

The Molecular Pathology of Melanoma: An Integrated Taxonomy of Melanocytic Neoplasia

Boris C. Bastian

Departments of Dermatology and Pathology, Cardiovascular Research Institute, University of California, San Francisco, California 94158-9001; email: boris.bastian@ucsf.edu

Annu. Rev. Pathol. Mech. Dis. 2014;9:239–71

The *Annual Review of Pathology: Mechanisms of Disease* is online at pathol.annualreviews.org

This article's doi:
10.1146/annurev-pathol-012513-104658

Copyright © 2014 by Annual Reviews.
All rights reserved

Keywords

genetics, pathogenesis, classification, mutation, nevi

Abstract

Melanomas comprise multiple biologically distinct categories, which differ in cell of origin, age of onset, clinical and histologic presentation, pattern of metastasis, ethnic distribution, causative role of UV radiation, predisposing germ-line alterations, mutational processes, and patterns of somatic mutations. Neoplasms are initiated by gain-of-function mutations in one of several primary oncogenes, which typically lead to benign melanocytic nevi with characteristic histologic features. The progression of nevi is restrained by multiple tumor-suppressive mechanisms. Secondary genetic alterations override these barriers and promote intermediate or overtly malignant tumors along distinct progression trajectories. The current knowledge about the pathogenesis and clinical, histologic, and genetic features of primary melanocytic neoplasms is reviewed and integrated into a taxonomic framework.

CLASSIFICATION OF MELANOCYTIC NEOPLASMS

CNS: central nervous system

WHO: World Health Organization

SSM: superficial spreading melanoma

LMM: lentigo maligna melanoma

NM: nodular melanoma

ALM: acral lentiginous melanoma

Melanocytes can give rise to a diverse set of neoplasms with varying anatomic distribution, clinical features, histopathological appearance, and biologic behavior. Although melanocytes are most abundant in the skin, where they play a critical role in skin pigmentation and sun protection, they are present in other locations throughout the body, where they have additional, less well understood functions. Melanocytic neoplasms most frequently arise from melanocytes in the skin, but they can also arise from autochthonous melanocytes from numerous internal organs, including the central nervous system (CNS).

As a rule, benign neoplasms of melanocytic lineage are termed melanocytic nevi (the plural of nevus), and malignant ones are termed melanomas; the often-used term malignant melanoma is thus a tautology. Although melanomas can arise from nevi, as can be inferred from the presence of an adjacent nevus remnant contiguous with a melanoma, most primary melanomas do not show an associated precursor nevus. In some cases this is because the precursor nevus was overgrown by the melanoma during its progression, but in many instances there likely was no detectable benign precursor state.

It has long been noted that melanoma comprises different subtypes with varying anatomic location and pathogenesis. The current World Health Organization (WHO) classification (1) is based on the melanoma classification proposed by Clark and colleagues (2) nearly 30 years ago, which uses morphologic aspects of the early (radial) growth phase and the body site of the primary melanoma to distinguish four main types: superficial spreading melanoma (SSM), lentigo maligna melanoma (LMM), nodular melanoma (NM), and acral lentiginous melanoma (ALM). The system captures archetypical patterns of clinical and histological presentation; however, a portion of melanomas cannot be unequivocally placed in any of the categories (3), and the system's im-

pact on clinical care, in particular that of advanced disease, is limited.

Over the past two decades, tremendous progress has been made in uncovering genetic alterations in melanocytic neoplasia, and an expanding panel of recurrent driver mutations that activate specific oncogenes or inactivate tumor-suppressor genes is emerging. Remarkably, many of these mutations are found in association with specific clinical or histopathological subsets of lesions, a finding that strongly supports the notion of biologically distinct types of melanocytic neoplasms. This is true for melanomas as well as for melanocytic nevi, which express a similar diversity of clinical appearance and histomorphology, age of onset, anatomic site, and genetic alterations.

A desirable classification system separates individual disease states by considering differences in cell of origin, pathogenesis, clinical and histologic aspects, genetic alterations, etc. It can serve as a framework to develop refined primary prevention approaches, objective diagnostic and staging algorithms, and tailored therapeutic strategies. Such a taxonomy differs from current approaches, which rely primarily on one of these dimensions (histology or mutation status), by integrating multiple aspects to capture individual disease subtypes.

The organizing principle for this overview is the current, but still incomplete, clinical and genetic data available in varying detail across the broad phenotypic spectrum of melanomagenesis. The proposed taxa are separated in one dimension. For some taxa, distinct evolutionary stages—ranging from benign, to intermediate, to malignant—are separated in the second dimension (**Figures 1 and 2**). The guiding principles for distinguishing taxa are genetic alterations that arise early during progression; clinical or histologic features of the primary tumor; characteristics of the host, such as age of onset, ethnicity, and skin type; and the role of environmental factors such UV radiation.

Mutations common to all progression steps within a taxon are considered probable initiating oncogenic events. They are gain-of-function mutations in oncogenes

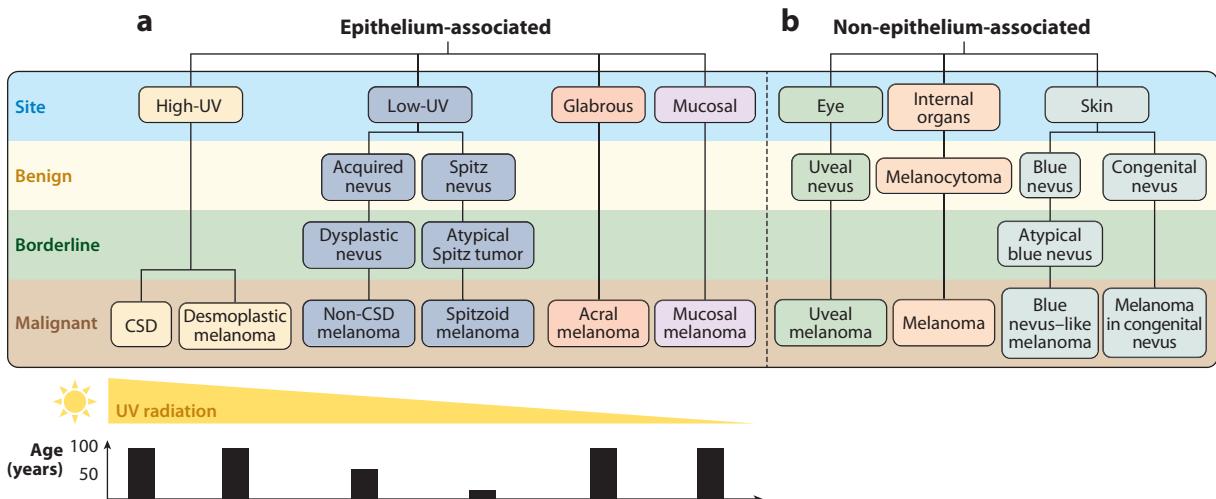


Figure 1

Taxonomy of melanocytic neoplasia. (a) Melanocytic neoplasms arising from epithelium-associated melanocytes. Where applicable, benign or intermediate progression stages are noted. Dysplastic nevus here refers to the histopathological definition as outlined in the text. The different classes have different relationships to UV radiation and different age distributions, shown at the bottom. (b) Melanocytic neoplasms arising from non-epithelium-associated melanocytes. The categories have no relationship to UV radiation, and with the exception of congenital nevus-associated melanomas, which occur primarily in prepubertal children, they have a wide age distribution. Abbreviations: CSD, cumulative sun-induced damage.

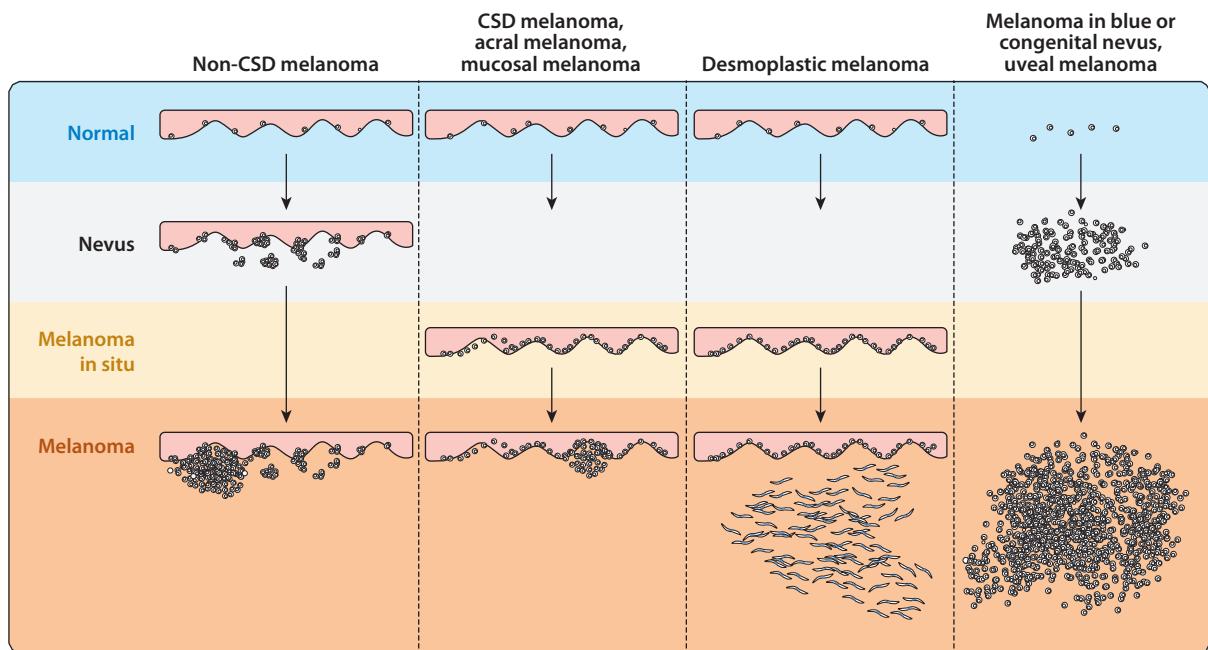


Figure 2

Histologic patterns of archetypical progression patterns for the classes depicted in Figure 1. Abbreviation: CSD, cumulative sun-induced damage.

Table 1 Primary oncogenic events in melanocytic neoplasia

Gene	Type of mutation	Effector pathways	Neoplasms affected
Primary oncogenic driver mutations			
<i>NRAS</i>	Point mutation	MAP kinase, PI3-kinase	Congenital nevi; CSD, non-CSD, acral, and mucosal melanomas
<i>HRAS</i>	Point mutation	MAP kinase, PI3-kinase	Spitz tumors
<i>BRAF</i>	Point mutation, kinase fusions	MAP kinase	Acquired nevi; non-CSD, CSD (V600K), and acral melanomas; Spitz tumors
<i>KIT</i>	Point mutation, amplification	PI3-kinase, MAP kinase, STAT3	Acral, mucosal, and CSD melanomas
<i>GNAQ</i>	Point mutation	Protein kinase C, MAP kinase	Blue nevi; blue nevus-like and uveal melanomas
<i>GNA11</i>	Point mutation	Protein kinase C, MAP kinase	Blue nevi; blue nevus-like and uveal melanomas
<i>ALK</i>	Kinase fusions	MAP kinase, PI3-kinase, STAT3	Spitz tumors and yet-to-be-defined melanoma subtypes
<i>ROS1</i>	Kinase fusions	MAP kinase, PI3-kinase, STAT3	Spitz tumors and yet-to-be-defined melanoma subtypes
<i>RET</i>	Kinase fusions	MAP kinase, PI3-kinase, STAT3	Spitz tumors and yet-to-be-defined melanoma subtypes
<i>NTRK1</i>	Kinase fusions	MAP kinase, PI3-kinase, STAT3	Spitz tumors and yet-to-be-defined melanoma subtypes

Abbreviations: CSD, cumulative sun-induce damage; MAP kinase, mitogen-activated protein kinase; PI3-kinase, phosphoinositide 3-kinase; STAT3, signal transducer and activator of transcription 3.

that tend to occur in a mutually exclusive pattern. The known initiating oncogenic events in melanocytic neoplasia are listed in **Table 1**. By contrast, oncogenic events that mark the transition to the next progression stage within a given taxon are considered secondary (or tertiary) oncogenic events. These are commonly loss-of-function alterations of tumor-suppressor genes such as *CDKN2A* (cyclin-dependent kinase inhibitor 2A), *TP53* (tumor protein 53), *PTEN* (phosphatase and tensin homolog), and *BAP1* [breast cancer 1 (BRCA1)-associated protein 1], some of which are also encountered as germ-line alterations predisposing to syndromes with various types of melanocytic neoplasms (**Table 2**). Somatic alterations considered progression events are listed in **Table 3**. As a consequence, initiating oncogenic events are most valuable for separating classes, whereas later events serve to separate progression steps within classes (**Figure 3**). Primary and secondary oncogenic events do not necessarily have a one-to-one relationship either to taxa or to the progression

steps within a taxon. Some classes may have identical primary oncogenic events and still deserve to be separated, if there are systematic differences in other relevant parameters. For example, both acral melanomas and cumulative sun-induced damage (CSD) melanomas can have *KIT* mutations, but they differ sufficiently in other parameters (e.g., type of genomic instability, pathogenetic role of UV radiation) to be classified separately. Similarly, stage-defining secondary oncogenic events may be shared among several classes.

Table 4 summarizes the proposed taxa, and the following paragraphs detail the basis for their distinction. Although remarkable progress has been made in bringing to light critical genetic alterations, the process is far from complete. This review attempts to capture the salient aspects of insight into the molecular pathology of melanocytic neoplasia and to present an organizational taxonomic framework that can be continually filled in and refined as additional defining features continue to emerge.

CSD: cumulative sun-induced damage

Table 2 Known germ-line mutations associated with melanocytic neoplasia

Gene (OMIM number)	Function	Melanocytic lesions	Other disease or trait associations
<i>CDKN2A</i> (600160)	Encodes two separate proteins: p16 is an inhibitor of cyclin-dependent kinases 4 and 6, and p14 is an inhibitor of MDM2, a ubiquitinase that degrades p53	Melanoma, dysplastic nevi	Pancreatic cancer
<i>CKD4</i> (609048)	Active at the G1-S transition; phosphorylates RB	Melanoma	—
<i>BAP1</i> (614327)	Deubiquitinase involved in chromatin modification	Uveal and cutaneous melanoma, atypical Spitz tumors	Mesothelioma, meningioma
<i>PRKARIA</i> (188830)	Regulatory subunit of protein kinase A	Lentigines, blue nevi	Carney complex
<i>PTPN11</i> (602216)	Protein tyrosine phosphatases that can activate the MAP kinase pathway	—	LEOPARD syndrome, Noonan syndrome, juvenile myelomonocytic leukemia, metachondromatosis
<i>STK11</i> (602216)	Serine/threonine protein kinase that controls the activity of AMP-activated protein kinase family members	Melanocytic macules of the lips, buccal mucosa, and digits	Peutz-Jeghers syndrome
<i>TERT</i> (615134)	Catalytic subunit of telomerase	Melanoma	Other cancers
<i>PTEN</i> (601728)	Lipid phosphatase opposing the action of PI3-kinase	Pigmented lesions of the genitalia	Bannayan-Riley-Ruvalcaba syndrome, Cowden syndrome
<i>XPA</i> (278700), <i>XPB</i> (610651), <i>XPC</i> (278720), <i>XPD</i> (278730), <i>XPE</i> (278740), <i>XPF</i> (133520), <i>XPG</i> (133530), <i>XPV</i> (278750)	Nucleotide-excision DNA repair	Melanoma, lentigines	Xeroderma pigmentosum
<i>WRN</i> (277700)	DNA helicase	Acral and mucosal melanoma	Werner syndrome
<i>MITF</i> (614456)	Helix-loop-helix leucine zipper protein important for melanocyte development and differentiation	Melanoma	Renal cell cancer
<i>MC1R</i> (613099)	Melanocortin 1 receptor, binds melanocyte stimulatory hormone	Freckles, melanoma	Red hair

Abbreviations: LEOPARD syndrome, lentigines, ECG conduction abnormalities, ocular hypertelorism, pulmonic stenosis, abnormal genitalia, retardation of growth, and sensorineural deafness syndrome; MAP kinase, mitogen-activated protein kinase; PI3-kinase, phosphoinositide 3-kinase.

