborns has probably been ruled out as a factor in the increased incidence of MM; on the contrary it is possible that phototherapy could be a protective factor just as outdoor activities in childhood are thought to be. Due to the even more common use of phototherapy today – 5-10% of all newborns – it would be of value to repeat the study with a longer follow-up period than the 18 years.

In the CN study – the largest so far, with nearly 4000 cases of CN – an overall incidence of 0.2% was found. One can always question this as being due to underreporting; but quality studies show that some other diagnoses such as haemangioma is also sometimes regarded by non-specialists as congenital nevi.

With a removal rate of 40% of CN no MM developed, so perhaps the lesions with the highest risk had been removed. However, there is no direct evidence that MM is actually prevented by the excision of CN. The larger the nevi the more common surgical removal is, as well as surgical techniques such as dermabrasion which do not remove all melanocytic cells. The medium age at surgery was 9.7 years (0-25). Almost 90% of the excisions were done by plastic surgeons or general surgeons. One problem with this way of dealing with nevi is that many patients never obtain a dermatologist's opinion. The absolute highest-density location of CN was the head, just as in the case of MM.

The CN seem to have had limited effects on social life, but had resulted in greater caution with regard to sun exposure. Many authors consider that without additional basic data, any recommendation to remove all CN to decrease the incidence of MM is premature.

After histopathological review we found no cases of MM in nearly 4000 CN of which at least 146 could be regarded as large. We have also up-to date histopathological techniques. The great variation in estimated raw risk of malignant transformation (10.7-0.0 %) in ten different studies of large CN, is so great that one can presume that many of them includes selection biases.

We found no clear association between CN and maternal illness/infection during pregnancy.

Congenital nevi, or treatment of CN, seem to play no part in the increased incidence of MM below the age of 20 years.

In the present investigation we have shown that among patients with MM diagnosed during childhood/adolescence, CDKN2A germline mutations are rare, although ten percent of patients reported a familial history of relatives with MM. It is therefore likely that there are other genetic factors of importance for the development of childhood/adolescence MM.

I believe Sweden is in an excellent position for epidemiological studies thanks to our personal identification number-system and the national registers. Still it is important that new regulations do not impede the research they allow.

In summary, it seems likely that many different etiological factors affect the development of MM. As yet, however, we have found no solution for how to stop the disease. Further etiological and genetic research is necessary before the subgroups of MM can be differentiated.

-Eders majestät, tack för den fina utnämningen till ärkebiskop.
-Tacka fan för att du skulle bli ärkebiskop.
-Då får jag tacka båda herrarna.
(Natan Söderblom vid audiensen hos konung Gustav V, 1914)

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My greatest thanks to you all!

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There is no such thing as skin disease.
(E.Bazin 1855).

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