MELANOCYTIC NEOPLASMS ORIGINATING FROM EPITHELIAL MELANOCYTES

Most melanocytic neoplasms show an intraepithelial component; that is, neoplastic

melanocytes are present within the epithelium. Although this feature does not necessarily allow the conclusion that the cell of origin of these neoplasms resides within the epithelium, the high mutational burden and UV signature in

Table 3 Secondary oncogenic events in melanocytic neoplasia

Gene	Type of mutation	Effector pathways	Neoplasms affected
Secondary oncogenic driver mutations			
Gain-of-function alterations			
<i>CCND1</i>	Amplification	RB pathway	Acral, mucosal, and CSD melanomas
<i>CDK4</i>	Amplification	RB pathway	Mucosal and acral melanomas
<i>MITF</i>	Amplification	Upregulation of transcriptional targets including MET, BCL-2, CDK2	Acquired nevi; non-CSD, CSD (V600K), and acral melanomas; Spitz tumors
<i>TERT</i>	Promoter mutation, amplification	Telomere elongation	Subtype variation for mutations TBD; amplification in acral melanomas
<i>CDK4</i>	Amplification	RB pathway	Mucosal and acral melanomas
<i>MEK1, MEK2</i>	Mutation	MAP kinase	Subtype spectrum TBD
<i>MITF</i>	Amplification	Upregulation of transcriptional targets including MET, BCL-2, CDK2	Acquired nevi; non-CSD, CSD (V600K), and acral melanomas; Spitz tumors
<i>CTNNB1</i>	Mutation	WNT signaling	Subtype spectrum TBD
<i>EZH2</i>	Mutation	Chromatin remodeling	Subtype spectrum TBD
<i>RAC1</i>	Point mutation	Adhesion, migration, invasion	Subtype spectrum TBD
Loss-of-function alterations			
<i>CDKN2A</i>	Deletion, mutation	RB pathway (via p16), p53 pathway (via p14ARF)	All melanoma types, dysplastic nevi, and atypical Spitz tumors
<i>PTEN</i>	Deletion, mutation	PI3-kinase	non-CSD, acral, and mucosal melanomas
<i>BAP1</i>	Deletion, mutation	Chromatin remodeling	Uveal melanoma
<i>TP53</i>	Mutation	p53 pathway	CSD melanomas
<i>ARID1A, ARID1B, and ARID2</i>	Deletion, mutation	Chromatin remodeling	Subtype spectrum TBD
<i>NFI</i>	Deletion, mutation	MAP kinase pathway	CSD and desmoplastic melanoma
<i>SMARCA4</i>	Deletion, mutation	Chromatin remodeling	Subtype spectrum TBD
<i>BAP1</i>	Deletion, mutation	Chromatin remodeling	Uveal melanoma
<i>ARID2</i>	Deletion, mutation	Chromatin remodeling	Subtype spectrum TBD
Unknown-function alterations			
<i>PPP6C</i>	Mutation	Cell cycle regulation	Subtype spectrum TBD
<i>SF3B1</i>	Mutation	RNA splicing	Uveal melanoma
<i>STK19</i>	Mutation	Serine/threonine protein kinase	Subtype spectrum TBD

Abbreviations: CSD, cumulative sun-induced damage; MAP kinase, mitogen-activated protein kinase; PI3-kinase, phosphoinositide 3-kinase; RB pathway, retinoblastoma pathway; TBD, to be determined; WNT signaling, wingless-type signaling.

melanomas originating from the nonglabrous skin strongly suggest that many originate from epidermal melanocytes. Lesions of intraepithelial origin are distinct from melanocytic neoplasms, which consistently lack an epithelial involvement; these include uveal melanoma and intradermal melanocytic proliferations, which share common mutations in genes encoding

the two G protein α -subunits of the G α q family, GNAQ and GNA11. They are separated here as a distinct clade (**Figure 2**) and discussed at the end of the article. The other clade of epithelium-associated melanocytic neoplasms is divided further into several classes on the basis of anatomic site and degree of sun exposure. Furthermore, melanomas on glabrous skin and

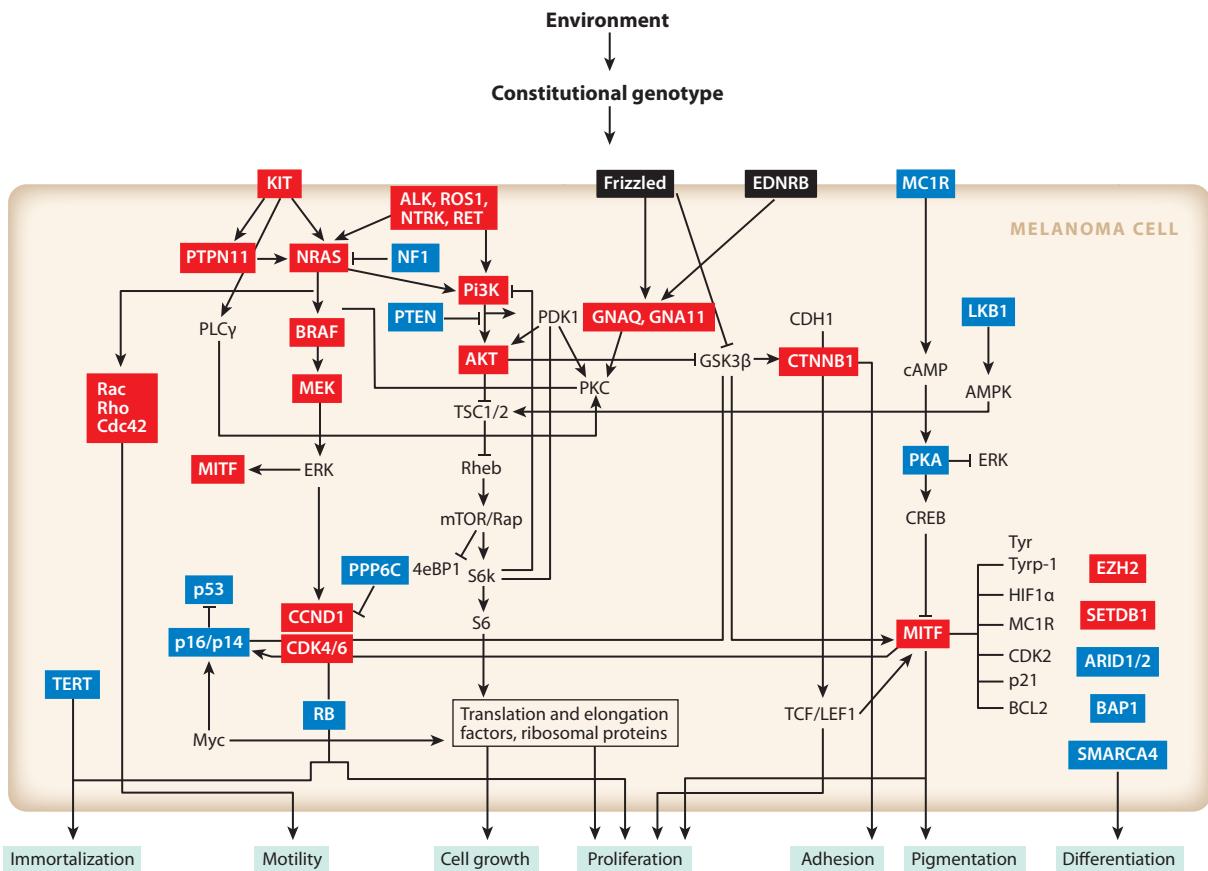


Figure 3

Signaling pathways disrupted by genetic alterations and their relationship to the hallmarks of melanoma. Proteins boxed in red are affected by gain-of-function mutations; those boxed in blue are affected by loss-of-function mutations.

mucosa also have multiple distinctive features and will be discussed separately. These considerations and those detailed below form the background for the classification schema outlined in Figure 1.

Melanocytic Neoplasms on Hair-Bearing Skin

In Caucasians, who have the highest burden of melanocytic neoplasms, the overwhelming majority of lesions present on skin that bears hair follicles and is subject to UV radiation; only a small fraction present on the hairless (glabrous) skin of the palms and soles or on sites covered with mucosa. As discussed below, there are major phenotypic and molecular differences

among melanocytic neoplasms on sun-exposed skin, dependent on the degree of sun exposure and the anatomic site, so these classes are presented separately.

Melanomas on sun-exposed skin without cumulative sun-induced damage. Shortly after the discovery of frequent *BRAF* mutations in melanoma (4), it became apparent that these mutations were unequally distributed across the phenotypic spectrum of melanocytic neoplasms. Mutations were more common in younger patients whose melanomas arose on skin that was sun exposed but not heavily sun damaged (i.e., non-CSD, as evidenced by the absence of marked solar elastosis, an

Table 4 Summary of melanoma classes

Type of melanoma	Non-CSD	CSD	Acral	Mucosal	Uveal	Desmoplastic	Spirzoid	In congenital nevus	Blue nevus-like (130, 142)
Main age distribution	Third to sixth decade	Seventh decade and later	Sixth decade and later	Sixth decade and later	Wide distribution, average age 60 years	Seventh decade and later (143)	Mostly children	Mostly children	All age groups
Anatomic site	Intermittently sun-exposed skin of trunk and extremities (except glabrous sites and nail apparatus); bulbar conjunctiva	Chronically sun-exposed skin of the head, neck, lower arms, and lower legs	Palms, soles, nail apparatus	Any mucosal membrane, but primarily nasal cavity and sinuses, oral cavity, anorectum, vulva and vagina, and tarsal conjunctiva	Posterior and anterior choroid, ciliary body, iris	Mostly head and neck (53%) but also trunk (21%) and extremities (27%) (143)	Mostly head, neck, and lower extremities, but can arise anywhere on the skin, including glabrous skin	Primarily the skin but can arise in all tissues involved by congenital nevi	Most common on scalp, trunk
Typical initial clinical presentation	Irregularly pigmented, asymmetrical patches or nodules; more common in patients with pigmented acquired nevi	Irregularly pigmented, asymmetrical patches or nodules; more common in patients with nonmelanoma skin cancers	Pigmented macule on glabrous skin or nail apparatus followed by nodular growth	Pigmented macule on mucosal skin or nail apparatus followed by nodular growth	Blurred vision and visual field defects	Unpigmented nodule or plaque, often on skin with high cumulative sun exposure; can have associated brown macule as in LMM	Unpigmented rapidly growing nodule	Nodular growth within congenital nevus	Black nodular growth
Typical histologic features	Early lesions composed of enlarged, round melanocytes with dusty melanin, arranged as nests with pagetoid scatter	Pronounced solar elastosis in adjacent skin; early lesions often show lentiginous growth pattern	Early lesions often show extensive lentiginous growth pattern	Early lesions often show extensive lentiginous growth pattern	Solid nodules of spindled or epithelioid melanocytes	Pauctacellular proliferation of atypical spindled melanocytes set in a fibrotic dermis; lentiginous component in 80%, neurotropism in 30–40% (144)	Compound or intradermal proliferation of large atypical epithelioid or spindled cells with ample cytoplasm, with mitotic figures	Nodular aggregates of atypical melanocytes with mitotic figures within a congenital nevus	Dermal nodules of large atypical spindled and/or epithelioid melanocytes with mitoses and areas of necrosis; often adjacent blue nevus of the cellular variant
Pattern of metastasization	Frequent involvement of lymph nodes as first manifestation	Equal proportions of satellite or in-transit, lymph node, and distant metastases (145)	Frequent satellite and in-transit lymph node metastases as first manifestation (145)	Frequent local recurrence, lymph node metastases	Liver and bone	Primarily to the lung; infrequent involvement of lymph nodes (7%) (143)	Frequent involvement of lymph nodes as first manifestation	Not enough data	Lymph nodes, liver, lung, bones (130)

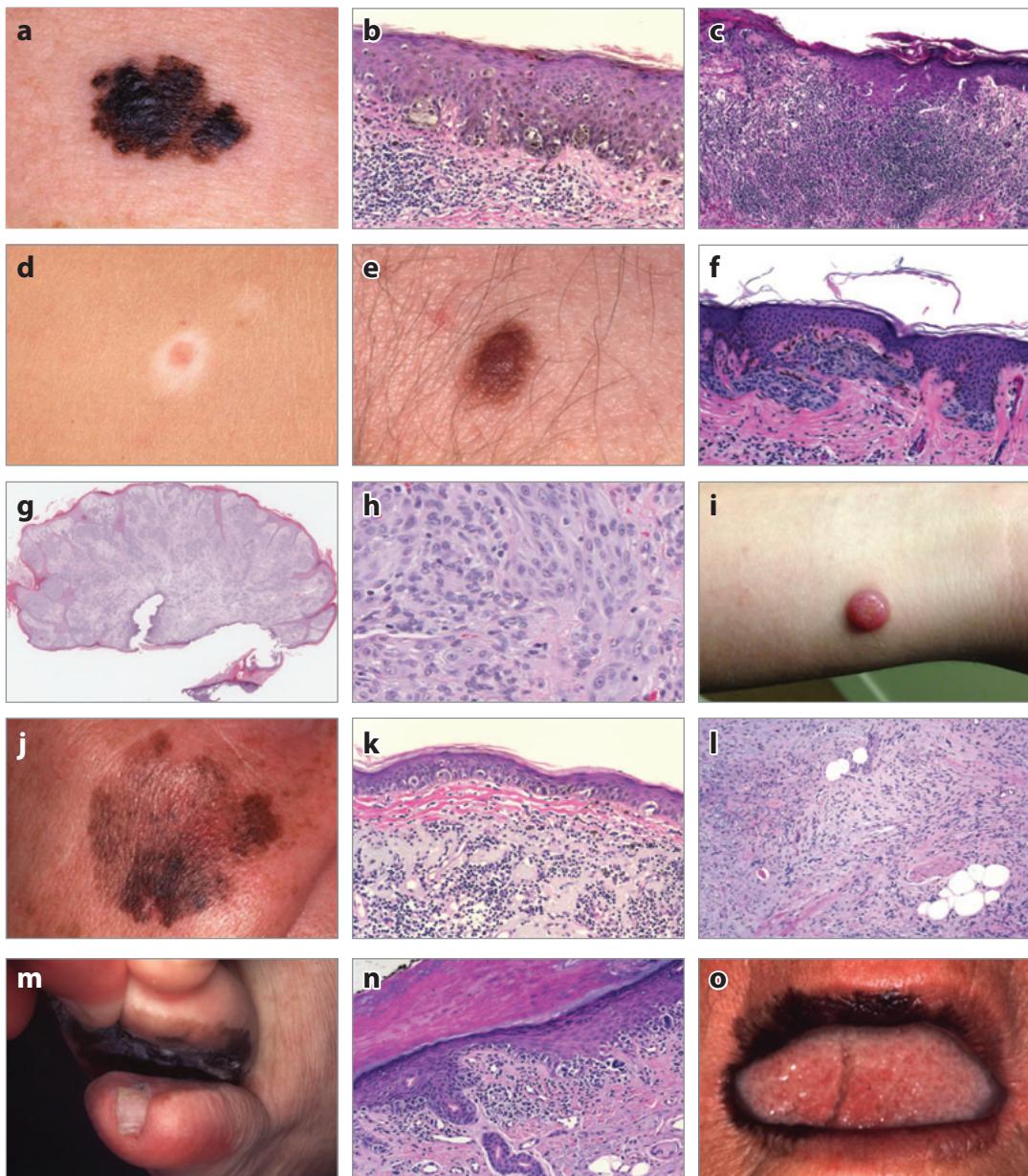
Female-to-male incidence ratio	1	0.6	1:1 (46)	2:1 (110)	0.76 (133)	0.6 (143)	1.1	1.3 (118)	0.53 (130)
Estimated incidence (per million) (108, 110, 133, 143, 147)	1.35	55	2	2.2	5.1 (133)	5 (144)	Not enough data	Not enough data	Not enough data
White-to-nonwhite incidence ratio	6	1.3	2	5	Not enough data	5 (148)	Not enough data	Not enough data	Not enough data
Related melanoma categories	SSM, NM on skin without CSD	LMM, NM on skin with CSD	ALM, SSM, or NM on glabrous skin	Blue nevus-like melanoma	Choroidal, iris, and ciliary tract melanoma	Spindle cell melanoma	Atypical Spitz tumor; pediatric melanoma	Not applicable	Uveal melanoma
Role of UVa	++	+++	(+)	—	(+)?	+++	(+)?	(+)	(+)
Primary oncogenic alterations	BRAF (70%), NRAS (15%)	NRAS (15%), KIT (10%)	KIT (15%), BRAF (15%), NRAS (15%)	KIT (15%), NRAS (15%)	GNAQ and GNA11 (90%)	NF1 (25%)	NRAS (85%); fusions of ROS1, NTRK1, ALK, RET, BRAF, NTRK3	NRAS and GNA11 fusions	GNAS and GNAT1
Secondary genetic alterations	TERT mutations; mutations and deletion of CDKN2A, PTEN	TPX3	TERT amplification	TERT amplification	BAP1	?	CDKN2A?	?	?
Benign precursor	Common acquired nevus	?	?	Melanotic macule	Uveal nevus	—	Spitz nevus	Congenital nevus	Blue nevus
Chromosomal aberrations	Losses of 9, 10, 6q, and 8p Gains of 6p, 7, 8q, 1q, 20q, and 17q	Losses of 9, 6q, 8p Gains of 6p, 7, 11q, 3, 8q, 1q, 20q, and 17q	Losses of 6q, 9p, and 11 Gains of 6p, 7, 8q, 15q, and 20 Amplification of hTERT, RICTOR, and CCND1 loci	Losses of 3q, 4q, 6q, 8p, 9p, 10, 11, and 22 Gains of 1q, 6p, 7, 8q, and 17q Amplification of CDK4 and KIT loci	Losses of 1p, 3, 6q, 8, and 9p Gains of 1q, 6p, and 8q	?	?	?	?
Mutator mechanism	UV radiation	UV radiation	Amplifications	Amplifications	?	UV radiation	Rearrangements	?	?

Abbreviations: ALM, acral lentiginous melanoma; CSD, cumulative sun-induced damage; LMM, lentigo maligna melanoma; NM, nodular melanoma; SSM, superficial spreading melanoma.

^a Parentheses indicate that affected sites have some exposure to UV radiation, but there are no strong indications of a pathogenetic role of UV radiation.

accumulation of degenerated elastic fibers) (5, 6). Follow-up studies confirmed these associations and added additional distinguishing features of a melanoma type characterized by a high frequency of specific *BRAF* mutations with associated clinical features such as increased pigmentation of the primary melanoma by clinical examination (Figure 4a)

and histopathological features such as a predominance of enlarged, hyperpigmented tumor cells of round rather than spindled shape; increased upward intraepidermal scatter; a predominance of tumor cell nests over single cells; and thickening of the involved epidermis (Figure 4b) (6–10). Other genetic alterations associated with this melanoma type



include copy-number increases of chromosome 7, favoring the chromosome harboring the mutant *BRAF* allele (5), and losses of chromosome 10, primarily driven by *PTEN* (6, 11, 12).

Melanomas of this category also more frequently show an associated nevus remnant (9). Independent studies have shown that, compared with melanomas on the head and neck, melanomas on the trunk are associated with increased nevus count, lower self-reported cumulative sun exposure, and fewer nonmelanoma skin cancers (13). These studies also suggest differences among the melanomas arising on the sun-exposed skin, which as a group represent the most common form of melanoma and primarily affect Caucasians. Although these non-CSD melanomas are most commonly of the SSM type, they are more strongly associated with the *BRAF* mutation status (7) than with the SSM category of the WHO classification.

Common acquired nevi. Studies by Pollock et al. (14) and others have shown that *BRAF* mutations are also present in melanocytic nevi; common acquired nevi have the highest *BRAF* mutation frequency. These nevi arise mostly during the first two decades of life. They primarily affect the trunk and extremities, mostly sparing sun-protected sites, which implicates UV radiation as a contributing pathogenetic factor. Histologically, they are separated into three types, depending on whether they are

confined to the epidermis (junctional nevi) or the dermis (dermal nevi) or show both a junctional and a dermal component (compound nevi). There is an unresolved debate among pathologists as to whether the nevi (*a*) form at the dermoepidermal junction, and some melanocytes subsequently drop into the subjacent dermis, or (*b*) originate in deeper structures, and melanocytes subsequently ascend into the overlying epidermis.

The symmetric distribution of the neoplastic cells in most nevi, the monotony of their constituent melanocytes, and the presence of identifiable mutations in bulk populations of nevus cells suggest that nevi result from the clonal expansion of a single cell. Although some studies found that not all melanocytes within an acquired nevus have detectable *BRAF* mutations and concluded that nevi are not necessarily clonal or that *BRAF* mutations are not an initiating event, this finding may have resulted from the technical difficulties of quantifying mutant alleles in small numbers of cells (15, 16). Recent immunohistochemistry studies using a *BRAF^{V600E}*-specific antibody show labeling of the majority of neoplastic cells in melanocytic nevi harboring *BRAF* mutations but no labeling in nevi without *BRAF* mutations. In studies that used digital droplet PCR to quantify the ratios of mutant to wild-type alleles, *BRAF^{V600E}* mutations were fully clonal in neoplastic populations of melanocytes (17).

Figure 4

Clinical and histologic presentations of melanocytic neoplasms originating from melanocytes associated with epithelial structures. (*a*) Melanoma arising on skin without cumulative sun-induced damage (non-CSD): heavily pigmented asymmetrical patch with different shades of black and brown, irregular outline, and altered skin relief. (*b*) Irregular nests of neoplastic melanocytes at the junction between epidermis and dermis. Single melanocytes with dusty pigmentation are scattered throughout upper layers of the epidermis. The dermis harbors a patch infiltrate of lymphocytes and does not show solar elastosis. (*c*) Halo nevus with a dense lymphocytic infiltrate that obscures a proliferation of melanocytes at the dermoepidermal junction. (*d*) Halo nevus with a surrounding depigmented halo. (*e*) Enlarged acquired nevus with clinical features typical of a dysplastic nevus. (*f*) Nevus with bridging of junctional nests of melanocytes and papillary dermal fibrosis, features ascribed to the histopathological aspects of dysplastic nevi. (*g*) Atypical Spitz tumor: exophytic proliferation of spindled and epithelioid melanocytes. (*h*) Close-up of the lesion shown in panel *g*. (*i*) Clinical presentation of an atypical Spitz tumor in a young child. (*j*) Cumulative sun-induced damage (CSD) melanoma: irregularly pigmented patch on skin with CSD. (*k*) CSD melanoma with lentiginous growth pattern. The epidermis is atrophic with atypical melanocytes along the basal layer. The dermis contains amorphous gray material representing solar elastosis, along with an infiltrate of lymphocytes. (*l*) Desmoplastic melanoma: proliferation of atypical spindle cells in the dermis that shows increased amounts of collagen fiber. (*m*) Acral melanoma: irregularly pigmented patch in the web space between toes. (*n*) Markedly atypical melanocytes along the basilar epidermis (lentiginous growth pattern). (*o*) Mucosal melanoma: irregular pigmentation of the lips and oral mucosa.

Tumor-suppressive mechanisms at the junction between nevi and melanomas. The finding in benign nevi of mutations in potent oncogenes such as *NRAS* (in congenital nevi) (18), *HRAS* (in Spitz nevi) (19), *BRAF* (in acquired nevi) (14), and *GNAQ* or *GNA11* (in blue nevi) (20, 21) originally came as a surprise, and significant interest developed in the mechanisms suppressing the expansion of partially transformed melanocytes in nevi. Many of the relevant studies have been performed in the context of *BRAF* mutations and therefore are discussed in this section.

It has been posited, on the basis of the expression of β -galactosidase and other senescence-associated markers, that the melanocytes constituting a nevus are senescent, whereas melanomas have acquired ways to bypass this arrest (22). However, the situation is more complex, because nevi routinely contain cells that label with proliferation markers. Moreover, melanomas, and even melanoma metastases, can also have significant numbers of β -galactosidase-positive cells (23).

This complexity can in part be attributed to the fact that senescence summarizes a growth-arrested cellular state that can be reached by a range of different mechanisms (24). One such mechanism, oncogene-induced senescence, has been proposed as an immediate reaction in which a mutation within a critical signaling pathway such as the mitogen-activated protein (MAP) kinase pathway leads to nonphysiologically high activation, which triggers a stress response that induces cyclin-dependent kinase inhibitors such as p21 and p16, leading to permanent G₁ arrest (25, 26). In most melanomas p16 is disabled by deletion, mutation, or silencing of *CDKN2A*, and germ-line mutations in *CDKN2A* predispose to melanoma with high penetrance; these observations demonstrate the critical role of p16 in melanoma (27, 28). Among melanoma patients, 10% have a family history of melanoma, and of these, 20–40% carry germ-line mutations of *CDKN2A*. Most of these mutations are in the exons or in the reading frame that affects p16 and not the p14

protein that is transcribed from the same gene (29, 30). Approximately 2–3% of melanoma families have *CDK4* (cyclin-dependent kinase 4) mutations in the p16-binding domain (31), and a similar percentage have mutations that specifically disable p14ARF (p14 alternate reading frame) (30, 32), which acts upstream of p53, by inhibiting its ubiquitinase, MDM2 (mouse double minute 2 homolog).

However, nevi are composed of several tens of thousands of cells or more, indicating that the senescence mechanisms at work are not immediate in nature but engage with some latency. It is conceivable, but difficult to prove, that individuals without nevi are able to effectively induce such an immediate senescence response and thereby subdue any oncogene-induced melanocytic proliferation before a clinically detectable lesion can form. By contrast, individuals with constitutional defects in immediate-type senescence may engage secondary mechanisms that operate with a longer latency and thus lead to the formation of populations of neoplastic melanocytes large enough to constitute a nevus. Patients with inherited *CDKN2A* mutations have more and larger nevi than their wild-type relatives do (33, 34), indicating that p16 exerts its tumor-suppressive function from nevus initiation to later phases of nevus growth.

Relicative senescence induced by progressive telomere shortening is an inherently latent mechanism. However, critical telomere shortening occurs after approximately 60 population doublings, far more than would be expected to occur in a nevus if all progeny from the initial founder cell were proliferating equally. Other latent mechanisms linked to senescence include stochastic events triggered by DNA-replication stress. Constitutive proliferative signals from activated oncogenes can lead to DNA hyperreplication, resulting in genomic instability in the form of prematurely terminated DNA replication forks and subsequent double-stranded DNA breaks. Markers of DNA damage cosegregate with markers of senescence in precancerous lesions, including acquired and dysplastic nevi, implicating oncogene-mediated

replication stress as an additional barrier to transformation (35). This mechanism should operate stochastically and may explain why cell proliferation is exhausted in nevi following a period of latency. In support of this model, random DNA copy-number changes can be observed within the melanocytes of a nevus by fluorescence *in situ* hybridization, whereas clonal chromosomal aberrations are typically present only at the melanoma stage (36). Exhaustion of nucleotides by oncogene-induced suppression of their synthesis precedes and directly contributes to the DNA-damage response (37). Additional mechanisms implicated in melanocyte senescence include paracrine factors such as interleukin-6 and interleukin-8 (38). In summary, these findings indicate that multiple, independent barriers restrain the proliferation of partially transformed melanocytes. A nevus that has reached a stable size thus might represent a population of cells that were growth arrested by different mechanisms. This model would explain the mosaic expression of individual senescence markers among the cells of a given nevus (23, 39). It would also explain why the removal of individual mechanisms, even critical ones such as p16, is not sufficient to bypass the senescence process of melanocytes (22), because other senescence mechanisms remain intact. The more such barriers become disabled by inherited or somatic mutations, the larger the nevus will grow—and with it the probability of full transformation to melanoma.

Activation of telomerase is emerging as a critical barrier to full transformation of melanomas. Although telomerase activity is low or absent in nevi, melanoma metastases typically have significantly increased activity; telomerase activity in primary melanomas lies somewhere in between (40, 41). Amplification of the *TERT* (telomerase reverse transcriptase) gene is a common event in acral melanoma (6, 42) and coincides with the transition to more advanced primaries (43). Recently, frequent mutations in the telomerase promoter were found in melanoma (44, 45). Two mutational hotspots were found in a mutually exclusive pattern in more than 85% of melanoma metas-

tases. The frequency was significantly lower in primary melanomas (33%), and mutations were not found in 25 nevi that were examined, consistent with the pattern of telomerase activation reported previously. These findings implicate telomere length as a factor limiting net cell expansion at some point during the evolution of the primary melanoma. The finding of mutations activating telomerase in primary melanomas indicates that they convey a selective advantage while the number of neoplastic cells is significantly below the Hayflick limit. Therefore the neoplastic population is likely to have undergone constant pruning by growth arrest or cell death of individual cells. It is noteworthy that primary melanomas frequently show areas of regression, that is, areas in which neoplastic melanocytes vanished but left behind a telltale sign in the form of melanophages (macrophages that have ingested leftover melanin pigment). Immune cell-mediated killing of melanoma cells is proposed to be the main mechanism leading to melanoma regression, but telomere crisis has emerged as a possible alternative mechanism (46).

However, in addition to the cell-autonomous mechanisms outlined above, immune-mediated mechanisms undoubtedly play an important role in restraining the expansion of the partially transformed melanocytes of nevi and early melanomas. Nevi often show infiltrates of lymphocytes and are known to acutely regress when the inflammation is more pronounced (**Figure 4c**). The latter scenario is often accompanied by a clinically visible depigmented halo surrounding the nevus (**Figure 4d**). This phenomenon can affect multiple nevi simultaneously and can even be accompanied by vitiligo, suggesting that immune responses against one nevus can extend to others. Nevi can also enlarge or erupt *de novo* in large numbers under immunosuppression. Such eruptive nevi also frequently have *BRAF* mutations and favor sun-exposed sites, indicating that the initiating mechanisms are identical to those of other acquired nevi (47). In a liver model, hepatocytes that became senescent due to oncogenic *RAS* were effectively eliminated by immune cells

5-hmC: 5-hydroxymethylcytosine

(48). The phenomenon of eruptive nevi in immunosuppressed patients may indicate a similar immune surveillance that restricts the number of neoplastic melanocytes in melanocytic nevi. However, it does not happen in all individuals that become immunosuppressed, indicating that other factors are involved.

Epigenetic alterations are emerging as additional tumor-suppressive mechanisms in melanoma, as evidenced by recurrent mutations in genes involved in chromatin remodeling and by dramatic changes in the chromatin state and DNA modifications in melanoma. Sequencing studies have revealed recurrent mutations in components of the SWI/SNF (switt/sucrose nonfermentable) complex, including *ARID1A*, *ARID1B*, and *ARID2* as well as *SMARCA4* (12, 49). *SETDB1*, which encodes a histone-methylating enzyme, is amplified in melanoma, and its expression is increased compared with that in melanocytic nevi (50). *SETDB1* resides within a narrow locus on chromosome 1q21.3 that has also been implicated in melanoma susceptibility by genome-wide association studies (51). The histone methyltransferase EZH2, a member of the Polycomb complex, is more highly expressed in melanomas than in nevi (52), and mutations have been found in a small proportion of melanomas (49). Several studies have reported marked alteration of the chromatin landscape and DNA-methylation status during the progression to melanoma. The histone variant macroH2A is markedly less abundant in melanomas than in nevi, due to transcriptional downregulation (53). Genome-wide hypomethylation and hypermethylation of certain promoter regions, compared with that in normal melanocytes or neoplastic melanocytes of nevi, have been observed in melanoma (54–56). In melanocytic nevi, 5-methyl hydroxylation of cytosine is abundant, but there is a genome-wide loss of 5-hydroxymethylcytosine (5-hmC) at some point during the transition to melanoma (57). The genetic basis underlying this dramatic change in 5-hmC is not clear. In myelodysplastic syndrome and acute myeloid leukemia, loss of 5-hmC is caused by loss of function of the 5-methyl cytosine hydroxylat-

ing enzymes of the TET (ten-eleven translocation) family, which can occur as a result of inactivating mutations or functional inhibition via the production of 2-ketoglutarate that is caused by mutations in the isocitrate dehydrogenase (IDH) genes *IDH1* or *IDH2* (58, 59). Although mutations in TET family members and IDH proteins appear to be infrequent in melanoma, decreased expression levels of these proteins have been implicated as a mechanism underlying the loss of 5-hmC in melanoma progression (57). Inactivating mutations in the deubiquitinase BAP1 are frequent somatic events during the progression of uveal melanoma, as discussed below (60). Germ-line mutations in *BAP1* were independently discovered in two families with uveal and cutaneous melanoma and atypical Spitz tumors. In this setting, somatic loss of the remaining wild-type allele in acquired nevi with *BRAF^{V600E}* led to clonal expansion of enlarged epithelioid melanocytes with nuclear pleomorphism, but not to clear-cut melanoma (61, 62). The resulting lesions show a biphasic morphology and represent an example of stepwise tumor progression in which a common acquired nevus is initiated by *BRAF* mutation from which a morphologically distinct clone emerges upon biallelic inactivation of BAP1. BAP1 interacts with the Polycomb additional sex combs-like (ASXL) factors ASXL1 and ASXL2 and their substrates HCF1 (host cell factor 1) and OGT (*O*-glucosyltransferase), but the precise mechanism by which its loss of function bypasses growth arrest in *BRAF*-mutated nevi remains to be elucidated.

Dysplastic nevi. The concept of the dysplastic nevus was introduced in 1978 to identify a melanoma risk indicator and potential melanoma precursor (63). In a series of melanoma families, family members were noted to have increased numbers of acquired nevi that were unusually large, often exceeding 10 mm, and showed irregular pigmentation (**Figure 4e**). The nevi observed in patients with this syndrome were proposed to have specific histopathological features such as nuclear pleomorphism, bridging of rete ridges, and lamellar

fibrosis of the papillary dermis (**Figure 4f**). It later became clear that these histopathological features are very common in acquired nevi in general and are not specifically associated with the syndrome or with the size of the nevi (64, 65). It also became clear that in the general population, the risk of progression to melanoma is very low. Dysplastic nevi remain stable for decades and tend to fade away later in life, and removal of clinically dysplastic nevi is not recommended as long as a lesion is not suspicious for melanoma.

Despite this shift in perception, the term dysplastic remains widely used among clinicians and pathologists today, but with varying connotations. Clinicians use it to describe acquired nevi measuring 7 mm or more that are flat, are brown to orange-brown, do not show the elongated hair of congenital nevi, and may have a darker or lighter center and fuzzy borders. However, similar features are frequently present in smaller nevi as well. Pathologists use the term to describe nevi that have irregular architectural and cytological features. Some schools of thought grade lesions as having mild, moderate, or severe dysplasia in an attempt to position the lesion on a progressive scale between a benign nevus and a melanoma (66). This view contradicts the notion that they are entirely benign lesions, implying that they instead represent a spectrum. Another school uses the term to convey histopathological correlation to the clinician's perception of dysplasia but considers them entirely benign. The latter approach is based on the proposal of Ackerman & Milde (67) to term flat nevi with bridging of nests and papillary dermal fibroplasia Clark's nevi. The inconsistent usage and different meanings of the term dysplastic nevus have generated significant and unresolved controversy and confusion about treatment, in particular of incompletely excised lesions.

What remains uncontroversial is that the presence of multiple enlarged acquired nevi is associated with increased melanoma risk. For this assessment, clinical examination is sufficient, and histological examination does not provide additional information. It is also firmly

established that nevus size is a heritable trait (68). The higher the count of large nevi, the greater the melanoma risk (69). Clinically dysplastic nevi are thus a symptom of systemic melanoma susceptibility and reflect the inherited loss of genes functionally involved in restraining the proliferation of melanocytes that have acquired oncogenic mutations in genes such as *BRAF*. The phenotype is therefore useful to identify patients with increased melanoma risk. By contrast, the degree of histopathologic dysplasia, if used to determine how closely a given lesion comes to bona fide melanoma, provides information only about the lesion at hand and not about the patient's overall melanoma risk. Nevi with histopathological dysplasia have been analyzed for allelic imbalance and have recurrent losses of heterozygosity of the *CDKN2A* and *TP53* loci (70) as well as *BRAF^{V600E}* mutations (14).

The collective findings outlined above indicate that the neoplastic proliferation initiated by mutations in oncogenes is restrained by a multitude of independent mechanisms, which can be overridden by additional mutations that lead to loss of function in multiple tumor-suppressor genes. These mutations can be inherited or somatically acquired. The sizes, shapes, and colorations of acquired nevi are more homogeneous within an individual than among different individuals. In twin studies, the average nevus size is more tightly correlated among monozygotic twins than among dizygotic twins (71). This fact is used clinically to identify pigmented lesions outside the range defined by most of an individual's nevi as suspicious for melanoma (the ugly duckling sign). In this light, the phenotype of dysplastic nevus syndrome—that is, the presence of multiple enlarged acquired nevi—indicates the inherited loss or impairment of senescence barriers. By contrast, the ugly duckling lesion arises through the somatic loss of barriers only within the neoplastic population of that lesion.

Spitz tumors. The term Spitz nevus makes reference to Sophie Spitz's description in 1948 of a form of melanoma in children that

expresses a benign behavior. This contradiction was resolved later by renaming lesions with these features as Spitz nevi. However, this type of melanocytic neoplasm, characterized by a predominance of large polygonal (epithelioid) or spindled melanocytes with enlarged nuclei and often multinucleated cells (**Figure 4g,h**), has retained considerable notoriety, because some cases diagnosed as such by even expert consultants have metastasized widely. Here the term Spitz tumor is used to capture a range of melanocytic neoplasms with overlapping histomorphology that includes Spitz nevi on the benign end of the spectrum and spitzoid melanomas with lethal potential on the other. The term atypical Spitz tumor captures borderline lesions with intermediate histological features. Spitz tumors occur more commonly in children and young adults but can occur in all age groups. Most neoplasms diagnosed as melanoma of childhood fall in the category of Spitz tumors and tend to have lower mortality than melanomas of similar thickness in older patients do. Paradoxically, they often grow rapidly (**Figure 4i**), show numerous mitotic figures, and commonly have lymph node involvement (72, 73). In one study, 47% of 52 cases had lymph node metastases, but only a single patient died of metastatic melanoma (73). In conventional melanoma, less than 20% of patients have a positive sentinel lymph node, and those that do have increased risk for distant metastasis and death (74).

It is becoming increasingly clear that Spitz tumors are a heterogeneous group of genetically and biologically distinct categories. Approximately 20% of cases show oncogenic mutations of *HRAS*, typically accompanied by copy-number increases of the entire short arm of chromosome 11 as the only chromosomal aberration (19). This type of Spitz nevus often presents as a predominantly intradermal, horizontally oriented proliferation of large epithelioid melanocytes widely dispersed singly between thickened collagen bundles in the deep reticular dermis. Another variant has a combination of *BRAF^{V600E}* mutations and biallelic loss of the tumor suppressor *BAP1* on chromosome

3p21 (62). These lesions present as intradermal, often dome-shaped or polypoid, and vertically oriented proliferations of large epithelioid melanocytes arranged in large cellular aggregates, often in continuity with a conventional acquired nevus. In these combined lesions, the transition to the epithelioid phenotype coincides with the loss of *BAP1*, demonstrating that this Spitz tumor variant represents a progression from an acquired nevus (61).

The driving oncogenic alterations of the remaining Spitz tumors are now rapidly emerging. Recent studies have revealed a high frequency of rearrangements of kinases in the remainder of Spitz tumors. Rearrangements resulting in fusion kinases of *ROS1*, *ALK*, *RET*, *NTRK1*, and *BRAF* were observed in 60% of cases (T. Wiesner, J. He, R. Yelensky, R. Esteve-Puig, T. Botton, et al., submitted manuscript). The rearrangements fuse the intact kinase domains in frame to a wide range of 5' fusion partners, including genes involved in similar rearrangements in lung, colon, and thyroid tumors and lymphoma as well as several novel fusion partners. Most of the 5' fusion partners have coiled-coil domains, suggesting that they allow the kinase domains to dimerize and autophosphorylate, resulting in ligand-independent constitutive activation of multiple oncogenic signaling pathways, including the MAP kinase, PI3 (phosphoinositide 3)-kinase, and STAT (signal transducer and activator of transcription) pathways, with potent induction of proliferation. Many of the kinases involved in the rearrangements are expressed only during development, including neural crest development, and are silenced in adult tissues. The kinase fusions thus lead simultaneously to expression and kinase activation, which explains why rearrangements rather than point mutations are the predominant mode of oncogenic activation.

It is unclear how the specific nature of these oncogenic alterations is related to the unique biological behavior of atypical Spitz tumors and spitzoid melanomas of childhood, with their rapid growth and frequent metastasis but rare lethal outcomes. The clinical behavior indicates that progression is not constrained by the

nature of the oncogenic signaling, as it can potently drive tumor growth and metastasis. It is conceivable that Spitz tumors are not immortal and lack the high mutation burden that can make mutations in the telomerase promoter likely, as occurs in most lethal melanomas. They also lack the chromosome-level genomic instability required to amplify the *TERT* locus, as occurs in acral melanoma. The immune system is another possible mechanism to restrain the proliferation of fusion-driven melanocytic neoplasms. Fusion kinases are chimeric proteins with neoantigenic properties, which could make them an easier target for the immune system than are oncogenes activated by point mutations.

Pediatric melanoma. Pediatric melanomas represent a heterogeneous group comprising lesions related to atypical Spitz tumors, melanomas arising in congenital nevi that occur primarily in younger children, and non-CSD melanomas in older, postpubertal children. The latter have clinical features and outcomes similar to those in adults. Prepubertal melanomas also occur in nonwhites, but the overwhelming majority of melanomas in postpubertal children affect whites (75). These melanomas are discussed separately with Spitz tumors and non-CSD melanomas, above. Not all melanomas in prepubertal children are spitzoid or arise in congenital nevi, which raises the possibility that additional subtypes exist.

The roles of UV radiation and skin pigmentation. People living in geographic regions with increased UV exposure have an increased melanoma risk (76). However, the relationship between melanoma risk and degree of exposure to UV radiation is complex. Paradoxically, melanomas on intermittently exposed skin, such as the trunk and proximal extremities, develop at a younger age, whereas melanomas on sites that are more frequently exposed, such as the face, ears, and neck, develop significantly later in life (10, 77). Whole-genome and whole-exome sequencing studies have revealed a high burden of mutations, pre-

dominantly cytosine-to-thymidine transitions, in the genomes of melanomas originating from sun-exposed sites, which provides compelling genetic validation that UV radiation is a major mutagenic factor (12, 49, 78). Consistent with the finding that melanomas with *BRAF* mutations primarily occur on intermittently exposed skin, the genomes of *BRAF*-mutant melanomas have intermediate mutation burdens (~30,000) when compared with those of chronically exposed (~100,000) and unexposed (<1,000) sites (12). Approximately 80% of mutations are compatible with UV-related mutagenesis, which confirms that UV radiation is the predominant mutagen involved in the pathogenesis of these types of melanoma. The exceptionally high mutation burden in melanomas originating from sun-exposed skin raise the question of whether inherited defects in DNA repair contribute to mutation burden and melanoma susceptibility (12, 49). Some studies have found that polymorphisms in DNA-repair genes such as *ERCC2* (excision repair cross-complementing rodent repair deficiency 2) are associated with melanoma risk (79), and others have found high frequencies of somatic loss-of-function mutations of genes in the DNA-damage response pathways that include *ERCC2* (80).

Although the above associations identify UV radiation as a pathogenetic factor for melanocytic neoplasia, the most common mutation in *BRAF* at codon V600 is a thymidine-to-adenine transversion and thus not a classic UV-signature mutation. *BRAF^{V600E}* mutations also arise in thyroid or colorectal cancer, which indicates that UV radiation is not required for their formation. This finding, however, does not rule out a causal role for UV radiation in the formation of *BRAF^{V600E}* mutations in melanocytic neoplasms. Although UV radiation causes mutations primarily at pyrimidine dimers, it causes a broad range of other mutations as well (81). Indirect effects of UV radiation must also be considered, because the interaction of UV radiation with melanins generates free radicals, which could act as secondary mutagens. Data from genetically engineered mouse models have shown that

MELANOCYTE DEVELOPMENT AND THE TANNING RESPONSE

Melanocytes are derived from the neural crest. Two developmental pathways give rise to melanocyte progenitor cells (140). The dorsolateral pathway generates melanoblasts that travel through the mesoderm to colonize the epidermis, mucosa, and hair follicles. A second population of neural crest cells gives rise to bipotent precursors of Schwann cells and melanocytes that migrate ventromedially to reach the skin. There are differences in the signals involved in fate determination and migration of the two developmental pathways; END3 signaling plays a major role in the ventromedial pathway.

After migration is completed, most differentiated melanocytes reside within epithelia, primarily in the epidermis and hair follicles of the skin but also in mucosal epithelium. They are also found in significant numbers in the leptomeninges and the inner ear and at lower density in many internal organs. The life span and life cycle of melanocytes are not fully understood. In hair follicles, the number of melanocytes waxes and wanes with the cycling of the follicle, and the stem cells to replenish melanocyte populations early in the anagen phase reside in the bulge region of the hair follicle (141). These melanocyte stem cells can also replenish lost epidermal melanocytes in conditions such as vitiligo. However, the degree to which stem cells within the hair follicle contribute to melanocytes of the interfollicular epidermis under homeostatic conditions and the kinetics with which they do so remain to be determined.

The main function of differentiated melanocytes is to produce melanin and distribute it to surrounding epithelial keratinocytes to protect them from UV radiation. There are two chemically distinct types of melanin. One, eumelanin, is black and is the more abundant form in dark-skinned people. The other, pheomelanin, contains benzothiazine and benzothiazole, and its color is more orange. Although pheomelanin is also present in higher concentrations in the melanocytes of dark-skinned individuals, its relative abundance is much higher in individuals with light complexion, who have lower amounts of eumelanin. Eumelanin production is subject to dynamic regulation as part of the tanning response, in which activation of MC1R by MSH, synthesized by keratinocytes, stimulates pigment synthesis via the induction of the transcription factor MITF (microphthalmia-associated transcription factor).

UVA radiation requires melanin to be present to induce melanoma formation (82). The specific mutations that induce melanoma formation in this model are not yet known, but the results nonetheless clearly establish that an interaction between UV radiation and melanin is a causative mechanism. Additional studies have confirmed and further refined the role of melanin. In a mouse model in which mutant *BRAF^{V600E}* can be conditionally activated in melanocytes, melanomas develop only if melanin, specifically pheomelanin, is present (see the sidebar). Significant oxidative DNA and

lipid damage occurs even in the absence of UV radiation in the normal skin of “redhead” mice that, due to a loss of MC1R (melanocortin 1 receptor) function, produce only pheomelanin. In this model, the *BRAF* mutations are pre-engineered so that melanoma formation depends on the occurrence of additional, as yet unknown mutations. However, the experiment provided additional evidence of the mutagenic effect of pheomelanin or an increased pheomelanin-to-eumelanin ratio, which are expected to be enhanced under exposure to UV radiation. How the balance of pheomelanin and

eumelanin affects mutagenesis is not known, but because the melanosome ultrastructure differs between light- and dark-skinned individuals, it has been proposed that eumelanin encases pheomelanin as an antioxidative coating in dark-skin melanosomes and that this function is impaired in patients with light skin (83, 84). Emerging evidence also suggests that induction of the DNA-damage response may be hardwired into the tanning response. Stimulation of MC1R with melanocyte stimulatory hormone (MSH) not only is the main initiator of pigment synthesis in melanocytes but can also directly activate scavenging of reactive oxygen species (ROS) (85).

Most individuals of European descent have inherited variants in *MC1R* that blunt the receptor's ability to activate downstream signaling and to induce tanning in response to binding of MSH. Several loss-of-function variants of *MC1R* are highly associated with red hair, poor tanning, freckling of the skin, and increased melanoma risk (86). Germ-line polymorphism in a range of other genes affecting skin pigmentation, including *ASIP*, *OCA2*, *SLC45A2*, *TYRP1*, and *TYR*, are associated with melanoma risk (51, 87–92), but inherited polymorphisms in *MC1R* are probably the most important genetic factor among risk alleles that occur at high frequency in populations. The risk conveyed by germ-line variants of *MC1R* might differ for the various molecular subtypes of melanoma. In non-CSD melanomas, *MC1R* variants were strongly associated with *BRAF* mutations in some studies (93, 94) but not in others (95, 96). This discrepancy may be due to differences in the ethnic compositions of the cohorts and/or their proportions of melanoma types. CSD melanomas have a low frequency of *BRAF* mutations. Those that occur are primarily of the V600K rather than V600E type (97). In addition, *BRAF* mutations in CSD melanomas are associated with wild-type *MC1R* alleles (98), indicating that any interactions between *BRAF* and *MC1R* may be complex.

The consistent observation that melanocytic neoplasms with V600E mutations in *BRAF* have a high frequency early in life that later de-

creases, whereas V600K mutations show the opposite pattern, is a puzzle that remains to be solved. The early onset and multiplicity of nevi with *BRAF* mutations in some individuals suggest a genetically determined susceptibility that manifests as a high mutation rate in melanocytes. The odds of mutating one of the single base pairs that can generate the C.1799T>A mutation that results in the V600E allele are exceedingly low, so the presence of multiple independent nevi with these mutations on the skin indicates an already high mutation burden in normal melanocytes at a young age. In addition to sun exposure in early life, neonatal blue-light therapy for the management of hyperbilirubinemia, which is administrated to a significant proportion of newborns and contains some UVA radiation, increases the number of nevi (99). Sun protection in children reduces nevus formation (100). The link between sun exposure in early life and nevus and melanoma risk has led to increased legislation to restrict access to tanning beds for minors, the use of which has been associated with melanoma risk (101).

In summary, both the number and the size of acquired nevi contributing to melanoma risk can be interpreted in the light of the factors outlined above. The number of nevi in an individual is proportional to the number of initiating mutations, which reflects the mutational burden in melanocytes of the skin. This mutation burden is a product of cumulative exposure to UV radiation and the host's defense and repair mechanisms, consisting of the production of melanin and detoxifying ROS as well as the repair of UV-radiation-mediated DNA damage.

Melanomas on skin with cumulative sun-induced damage. The second most common type of melanoma in Caucasians arises in areas chronically exposed to the sun, predominantly the face, ears, neck, and lower extremities. These melanomas tend to arise approximately two decades after the peak of non-CSD melanomas, typically in individuals over 60 years old (10). They are associated

ROS: reactive oxygen species

not with an increased number of acquired nevi but instead with signs of high cumulative exposure to UV radiation, such as solar elastosis and nonmelanoma skin cancers (5, 102). The genetic alterations in melanomas on chronically sun-exposed skin differ from those in non-CSD melanomas, as shown in **Table 1**: CSD melanomas have infrequent *BRAF* mutations (more often V600K than V600E) (5, 12, 97); inactivating mutations of *NF1* (30% of cases) (12); copy-number increase of *CCND1* (20% of cases) (6, 103); activating mutations of *KIT* (~10% of cases) (104); increased mutational frequencies in *TP53* and *ARID2* (12); and differences in the pattern of chromosomal aberrations (6). Their mutation burden is very high: Somatic mutations number 100,000 or more (49). The most common presentation is that of LMM, in which a pigmented macule (**Figure 4j**) that comprises a subtle proliferation of single intraepidermal melanocytes (**Figure 4k**) develops, often after many years, a nodular growth with dermal invasion. However, melanomas on skin with chronic sun-induced damage that present as nodular melanomas without an adjacent *in situ* component show indistinguishable genetic alterations, indicating that they represent an accelerated transition from *in situ* to invasive melanoma rather than a unique class of melanoma (6).

Desmoplastic melanomas. Desmoplastic melanomas are primarily intradermal proliferations of spindled melanocytes with atypical nuclei that are interspersed at varying density between thickened collagen bundles (**Figure 4l**). They most commonly arise on the chronically sun-exposed skin of the head and neck. Their distinctive clinical and genetic characteristics indicate that they represent a distinct subtype of melanoma. They lack activating mutations in the known melanoma oncogenes *BRAF* (105), *NRAS*, *KIT*, *GNAQ*, and *GNA11* but have loss-of-function mutations in *NF1* in approximately 25% of cases (A.H. Shain, B.C. Bastian, unpublished observation). In 30–40% of cases they show growth along nerves, a feature associated with an increased risk of recurrence.

Although little is known about their pathogenesis, their preference for heavily sun-exposed sites indicates that UV radiation is a causative factor. Most desmoplastic melanomas have a lentiginous component, as in LMM, and whole-genome sequencing studies have revealed a high mutation burden with a strong UV signature, as in CSD melanomas (A.H. Shain, B.C. Bastian, unpublished observation), which indicates a superficial location for their cell of origin.

Acral and Mucosal Melanomas

Melanomas originating from the glabrous (non-hair-bearing) skin (**Figure 4m**) and the nail apparatus have characteristic morphological, epidemiological, and genetic features that set them aside from other subtypes. They share several features with melanomas originating from mucosal epithelia (mucosal melanomas) and therefore are discussed together. Glabrous skin, by definition, lacks hair follicles but has eccrine glands. Mucosa lacks either structure, but may, depending on the site, have mucosal glands. These differences may be relevant for the cell of origin of these melanoma types. The bulge region of the hair follicle harbors melanocyte stem cells in the nonglabrous skin; whether an equivalent melanocyte stem cell reservoir exists in glabrous skin is unclear. Acral melanomas frequently involve the eccrine sweat ducts, and early acral melanoma *in situ* has a characteristic preference to grow around the openings of eccrine sweat ducts along the ridges of the dermatoglyphs, a pattern that provides diagnostic information (106). It remains to be determined whether the apparent tropism of acral melanoma to eccrine glands indicates that they harbor the cell of origin or that they play other important roles during the early phases of the disease.

Acral melanoma and mucosal melanoma often show a lentiginous component, in which melanocytes are found predominantly as single units in the basal layer of the epithelium (**Figure 4n**). For this reason, the term acral lentiginous melanoma (ALM) has been employed (107). However, the characteristic

genetic alterations detailed below are found in virtually all melanomas presenting on glabrous skin, so the term acral melanoma is used here to encompass all traditional melanoma types—ALM, NM, and SSM—that can present on glabrous skin.

As shown in **Table 4**, acral and mucosal melanomas do not have the dramatically increased frequency in Caucasians that melanomas on sun-exposed skin do (108, 109). Acral melanomas arise on relatively sun-protected sites and are additionally shielded by a thickened cornified layer or a nail plate. Epidemiologic studies do not indicate an increase in incidence with latitude or degree of sun exposure. These findings make UV radiation an unlikely pathogenetic factor, a conclusion supported by whole-genome sequencing studies, which do not reveal the high degree of UV-signature mutations found in melanomas originating from the nonglabrous skin (12, 49). Mucosal melanomas (**Figure 4b**) primarily involve the mucosa of the anogenital region, nasal cavity, and paranasal sinuses, but they can occur in all mucosal sites (110).

The spectrum of somatic mutations in acral and mucosal melanomas differs from that in melanomas on sun-exposed sites. In acral melanoma, mutations in *BRAF* occur in 15% of cases, much less frequently than they do in non-CSD melanomas. They are absent in mucosal melanoma. Activating mutations or amplifications of wild-type *KIT* are found in 15–40% of acral and mucosal melanomas, and approximately 15% have *NRAS* mutations (49, 104, 111). A unique feature of acral and mucosal melanomas is the high frequency of gene amplifications throughout the genome (42). Most acral melanomas have multiple (five, on average) focused gene amplifications, most commonly involving the loci that include *CCND1* (11q13), *bTERT* (5p15), *CDK4* (12q14), *RICTOR* (5p13), and *KIT* and *PDGFRα* (4q12) (6, 12, 42). Mucosal melanomas share this high frequency of amplifications, but the frequency at individual loci differs, which may indicate biological differences between these melanoma

types (6, 112). As opposed to those in other solid tumors, in which gene amplification typically arises later during progression, amplifications in acral melanoma are already present in the earliest detectable phases of the disease (42). The nature of the genomic instability that favors gene amplifications over other mutation types remains to be determined.

Although melanocytic nevi are not infrequent in acral sites, and can also affect the nail apparatus, they do not appear to play a significant role as precursors, as they are rarely found contiguously with acral melanomas. Limited information is available on the somatic mutations in acral nevi, but *KIT* mutations have not been found (113). Similarly, nevi are not found in association with mucosal melanomas. Instead, the earliest detectable manifestation of acral and mucosal melanomas is melanoma *in situ*, in which a subtle intraepithelial proliferation of single melanocytes with enlarged nuclei and uneven and thickened dendrites is present, often extending over a considerable distance. Studies in acral melanoma that use fluorescence *in situ* hybridization revealed individual, evenly spaced melanocytes, which appear morphologically normal but are genetically aberrant, colonizing stretches of apparently normal skin, up to 1 cm wide, immediately adjacent to obvious melanoma *in situ*. These so-called field cells share the same amplifications as the histopathologically apparent melanoma cells, albeit often at lower copy number, indicating that they are genetically not as evolved and represent an earlier progression phase (42, 43). Preliminary studies suggest that *KIT* mutations may arise early and are followed by amplification of *CCND1*, and subsequently *bTERT*, which coincides with a substantial increase in the neoplastic cell population (43). This finding is consistent with experimental studies, in which mutant *KIT* primarily induces migration and survival rather than significant cell proliferation (114, 115). Accordingly, *KIT* mutations may differ from *BRAF* mutations in that they are not sufficient to allow proliferation significant enough to result in an equivalent to the nevi observed with *BRAF* mutations.

The relatively high frequency of *CCND1* amplifications in these melanoma types may indicate that additional genetic alterations that drive proliferation may be required. A low to moderate frequency of *KIT* mutations is common to acral, mucosal, and CSD melanomas, which tend to share an often extensive lentiginous growth phase that progresses slowly, often over many years, before invasive melanoma develops.

The role of *KIT* in melanoma is complex; it inhibits proliferation or cell survival in certain settings and was therefore was originally considered to play a tumor-suppressive role in melanoma. This interpretation was based on the observations that *KIT* protein levels decline from *in situ* to invasive components and that forced expression of *KIT* in melanoma cell lines decreases their ability to proliferate and/or survive (116). However, in certain other types of melanocytic neoplasms, *KIT* clearly acts as an oncogene: Patients whose melanomas harbor activating mutations in *KIT* can have dramatic responses to *KIT* inhibitors (117).

MELANOCYTIC NEOPLASMS ORIGINATING FROM MELANOCYTES NOT ASSOCIATED WITH EPITHELIA

Congenital Nevi

Nevi that arise in utero are termed congenital melanocytic nevi. These, by definition, develop in the absence of UV radiation. However, the definition is often extended to include nevi that arise shortly after birth. Congenital nevi have several distinctive features that set them apart from acquired nevi. They tend to be significantly larger and can cover extensive portions of the body surface (**Figure 5a**). During childhood and adolescence, they increase in size proportionally with the body area. They are termed large or giant congenital melanocytic nevi when their predicted size at adulthood exceeds 20 or 60 cm, respectively. These larger congenital nevi can also involve extracutaneous sites, including the CNS. This condition, known as

neurocutaneous melanosis, can develop severe or even lethal compression symptoms of vital CNS structures. On the skin, congenital nevi often show marked hypertrichosis with hyperpigmented terminal hairs (**Figure 5b**), whereas acquired nevi do not. Their constituent melanocytes are also found in deeper structures of the skin and beyond, occasionally involving deep soft tissue and muscle. Large and giant congenital nevi frequently present with satellite lesions—discrete, morphologically similar, but smaller nevi elsewhere on the skin (**Figure 5a**). Of patients with large or giant congenital nevi, 2–3% develop melanomas, 70% of which develop before puberty. Melanomas can develop within any tissue involved by the nevus, and risk increases with the size and cellularity of the nevus (118). Primarily in neonates and mostly in giant congenital nevi, rapidly proliferating nodules can develop, which can reach several centimeters in diameter, can show an increased mitotic rate, and can be difficult to histologically distinguish from melanoma (119). These proliferating nodules have characteristic chromosomal aberrations that entail gains and losses of multiple entire chromosomes, rather than the copy-number changes involving chromosome fragments that are commonly observed in melanomas (120). This pattern of aberrations suggests a chromosome segregation defect in the constituent melanocytes. The proliferating nodules are not considered malignant as they frequently regress spontaneously.

In contrast to acquired nevi, in which *BRAF* mutations predominate, large and giant congenital nevi carry *NRAS* mutations in more than 80% of cases (121). In patients with satellite lesions, the same *NRAS* mutations are shared among anatomically separate lesions, which indicates that they originate from a common ancestor (122). The clonal relationship between discrete lesions indicates that the mutated progenitor cells can move apart and found anatomically separated, discrete lesions. Satellites are typically present at birth, and they continue to develop until age five; their continued emergence is linked to the risk of developing neurocutaneous melanosis (123). These findings

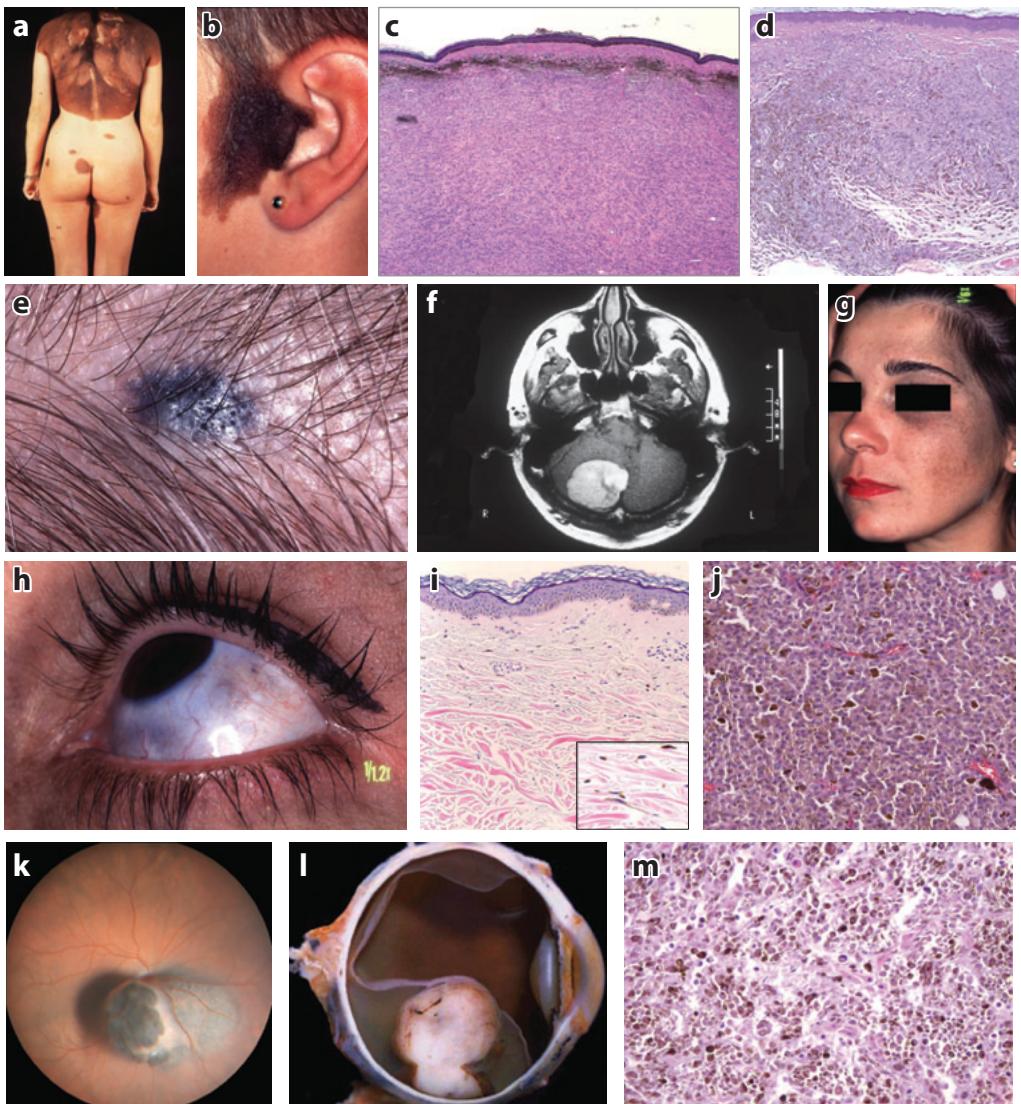


Figure 5

Clinical and histologic presentations of melanocytic neoplasms originating from melanocytes not associated with epithelial structures. (a) Giant congenital nevus with multiple satellite lesions. (b) Small congenital nevus with hypertrichosis. (c) Dense proliferation of melanocytes in the dermis. The superficial melanocytes are hyperpigmented. (d) Blue nevus: proliferation of pigmented melanocytes in the dermis interspersed with pigment-laden macrophages (melanophages). (e) Blue nevus on the scalp. (f) Melanocytoma of the central nervous system: hyperdense mass in the cerebellum. (g) Nevus of Ota: segmental hyperpigmentation in the innervation segment of the first branch of the trigeminal nerve. (h) Nevus of Ota: hyperpigmentation of the conjunctiva due to increased numbers of melanocytes. (i) Nevus of Ota: subtle increase of spindled melanocytes in the dermis, shown at higher magnification in the inset at the bottom right. (j) Melanocytoma: proliferation of epithelioid pigmented melanocytes interspersed with melanophages. (k) Fundoscopic view of a uveal melanoma. (l) Uveal melanoma growing as a mushroom-shaped tumor that detaches the retina. (m) Uveal melanoma: proliferation of atypical epithelioid pigmented melanocytes interspersed with melanophages.

indicate that satellites form from descendants of a single mutated progenitor cell that move along the axis of the neural crest and obtain access to different segments, from which they migrate and clonally expand. The founding *NRAS* mutation results in an expanded progenitor pool that exceeds the receiving capacity of the destination epithelia, so that excess cells pile up in the dermis and other tissues along the way (**Figure 5c**). A similar phenomenon occurs with hypermorphic mutations in *GNAQ* and *GNA11* in mouse models (124). Other oncogenic alterations reported in congenital nevi involve kinase fusions of *BRAF*, which have been reported in individual cases of congenital nevi (125). Patients with larger congenital nevi are also at increased risk for other neoplasms, including liposarcomas and rhabdomyosarcomas, which indicates that the cell of origin may retain pluripotency.

Blue Nevi and Related Neoplasms

Blue nevi comprise a group of neoplasms characterized by the proliferation of dendritic, spindled, ovoid, or epithelioid melanocytes in tissues without any significant epithelial involvement (**Figure 5d**) (126). Although most involve the skin (**Figure 5e**), they can be encountered in many other organs, such as the lung, intestinal tract, prostate, and CNS (**Figure 5f**), where they are called melanocytomas. Blue nevi can be acquired or congenital. The latter category includes a range of conditions, including Mongolian spot, nevus of Ito, and nevus of Ota (**Figure 5g**), all of which present as hyperpigmented patches comprising paucicellular proliferations of pigmented melanocytes in the dermis (**Figure 5b**). Because blue nevi spare the epidermis, they are also referred to as dermal melanocytoses. Nevi of Ota and Ito have a segmental pattern involving branches of the trigeminal nerve and cervical nerves, respectively. Their distribution along innervation segments and their cell morphology hint at a close relationship with Schwann cells, supporting the view that they arise from melanocytes that develop through the ventro-

medial pathway (see the sidebar) (127). Lesions with prominent areas of Schwann cell differentiation are termed neurocrystic hamartomas.

The term blue nevus, in the strict sense, refers to discrete, nonsegmental lesions that can vary in size from less than a centimeter to several centimeters in congenital lesions. Blue nevi express a range of cellularity, from paucicellular lesions similar to dermal melanocytoses to highly cellular nodules referred to as cellular blue nevi.

The molecular characteristics shared among neoplasms of this family are somatic mutations of *GNAQ* or *GNA11*, two closely related α -subunits of the G α q family (20, 21). Mutations in *GNAQ* and *GNA11* occur at the glutamine at position 209 or, in approximately 5% of cases, at the arginine at position 183. The Q209 mutations completely abrogate the subunits' GTPase activity, locking them in a GTP-bound, constitutively activated state. The R183 mutations maintain some residual enzymatic activity and thus are comparatively weakly activating. Mutations in these genes occur in a mutually exclusive pattern and are found in the majority of blue nevi, including the segmental dermal melanocytoses. *GNAQ* and *GNA11* convey signals from a broad range of G protein-coupled receptors. In melanocytes, endothelin receptors utilize G α q family members. Because endothelin signaling is critical for melanocyte development, in particular for melanocytes developing through the ventromedial pathway, the mutations likely simulate sustained endothelin signaling. To date, recurrent mutations in these genes have not been found in other neoplastic conditions. Their downstream effectors include phospholipase C, which releases two potent second messengers (diacylglycerol and inositol 3-phosphate) from membrane phospholipids, which activate protein kinase C, the MAP kinase pathway, and other signaling cascades.

Blue nevi can also arise as part of Carney's complex, a tumor predisposition syndrome caused by germ-line loss-of-function mutations in *PRKAR1A*, one of the two regulatory subunits of protein kinase A (PKA), resulting

in increased PKA activity (128). Blue nevi in individuals with Carney's complex often are composed of epithelioid melanocytes. The spectrum of somatic mutations in blue nevi arising in the context of Carney's complex is currently not known.

As with other melanocytic nevi, blue nevi have a malignant counterpart, referred to as blue nevus-like melanoma or sometimes, self-contradictorily, as malignant blue nevus. These melanomas can arise within blue nevi as a contiguous, morphologically benign lesion or can occur de novo. Although blue nevus-like melanomas are rare, blue nevi are also much less frequent than acquired nevi, and the ratio of benign to malignant lesions may be within the range of that in other categories. The genetic alterations in blue nevus-like melanoma have not been systematically studied but have similarities with those in blue nevi and uveal melanoma, such as frequent mutations in *GNAQ* or *GNA11* and losses of chromosome 3 (21, 129). Blue nevus-like melanomas also share with uveal melanoma a propensity to metastasize to the liver and bone (130).

Melanocytoma of the Central Nervous System

Melanocytomas of the CNS are primary melanocytic neoplasms that arise from autochthonous melanocytes of the leptomeninges and are typically discovered when they cause focal neurological symptoms by impinging on critical structures. They have strong morphological similarities to the spectrum of blue nevi described above and, like blue nevi, frequently harbor mutations of *GNAQ* or *GNA11* (131). Similarly to blue nevi, melanocytomas have a malignant counterpart with morphological features similar to those of blue nevus-like melanoma (132).

Uveal Melanoma

Uveal melanoma is the most common intraocular cancer and accounts for approximately 5%

of all melanomas in the United States. Unlike that of melanomas on the skin, its incidence has been stable, at approximately 5 per million (133). With 10-year survival rates at 50%, it ranks among the most lethal presentations of melanoma. The cells of origin are interstitial melanocytes that are found abundantly in the choroid, ciliary body, and iris of the eye, which together comprise the uveal tract. They are neural crest derived, as opposed to the pigmented cells of the retinal pigment epithelium, which are derived from the anterior neural plate. Unlike melanocytes of the skin, uveal melanocytes do not reside within an epithelium.

Clinical and histopathological evidence suggests that most uveal melanomas arise from uveal nevi. The prevalence of uveal nevi is between 4.6% and 7.9% in Caucasians, and on the basis of the number of reported uveal melanomas, the risk of transformation has been estimated to be 1 in 8,845 nevi per year (134). Mutations in *GNAQ* or *GNA11* occur in approximately 85% of uveal melanomas and are mutually exclusive. They are considered early events, because they are not associated with outcome and can already be detected in uveal nevi (21). Loss-of-function mutations in the deubiquitinase *BAP1* are found in 49% of uveal melanomas, and the frequency is significantly higher (85%) in tumors that become metastatic (60). A major selective force for the frequent loss of chromosome 3, a strong negative prognostic indicator in uveal melanoma (135), is the elimination of the remaining wild-type allele of *BAP1*. The precise mechanisms underlying how elimination of *BAP1* function promotes uveal melanoma growth are not fully understood. *BAP1* is a nuclear deubiquitinase that interacts with several proteins involved in chromatin modification, including HCF1, UDP-glucose-dependent OGT, and the Polycomb group proteins ASXL1 and ASXL2. The disruption of this complex by loss of *BAP1* is thought to result in altered histone modifications and a deregulated gene expression pattern (136). Gene expression profiling of uveal melanomas identified two separate

classes. Class 1 tumors have a better prognosis, whereas class 2 tumors have a significantly increased risk of metastasis. The latter pattern is closely associated with *BAP1* loss and may be a direct consequence of the ensuing epigenetic deregulation (137).

Another gene that is recurrently mutated in uveal melanoma is the splicing factor *SF3B1*. Mutations in this gene occur in 19% of uveal melanomas, primarily in class 1 tumors, and thus primarily affect melanomas without *BAP1* mutations (138). These mutations affect codon 625 and are typically heterozygous. It remains to be determined whether their effect is caused by gained or altered function or

by a dominant-negative effect. In zebra fish, germ-line loss-of-function mutations of *SF3B1* result in a pigmentation phenotype caused by a failure of neural crest development. Loss of *SF3B1* in this model leads to missplicing of critical neural crest transcription factors such as SOX10 and TFAP2A. Re-expression of properly spliced versions of these proteins can rescue the neural crest defect (139). Although the link between *SF3B1* function and neural crest development is intriguing, the specific *SF3B1* mutations found in uveal melanoma do not appear to be associated with missplicing of any of these specific transcription factors (138).

SUMMARY POINTS

1. There are multiple distinct categories of melanocytic neoplasms that differ in clinical and histologic presentation, cell of origin, age of onset, ethnic variation, pathogenetic role of UV radiation, predisposing germ-line alterations, patterns of somatic mutations, type of genomic instability, and preferential sites of metastasis.
2. Melanocytic neoplasms are initiated by somatic mutations that activate oncogenes. These mutations are not sufficient to form melanoma but in some categories of melanocytic neoplasia induce benign neoplasms, termed melanocytic nevi. The proliferation of melanocytes within nevi is constrained by a multiplicity of factors, including cell cycle checkpoints, telomere length, secreted factors, and the immune system. Some melanoma categories do not have a distinctively recognizable benign precursor stage.
3. Subsequent genetic alterations override the tumor-suppressive mechanisms that restrain tumor cell populations with imitating oncogenic alterations and lead to the progressive evolution of cells with an increasingly malignant phenotype. The subsequent genetic alterations differ among categories of melanocytic neoplasms.
4. In concert, the somatic mutations disrupt essential signaling pathways controlling cell proliferation, growth, motility, stromal interactions, differentiation status, and interaction with the immune system, giving rise to distinct phenotypic presentations of neoplasms.
5. Melanomas on the sun-exposed skin harbor a high burden of somatic mutations with the signature of UV radiation and begin as an intraepidermal growth, indicating that they arise from melanocytes within the epidermis.
6. Melanomas of the uveal tract of the eye and intradermal melanocytic proliferations such as blue nevi and related neoplasms arise from melanocytes not associated with epithelia and have distinct sets of genetic alterations.
7. UV radiation can cause mutations by direct interaction with DNA or indirectly through the generation of ROS. An increased ratio of pheomelanin to eumelanin may increase the mutation rate via ROS and thereby increase melanoma risk.

8. Separating categories of melanocytic neoplasms on the basis of reproducible associations between pathogenetically relevant genetic alterations and clinical phenotypes leads to an improved disease classification, which will facilitate future research and clinical management.

DISCLOSURE STATEMENT

The author is not aware of any affiliations, memberships, funding, or financial holdings that might be perceived as affecting the objectivity of this review.

ACKNOWLEDGMENTS

This work was supported by NIH grants R01-CA131524, R01-CA142873, and P01-CA025874. I would like to thank Drs. Daniel Pinkel, Philip LeBoit, Iwei Yeh, Hunter Shain, Thomas Botton, and Scott Dalton for critically reading the manuscript and providing helpful suggestions and Drs. Marc Rosenblum and David Abramson and the Department of Dermatology of the University of Würzburg, Germany, for providing clinical images. The amount of literature on this topic is enormous, and I apologize to my colleagues whose works I did not cite because of space limitations.

LITERATURE CITED

1. LeBoit PE, Burg G, Weedon D, Sarasin A, eds. 2006. *World Health Organization Classification of Tumours: Pathology and Genetics of Skin Tumours*. Lyon, Fr.: IARC Press. <http://www.iarc.fr/en/publications/pdfs-online/pat-gen/bb6/>
2. Clark WH, Elder DE, Van Horn M. 1986. The biologic forms of malignant melanoma. *Hum. Pathol.* 17:443–50
3. Weyers W, Euler M, Diaz-Casajo C, Schill WB, Bonczkowitz M. 1999. Classification of cutaneous malignant melanoma: a reassessment of histopathologic criteria for the distinction of different types. *Cancer* 86:288–99
4. Davies H, Bignell GR, Cox C, Stephens P, Edkins S, et al. 2002. Mutations of the *BRAF* gene in human cancer. *Nature* 417:949–54
5. Maldonado JL, Fridlyand J, Patel H, Jain AN, Busam K, et al. 2003. Determinants of *BRAF* mutations in primary melanoma. *J. Natl. Cancer Inst.* 95:1878–90
6. Curtin JA, Fridlyand J, Kageshita T, Patel HN, Busam KJ, et al. 2005. Distinct sets of genetic alterations in melanoma. *N. Engl. J. Med.* 353:2135–47
7. Viros A, Fridlyand J, Bauer J, Lasithiotakis K, Garbe C, et al. 2008. Improving melanoma classification by integrating genetic and morphologic features. *PLoS Med.* 5:e120
8. Liu W, Kelly JW, Trivett M, Murray WK, Dowling JP, et al. 2007. Distinct clinical and pathological features are associated with the *BRAF*^{T1799A(V600E)} mutation in primary melanoma. *J. Investigig. Dermatol.* 127:900–5
9. Purdue MP, From L, Armstrong BK, Kricker A, Gallagher RP, et al. 2005. Etiologic and other factors predicting nevus-associated cutaneous malignant melanoma. *Cancer Epidemiol. Biomark. Prev.* 14:2015–22
10. Lachiewicz AM, Berwick M, Wiggins CL, Thomas NE. 2008. Epidemiologic support for melanoma heterogeneity using the surveillance, epidemiology, and end results program. *J. Investigig. Dermatol.* 128:243–45
11. Tsao H, Goel V, Wu H, Yang G, Haluska FG. 2004. Genetic interaction between *NRAS* and *BRAF* mutations and *PTEN/MMAC1* inactivation in melanoma. *J. Investigig. Dermatol.* 122:337–41

12. Krauthammer M, Kong Y, Ha BH, Evans P, Bacchicocchi A, et al. 2012. Exome sequencing identifies recurrent somatic *RAC1* mutations in melanoma. *Nat. Genet.* 44:1006–14
13. Whiteman DC, Watt P, Purdie DM, Hughes MC, Hayward NK, Green AC. 2003. Melanocytic nevi, solar keratoses, and divergent pathways to cutaneous melanoma. *J. Natl. Cancer Inst.* 95:806–12
14. Pollock PM, Harper UL, Hansen KS, Yudt LM, Stark M, et al. 2003. High frequency of *BRAF* mutations in nevi. *Nat. Genet.* 33:19–20
15. Lin J, Takata M, Murata H, Goto Y, Kido K, et al. 2009. Polyclonality of *BRAF* mutations in acquired melanocytic nevi. *J. Natl. Cancer Inst.* 101:1423–27
16. Yancovitz M, Litterman A, Yoon J, Ng E, Shapiro RL, et al. 2012. Intra- and inter-tumor heterogeneity of *BRAFV600E* mutations in primary and metastatic melanoma. *PLoS ONE* 7:e29336
17. Yeh I, von Deimling A, Bastian BC. 2013. Clonal *BRAF* mutations in melanocytic nevi and initiating role of *BRAF* in melanocytic neoplasia. *J. Natl. Cancer Inst.* 105:917–19
18. Carr J, Mackie RM. 1994. Point mutations in the N-*ras* oncogene in malignant melanoma and congenital naevi. *Br. J. Dermatol.* 131:72–77
19. Bastian BC, LeBoit PE, Pinkel D. 2000. Mutations and copy number increase of *HRAS* in Spitz nevi with distinctive histopathological features. *Am. J. Pathol.* 157:967–72
20. Van Raamsdonk CD, Bezrookove V, Green G, Bauer J, Gaugler L, et al. 2009. Frequent somatic mutations of *GNAQ* in uveal melanoma and blue naevi. *Nature* 457:599–602
21. Van Raamsdonk CD, Griewank KG, Crosby MB, Garrido MC, Vemula S, et al. 2010. Mutations in *GNA11* in uveal melanoma. *N. Engl. J. Med.* 363:2191–99
22. Michaloglou C, Vredeveld LCW, Soengas MS, Denoyelle C, Kuilman T, et al. 2005. *BRAF^{E600}*-associated senescence-like cell cycle arrest of human naevi. *Nature* 436:720–24
23. Tran SL, Haferkamp S, Scurr LL, Gowrishankar K, Becker TM, et al. 2012. Absence of distinguishing senescence traits in human melanocytic nevi. *J. Investigig. Dermatol.* 132:2226–34
24. Kuilman T, Michaloglou C, Mooi WJ, Peper DS. 2010. The essence of senescence. *Genes Dev.* 24:2463–79
25. Kamijo T, Zindy F, Roussel MF, Quelle DE, Downing JR, et al. 1997. Tumor suppression at the mouse *INK4a* locus mediated by the alternative reading frame product p19^{ARF}. *Cell* 91:649–59
26. Serrano M, Lin AW, McCurrach ME, Beach D, Lowe SW. 1997. Oncogenic *ras* provokes premature cell senescence associated with accumulation of p53 and p16^{INK4a}. *Cell* 88:593–602
27. Kamb A. 1994. Role of a cell cycle regulator in hereditary and sporadic cancer. *Cold Spring Harb. Symp. Quant. Biol.* 59:39–47
28. Hussussian CJ, Struewing JP, Goldstein AM, Higgins PA, Ally DS, et al. 1994. Germline *p16* mutations in familial melanoma. *Nat. Genet.* 8:15–21
29. Fitzgerald MG, Harkin DP, Silva-Arrieta S, MacDonald DJ, Lucchini LC, et al. 1996. Prevalence of germ-line mutations in *p16*, p19^{ARF}, and *CDK4* in familial melanoma: analysis of a clinic-based population. *Proc. Natl. Acad. Sci. USA* 93:8541–45
30. Goldstein AM, Chan M, Harland M, Gillanders EM, Hayward NK, et al. 2006. High-risk melanoma susceptibility genes and pancreatic cancer, neural system tumors, and uveal melanoma across GenoME. *Cancer Res.* 66:9818–28
31. Zuo L, Weger J, Yang Q, Goldstein AM, Tucker MA, et al. 1996. Germline mutations in the *p16^{INK4a}* binding domain of *CDK4* in familial melanoma. *Nat. Genet.* 12:97–99
32. Randerson-Moor JA, Harland M, Williams S, Cuthbert-Heavens D, Sheridan E, et al. 2001. A germline deletion of p14^{ARF} but not *CDKN2A* in a melanoma–neural system tumour syndrome family. *Hum. Mol. Genet.* 10:55–62
33. Florell SR, Meyer LJ, Boucher KM, Porter-Gill PA, Hart M, et al. 2004. Longitudinal assessment of the nevus phenotype in a melanoma kindred. *J. Investigig. Dermatol.* 123:576–82
34. Bishop JAN, Wachsmuth RC, Harland M, Bataille V, Pinney E, et al. 2000. Genotype/phenotype and penetrance studies in melanoma families with germline *CDKN2A* mutations. *J. Investigig. Dermatol.* 114:28–33
35. Gorgoulis VG, Vassiliou L-VF, Karakaidos P, Zacharatos P, Kotsinas A, et al. 2005. Activation of the DNA damage checkpoint and genomic instability in human precancerous lesions. *Nature* 434:907–13

36. Bastian BC, Olshen AB, LeBoit PE, Pinkel D. 2003. Classifying melanocytic tumors based on DNA copy number changes. *Am. J. Pathol.* 163:1765–70
37. Aird KM, Zhang G, Li H, Tu Z, Bitler BG, et al. 2013. Suppression of nucleotide metabolism underlies the establishment and maintenance of oncogene-induced senescence. *Cell Rep.* 3:1252–65
38. Kuilman T, Michaloglou C, Vredeveld LCW, Douma S, van Doorn R, et al. 2008. Oncogene-induced senescence relayed by an interleukin-dependent inflammatory network. *Cell* 133:1019–31
39. Maldonado JL, Timmerman L, Fridlyand J, Bastian BC. 2004. Mechanisms of cell-cycle arrest in Spitz nevi with constitutive activation of the MAP-kinase pathway. *Am. J. Pathol.* 164:1783–87
40. Ramirez RD, D’Atri S, Pagani E, Faraggiana T, Lacal PM, et al. 1999. Progressive increase in telomerase activity from benign melanocytic conditions to malignant melanoma. *Neoplasia* 1:42–49
41. Rudolph P, Schubert C, Tamm S, Heidorn K, Hauschild A, et al. 2000. Telomerase activity in melanocytic lesions: a potential marker of tumor biology. *Am. J. Pathol.* 156:1425–32
42. Bastian BC, Kashani-Sabet M, Hamm H, Godfrey T, Moore DH, et al. 2000. Gene amplifications characterize acral melanoma and permit the detection of occult tumor cells in the surrounding skin. *Cancer Res.* 60:1968–73
43. North JP, Kageshita T, Pinkel D, LeBoit PE, Bastian BC. 2008. Distribution and significance of occult intraepidermal tumor cells surrounding primary melanoma. *J. Investig. Dermatol.* 128:2024–30
44. Horn S, Figl A, Rachakonda PS, Fischer C, Sucker A, et al. 2013. *TERT* promoter mutations in familial and sporadic melanoma. *Science* 339:959–61
45. Huang FW, Hodis E, Xu MJ, Kryukov GV, Chin L, Garraway LA. 2013. Highly recurrent *TERT* promoter mutations in human melanoma. *Science* 339:957–59
46. Bastian BC. 2003. Hypothesis: a role for telomere crisis in spontaneous regression of melanoma. *Arch. Dermatol.* 139:667–68
47. Sekulic A, Colgan MB, Davis MDP, DiCaudo DJ, Pittelkow MR. 2010. Activating *BRAF* mutations in eruptive melanocytic naevi. *Br. J. Dermatol.* 163:1095–98
48. Kang T-W, Yevsa T, Woller N, Hoenicke L, Wuestefeld T, et al. 2011. Senescence surveillance of pre-malignant hepatocytes limits liver cancer development. *Nature* 479:547–51
49. Hodis E, Watson IR, Kryukov GV, Arold ST, Imielinski M, et al. 2012. A landscape of driver mutations in melanoma. *Cell* 150:251–63
50. Ceol CJ, Houvras Y, Jane-Valbuena J, Bilodeau S, Orlando DA, et al. 2011. The histone methyltransferase SETDB1 is recurrently amplified in melanoma and accelerates its onset. *Nature* 471:513–17
51. MacGregor S, Montgomery GW, Liu JZ, Zhao ZZ, Henders AK, et al. 2011. Genome-wide association study identifies a new melanoma susceptibility locus at 1q21.3. *Nat. Genet.* 43:1114–18
52. McHugh JB, Fullen DR, Ma L, Kleer CG, Su LD. 2007. Expression of polycomb group protein EZH2 in nevi and melanoma. *J. Cutan. Pathol.* 34:597–600
53. Kapoor A, Goldberg MS, Cumberland LK, Ratnakumar K, Segura MF, et al. 2010. The histone variant macroH2A suppresses melanoma progression through regulation of CDK8. *Nature* 468:1105–9
54. Hoon DSB, Spugnardi M, Kuo C, Huang SK, Morton DL, Taback B. 2004. Profiling epigenetic inactivation of tumor suppressor genes in tumors and plasma from cutaneous melanoma patients. *Oncogene* 23:4014–22
55. Liu S, Ren S, Howell P, Fodstad O, Riker AI. 2008. Identification of novel epigenetically modified genes in human melanoma via promoter methylation gene profiling. *Pigment Cell Melanoma Res.* 21:545–58
56. Shen L, Kondo Y, Guo Y, Zhang J, Zhang L, et al. 2007. Genome-wide profiling of DNA methylation reveals a class of normally methylated CpG island promoters. *PLoS Genet.* 3:e181
57. Lian CG, Xu Y, Ceol C, Wu F, Larson A, et al. 2012. Loss of 5-hydroxymethylcytosine is an epigenetic hallmark of melanoma. *Cell* 150:1135–46
58. Ko M, Huang Y, Jankowska AM, Pape UJ, Tahiliani M, et al. 2010. Impaired hydroxylation of 5-methylcytosine in myeloid cancers with mutant *TET2*. *Nature* 468:839–43
59. Figueroa ME, Abdel-Wahab O, Lu C, Ward PS, Patel J, et al. 2010. Leukemic *IDH1* and *IDH2* mutations result in a hypermethylation phenotype, disrupt *TET2* function, and impair hematopoietic differentiation. *Cancer Cell* 18:553–67
60. Harbour JW, Onken MD, Roberson EDO, Duan S, Cao L, et al. 2010. Frequent mutation of *BAP1* in metastasizing uveal melanomas. *Science* 330:1410–13

61. Wiesner T, Obenauf AC, Murali R, Fried I, Griewank KG, et al. 2011. Germline mutations in *BAP1* predispose to melanocytic tumors. *Nat. Genet.* 43:1018–21
62. Wiesner T, Murali R, Fried I, Cerroni L, Busam K, et al. 2012. A distinct subset of atypical Spitz tumors is characterized by *BRAF* mutation and loss of BAP1 expression. *Am. J. Surg. Pathol.* 36:818–30
63. Reimer RR, Clark WH, Greene MH, Ainsworth AM, Fraumeni JF. 1978. Precursor lesions in familial melanoma: a new genetic preneoplastic syndrome. *JAMA* 239:744–46
64. Piepkorn M, Meyer LJ, Goldgar D, Seuchter SA, Cannon-Albright LA, et al. 1989. The dysplastic melanocytic nevus: a prevalent lesion that correlates poorly with clinical phenotype. *J. Am. Acad. Dermatol.* 20:407–15
65. Ackerman AB. 1988. What naevus is dysplastic, a syndrome and the commonest precursor of malignant melanoma? A riddle and an answer. *Histopathology* 13:241–56
66. Duncan LM, Berwick M, Bruijn JA, Byers HR, Mihm MC, Barnhill RL. 1993. Histopathologic recognition and grading of dysplastic melanocytic nevi: an interobserver agreement study. *J. Investigig. Dermatol.* 100:S318–21
67. Ackerman AB, Milde P. 1992. Naming acquired melanocytic nevi. Common and dysplastic, normal and atypical, or Unna, Miescher, Spitz, and Clark? *Am. J. Dermatopathol.* 14:447–53
68. Goldgar DE, Cannon-Albright LA, Meyer LJ, Pipekorn MW, Zone JJ, Skolnick MH. 1991. Inheritance of nevus number and size in melanoma and dysplastic nevus syndrome kindreds. *J. Natl. Cancer Inst.* 83:1726–33
69. Tucker MA, Halpern A, Holly EA, Hartge P, Elder DE, et al. 1997. Clinically recognized dysplastic nevi. A central risk factor for cutaneous melanoma. *JAMA* 277:1439–44
70. Hussein MRA-E, Wood GS. 2002. Molecular aspects of melanocytic dysplastic nevi. *J. Mol. Diagn.* 4:71–80
71. Whiteman DC, Pavan WJ, Bastian BC. 2011. The melanomas: a synthesis of epidemiological, clinical, histopathological, genetic, and biological aspects, supporting distinct subtypes, causal pathways, and cells of origin. *Pigment Cell Melanoma Res.* 24:879–97
72. Busam KJ, Murali R, Pulitzer M, McCarthy SW, Thompson JF, et al. 2009. Atypical spitzoid melanocytic tumors with positive sentinel lymph nodes in children and teenagers, and comparison with histologically unambiguous and lethal melanomas. *Am. J. Surg. Pathol.* 33:1386–95
73. Ludgate MW, Fullen DR, Lee J, Lowe L, Bradford C, et al. 2009. The atypical Spitz tumor of uncertain biologic potential. *Cancer* 115:631–41
74. Morton DL, Thompson JF, Cochran AJ, Mozzillo N, Elashoff R, et al. 2006. Sentinel-node biopsy or nodal observation in melanoma. *N. Engl. J. Med.* 355:1307–17
75. Lange JR, Palis BE, Chang DC, Soong S-J, Balch CM. 2007. Melanoma in children and teenagers: an analysis of patients from the National Cancer Data Base. *J. Clin. Oncol.* 25:1363–68
76. Crombie IK. 1979. Variation of melanoma incidence with latitude in North America and Europe. *Br. J. Cancer* 40:774–81
77. Elwood JM, Gallagher RP. 1998. Body site distribution of cutaneous malignant melanoma in relationship to patterns of sun exposure. *Int. J. Cancer* 78:276–80
78. Pleasance ED, Cheetham RK, Stephens PJ, McBride DJ, Humphray SJ, et al. 2009. A comprehensive catalogue of somatic mutations from a human cancer genome. *Nature* 463:191–96
79. Mocellin S, Verdi D, Nitti D. 2009. DNA repair gene polymorphisms and risk of cutaneous melanoma: a systematic review and meta-analysis. *Carcinogenesis* 30:1735–43
80. Nikolaev SI, Rimoldi D, Iseli C, Valsesia A, Robyr D, et al. 2012. Exome sequencing identifies recurrent somatic *MAP2K1* and *MAP2K2* mutations in melanoma. *Nat. Genet.* 44:133–39
81. Besaratinia A, Pfeifer GP. 2008. Sunlight ultraviolet irradiation and *BRAF* V600 mutagenesis in human melanoma. *Hum. Mutat.* 29:983–91
82. Noonan FP, Zaidi MR, Wolnicka-Glubisz A, Anver MR, Bahn J, et al. 2012. Melanoma induction by ultraviolet A but not ultraviolet B radiation requires melanin pigment. *Nat. Commun.* 3:884
83. Agrup G, Hansson C, Rorsman H, Rosengren E. 1982. The effect of cysteine on oxidation of tyrosine, dopa, and cysteinyldopas. *Arch. Dermatol. Res.* 272:103–15
84. Simon JD, Peles DN. 2010. The red and the black. *Acc. Chem. Res.* 43:1452–60

85. Song X, Mosby N, Yang J, Xu A, Abdel-Malek Z, Kadekaro AL. 2009. α -MSH activates immediate defense responses to UV-induced oxidative stress in human melanocytes. *Pigment Cell Melanoma Res.* 22:809–18
86. Valverde P, Healy E, Jackson I, Rees JL, Thody AJ. 1995. Variants of the melanocyte-stimulating hormone receptor gene are associated with red hair and fair skin in humans. *Nat. Genet.* 11:328–30
87. Brown KM, MacGregor S, Montgomery GW, Craig DW, Zhao ZZ, et al. 2008. Common sequence variants on 20q11.22 confer melanoma susceptibility. *Nat. Genet.* 40:838–40
88. Han J, Kraft P, Nan H, Guo Q, Chen C, et al. 2008. A genome-wide association study identifies novel alleles associated with hair color and skin pigmentation. *PLoS Genet.* 4:e1000074
89. Bishop DT, Demenais F, Iles MM, Harland M, Taylor JC, et al. 2009. Genome-wide association study identifies three loci associated with melanoma risk. *Nat. Genet.* 41:920–25
90. Falchi M, Bataille V, Hayward NK, Duffy DL, Bishop JAN, et al. 2009. Genome-wide association study identifies variants at 9p21 and 22q13 associated with development of cutaneous nevi. *Nat. Genet.* 41:915–19
91. Duffy DL, Zhao ZZ, Sturm RA, Hayward NK, Martin NG, Montgomery GW. 2010. Multiple pigmentation gene polymorphisms account for a substantial proportion of risk of cutaneous malignant melanoma. *J. Investig. Dermatol.* 130:520–28
92. Barrett JH, Iles MM, Harland M, Taylor JC, Aitken JF, et al. 2011. Genome-wide association study identifies three new melanoma susceptibility loci. *Nat. Genet.* 43:1108–13
93. Landi MT, Bauer J, Pfeiffer RM, Elder DE, Hulley B, et al. 2006. *MC1R* germline variants confer risk for *BRAF*-mutant melanoma. *Science* 313:521–22
94. Farnholz MC, Pike K, Pfeiffer RM, Tsang S, Rozenblum E, et al. 2008. *MC1R* variants increase risk of melanomas harboring *BRAF* mutations. *J. Investig. Dermatol.* 128:2485–90
95. Thomas NE, Kanetsky PA, Edmiston SN, Alexander A, Begg CB, et al. 2010. Relationship between germline *MC1R* variants and *BRAF*-mutant melanoma in a North Carolina population-based study. *J. Investig. Dermatol.* 130:1463–65
96. Hacker E, Hayward NK, Dumenil T, James MR, Whiteman DC. 2010. The association between *MC1R* genotype and *BRAF* mutation status in cutaneous melanoma: findings from an Australian population. *J. Investig. Dermatol.* 130:241–48
97. Menzies AM, Haydu LE, Visintin L, Carlino MS, Howle JR, et al. 2012. Distinguishing clinicopathologic features of patients with V600E and V600K *BRAF*-mutant metastatic melanoma. *Clin. Cancer Res.* 18:3242–49
98. Hacker E, Nagore E, Cerroni L, Woods SL, Hayward NK, et al. 2013. *NRAS* and *BRAF* mutations in cutaneous melanoma and the association with *MC1R* genotype: findings from Spanish and Austrian populations. *J. Invest. Dermatol.* 133:1027–33
99. Oláh J, Tóth-Molnár E, Kemény L, Csoma Z. 2013. Long-term hazards of neonatal blue light phototherapy. *Br. J. Dermatol.* 169:243–49
100. Bauer J, Buttner P, Wiecker TS, Luther H, Garbe C. 2005. Effect of sunscreen and clothing on the number of melanocytic nevi in 1,812 German children attending day care. *Am. J. Epidemiol.* 161:620–27
101. Green A, Autier P, Boniol M, Boyle P, Doré J-F, et al. (Int. Agency Res. Cancer Work. Group Artif. Ultrav. Light Skin Cancer). 2007. The association of use of sunbeds with cutaneous malignant melanoma and other skin cancers: a systematic review. *Int. J. Cancer* 120:1116–22
102. Whiteman DC, Watt P, Purdie DM, Hughes MC, Hayward NK, Green AC. 2003. Melanocytic nevi, solar keratoses, and divergent pathways to cutaneous melanoma. *J. Natl. Cancer Inst.* 95:806–12
103. Glatz-Krieger K, Pache M, Tapia C, Fuchs A, Savic S, et al. 2006. Anatomic site-specific patterns of gene copy number gains in skin, mucosal, and uveal melanomas detected by fluorescence in situ hybridization. *Virchows Arch.* 449:328–33
104. Curtin JA, Busam K, Pinkel D, Bastian BC. 2006. Somatic activation of KIT in distinct subtypes of melanoma. *J. Clin. Oncol.* 24:4340–46
105. Davison JM, Rosenbaum E, Barrett TL, Goldenberg D, Hoque MO, et al. 2005. Absence of V599E *BRAF* mutations in desmoplastic melanomas. *Cancer* 103:788–92
106. Saida T. 2000. Malignant melanoma on the sole: how to detect the early lesions efficiently. *Pigment Cell Res.* 13:135–39

107. Arrington JH 3rd, Reed RJ, Ichinose H, Krementz ET. 1977. Plantar lentiginous melanoma: a distinctive variant of human cutaneous malignant melanoma. *Am. J. Surg. Pathol.* 1:131–43
108. Chang AE, Karnell LH, Menck HR. 1998. The National Cancer Data Base report on cutaneous and noncutaneous melanoma: a summary of 84,836 cases from the past decade. *Cancer* 83:1664–78
109. Kuchelmeister C, Schaumburg-Lever G, Garbe C. 2000. Acral cutaneous melanoma in Caucasians: clinical features, histopathology and prognosis in 112 patients. *Br. J. Dermatol.* 143:275–80
110. McLaughlin CC, Wu XC, Jemal A, Martin HJ, Roche LM, Chen VW. 2005. Incidence of noncutaneous melanomas in the U.S. *Cancer* 103:1000–7
111. Beadling C, Jacobson-Dunlop E, Hodi FS, Le C, Warrick A, et al. 2008. *KIT* gene mutations and copy number in melanoma subtypes. *Clin. Cancer Res.* 14:6821–28
112. Van Dijk M, Sprenger S, Rombout P, Marres H, Kaanders J, et al. 2003. Distinct chromosomal aberrations in sinonasal mucosal melanoma as detected by comparative genomic hybridization. *Genes Chromosomes Cancer* 36:151–58
113. Park E, Yang S, Emley A, DeCarlo K, Richards J, Mahalingam M. 2012. Lack of correlation between immunohistochemical expression of CKIT and *KIT* mutations in atypical acral nevi. *Am. J. Dermatopathol.* 34:41–46
114. Alexeev V, Yoon K. 2006. Distinctive role of the cKit receptor tyrosine kinase signaling in mammalian melanocytes. *J. Investigig. Dermatol.* 126:1102–10
115. Monsel G, Ortonne N, Bagot M, Bensussan A, Dumaz N. 2009. c-Kit mutants require hypoxia-inducible factor 1 α to transform melanocytes. *Oncogene* 29:227–36
116. Huang S, Jean D, Luca M, Tainsky MA, Bar-Eli M. 1998. Loss of AP-2 results in downregulation of c-KIT and enhancement of melanoma tumorigenicity and metastasis. *EMBO J.* 17:4358–69
117. Carvajal RD, Antonescu CR, Wolchok JD, Chapman PB, Roman R-A, et al. 2011. KIT as a therapeutic target in metastatic melanoma. *JAMA* 305:2327–34
118. Watt AJ, Kotsis SV, Chung KC. 2004. Risk of melanoma arising in large congenital melanocytic nevi: a systematic review. *Plast. Reconstr. Surg.* 113:1968–74
119. Mancianti ML, Clark WH, Hayes FA, Herlyn M. 1990. Malignant melanoma simulants arising in congenital melanocytic nevi do not show experimental evidence for a malignant phenotype. *Am. J. Pathol.* 136:817–29
120. Bastian BC, Xiong J, Frieden IJ, Williams ML, Chou P, et al. 2002. Genetic changes in neoplasms arising in congenital melanocytic nevi: differences between nodular proliferations and melanomas. *Am. J. Pathol.* 161:1163–69
121. Bauer J, Curtin JA, Pinkel D, Bastian BC. 2007. Congenital melanocytic nevi frequently harbor *NRAS* mutations but no *BRAF* mutations. *J. Investigig. Dermatol.* 127:179–82
122. Kinsler VA, Thomas AC, Ishida M, Bulstrode NW, Loughlin S, et al. 2013. Multiple congenital melanocytic naevi and neurocutaneous melanosis are caused by post-zygotic mutations in codon 61 of *NRAS*. *J. Investigig. Dermatol.* 133:2229–36
123. Slutsky JB, Barr JM, Femia AN, Marghoob AA. 2010. Large congenital melanocytic nevi: associated risks and management considerations. *Semin. Cutan. Med. Surg.* 29:79–84
124. Van Raamsdonk CD, Fitch KR, Fuchs H, de Angelis MH, Barsh GS. 2004. Effects of G-protein mutations on skin color. *Nat. Genet.* 36:961–68
125. Dessars B, De Raeve LE, Housni HE, Debouck CJ, Sidon PJ, et al. 2007. Chromosomal translocations as a mechanism of *BRAF* activation in two cases of large congenital melanocytic nevi. *J. Investigig. Dermatol.* 127:1468–70
126. Murali R, McCarthy SW, Scolyer RA. 2009. Blue nevi and related lesions: a review highlighting atypical and newly described variants, distinguishing features and diagnostic pitfalls. *Adv. Anat. Pathol.* 16:365–82
127. Sherman L, Stocker KM, Morrison R, Ciment G. 1993. Basic fibroblast growth factor (bFGF) acts intracellularly to cause the transdifferentiation of avian neural crest-derived Schwann cell precursors into melanocytes. *Development* 118:1313–26
128. Kirschner LS, Carney JA, Pack SD, Taymans SE, Giatzakis C, et al. 2000. Mutations of the gene encoding the protein kinase A type I- α regulatory subunit in patients with the Carney complex. *Nat. Genet.* 26:89–92

129. Maize JC, McCalmont TH, Carlson JA, Busam KJ, Kutzner H, Bastian BC. 2005. Genomic analysis of blue nevi and related dermal melanocytic proliferations. *Am. J. Surg. Pathol.* 29:1214–20
130. Martin RCW, Murali R, Scolyer RA, Fitzgerald P, Colman MH, Thompson JF. 2009. So-called “malignant blue nevus.” *Cancer* 115:2949–55
131. Murali R, Wiesner T, Rosenblum M, Bastian B. 2012. *GNAQ* and *GNA11* mutations in melanocytomas of the central nervous system. *Acta Neuropathol.* 123:457–59
132. Brat DJ. 2010. Melanocytic neoplasms of the central nervous system. In *Practical Surgical Neuropathology*, ed. A Perry, DJ Brat, pp. 353–59. Philadelphia: Churchill Livingstone
133. Singh AD, Turell ME, Topham AK. 2011. Uveal melanoma: trends in incidence, treatment, and survival. *Ophthalmology* 118:1881–85
134. Singh AD, Kalyani P, Topham A. 2005. Estimating the risk of malignant transformation of a choroidal nevus. *Ophthalmology* 112:1784–89
135. Prescher G, Bornfeld N, Hirche H, Horsthemke B, Jöckel KH, Becher R. 1996. Prognostic implications of monosomy 3 in uveal melanoma. *Lancet* 347:1222–25
136. Dey A, Seshasayee D, Noubade R, French DM, Liu J, et al. 2012. Loss of the tumor suppressor BAP1 causes myeloid transformation. *Science* 337:1541–46
137. Onken MD, Ehlers JP, Worley LA, Makita J, Yokota Y, Harbour JW. 2006. Functional gene expression analysis uncovers phenotypic switch in aggressive uveal melanomas. *Cancer Res.* 66:4602–9
138. Harbour JW, Roberson EDO, Anbunathan H, Onken MD, Worley LA, Bowcock AM. 2013. Recurrent mutations at codon 625 of the splicing factor *SF3B1* in uveal melanoma. *Nat. Genet.* 45:133–35
139. An M, Henion PD. 2012. The zebrafish *sf3b1^{b460}* mutant reveals differential requirements for the *sf3b1* pre-mRNA processing gene during neural crest development. *Int. J. Dev. Biol.* 56:223–37
140. Sommer L. 2011. Generation of melanocytes from neural crest cells. *Pigment Cell Melanoma Res.* 24:411–21
141. Nishimura EK, Jordan SA, Oshima H, Yoshida H, Osawa M, et al. 2002. Dominant role of the niche in melanocyte stem-cell fate determination. *Nature* 416:854–60
142. Granter SR, McKee PH, Calonje E, Mihm MC Jr, Busam K. 2001. Melanoma associated with blue nevus and melanoma mimicking cellular blue nevus: a clinicopathologic study of 10 cases on the spectrum of so-called “malignant blue nevus.” *Am. J. Surg. Pathol.* 25:316–23
143. Lens MB, Newton-Bishop JA, Boon AP. 2005. Desmoplastic malignant melanoma: a systematic review. *Br. J. Dermatol.* 152:673–78
144. Busam KJ. 2011. Desmoplastic melanoma. *Clin. Lab. Med.* 31:321–30
145. Meier F, Will S, Ellwanger U, Schlagenhauff B, Schittek B, et al. 2002. Metastatic pathways and time courses in the orderly progression of cutaneous melanoma. *Br. J. Dermatol.* 147:62–70
146. Bradford PT, Goldstein AM, McMaster ML, Tucker MA. 2009. Acral lentiginous melanoma: incidence and survival patterns in the United States, 1986–2005. *Arch. Dermatol.* 145:427–34
147. Hu D-N, Yu G-P, McCormick SA, Schneider S, Finger PT. 2005. Population-based incidence of uveal melanoma in various races and ethnic groups. *Am. J. Ophthalmol.* 140:612e1–8
148. Moore-Olufemi S, Herzog C, Warneke C, Gershenwald JE, Mansfield P, et al. 2011. Outcomes in pediatric melanoma. *Ann. Surg.* 253:1211–15

Contents



Annual Review of
Pathology:
Mechanisms of
Disease

Volume 9, 2014

Glioblastoma: From Molecular Pathology to Targeted Treatment <i>Timothy F. Cloughesy, Webster K. Cavenee, and Paul S. Mischel</i>	1
Origin and Pathogenesis of Pelvic (Ovarian, Tubal, and Primary Peritoneal) Serous Carcinoma <i>Nilofar N. Nik, Russell Vang, Ie-Ming Shib, and Robert J. Kurman</i>	27
Oxygen Sensing, Hypoxia-Inducible Factors, and Disease Pathophysiology <i>Gregg L. Semenza</i>	47
The Influence of Innate and Adaptive Immune Responses on Atherosclerosis <i>Joseph L. Witztum and Andrew H. Lichtman</i>	73
The Pathogenesis of Chronic Lymphocytic Leukemia <i>Suping Zhang and Thomas J. Kipps</i>	103
Nox Enzymes and New Thinking on Reactive Oxygen: A Double-Edged Sword Revisited <i>J. David Lambeth and Andrew S. Neish</i>	119
Mechanisms of Autoimmune Thyroid Diseases: From Genetics to Epigenetics <i>Yaron Tomer</i>	147
Pathogenesis of Idiopathic Pulmonary Fibrosis <i>Paul J. Wolters, Harold R. Collard, and Kirk D. Jones</i>	157
The Multifaceted Functions of Neutrophils <i>Tanya N. Mayadas, Xavier Cullere, and Clifford A. Lowell</i>	181
The Intracellular Life of <i>Cryptococcus neoformans</i> <i>Carolina Coelho, Anamelia L. Bocca, and Arturo Casadevall</i>	219
The Molecular Pathology of Melanoma: An Integrated Taxonomy of Melanocytic Neoplasia <i>Boris C. Bastian</i>	239

Sjögren's Syndrome	
<i>Clio P. Mavragani and Haralampos M. Moutsopoulos</i>	273
MicroRNAs in Cancer	
<i>Gianpiero Di Leva, Michela Garofalo, and Carlo M. Croce</i>	287
IgG4-Related Disease	
<i>Vinay S. Mahajan, Hamid Mattoo, Vikram Deshpande, Shiv S. Pillai, and John H. Stone</i>	315
Gammaherpesviruses and Lymphoproliferative Disorders	
<i>Ethel Cesarman</i>	349

Indexes

Cumulative Index of Contributing Authors, Volumes 1–9	373
Cumulative Index of Article Titles, Volumes 1–9	377

Errata

An online log of corrections to *Annual Review of Pathology: Mechanisms of Disease* articles may be found at <http://www.annualreviews.org/errata/pathmechdis>



ANNUAL REVIEWS

It's about time. Your time. It's time well spent.

New From Annual Reviews:

Annual Review of Statistics and Its Application

Volume 1 • Online January 2014 • <http://statistics.annualreviews.org>

Editor: **Stephen E. Fienberg, Carnegie Mellon University**

Associate Editors: **Nancy Reid, University of Toronto**

Stephen M. Stigler, University of Chicago

The *Annual Review of Statistics and Its Application* aims to inform statisticians and quantitative methodologists, as well as all scientists and users of statistics about major methodological advances and the computational tools that allow for their implementation. It will include developments in the field of statistics, including theoretical statistical underpinnings of new methodology, as well as developments in specific application domains such as biostatistics and bioinformatics, economics, machine learning, psychology, sociology, and aspects of the physical sciences.

Complimentary online access to the first volume will be available until January 2015.

TABLE OF CONTENTS:

- *What Is Statistics?* Stephen E. Fienberg
- *A Systematic Statistical Approach to Evaluating Evidence from Observational Studies*, David Madigan, Paul E. Stang, Jesse A. Berlin, Martijn Schuemie, J. Marc Overhage, Marc A. Suchard, Bill Dumouchel, Abraham G. Hartzema, Patrick B. Ryan
- *The Role of Statistics in the Discovery of a Higgs Boson*, David A. van Dyk
- *Brain Imaging Analysis*, F. DuBois Bowman
- *Statistics and Climate*, Peter Guttorp
- *Climate Simulators and Climate Projections*, Jonathan Rougier, Michael Goldstein
- *Probabilistic Forecasting*, Tilmann Gneiting, Matthias Katzfuss
- *Bayesian Computational Tools*, Christian P. Robert
- *Bayesian Computation Via Markov Chain Monte Carlo*, Radu V. Craiu, Jeffrey S. Rosenthal
- *Build, Compute, Critique, Repeat: Data Analysis with Latent Variable Models*, David M. Blei
- *Structured Regularizers for High-Dimensional Problems: Statistical and Computational Issues*, Martin J. Wainwright

- *High-Dimensional Statistics with a View Toward Applications in Biology*, Peter Bühlmann, Markus Kalisch, Lukas Meier
- *Next-Generation Statistical Genetics: Modeling, Penalization, and Optimization in High-Dimensional Data*, Kenneth Lange, Jeanette C. Papp, Janet S. Sinsheimer, Eric M. Sobel
- *Breaking Bad: Two Decades of Life-Course Data Analysis in Criminology, Developmental Psychology, and Beyond*, Elena A. Erosheva, Ross L. Matsueda, Donatello Telesca
- *Event History Analysis*, Niels Keiding
- *Statistical Evaluation of Forensic DNA Profile Evidence*, Christopher D. Steele, David J. Balding
- *Using League Table Rankings in Public Policy Formation: Statistical Issues*, Harvey Goldstein
- *Statistical Ecology*, Ruth King
- *Estimating the Number of Species in Microbial Diversity Studies*, John Bunge, Amy Willis, Fiona Walsh
- *Dynamic Treatment Regimes*, Bibhas Chakraborty, Susan A. Murphy
- *Statistics and Related Topics in Single-Molecule Biophysics*, Hong Qian, S.C. Kou
- *Statistics and Quantitative Risk Management for Banking and Insurance*, Paul Embrechts, Marius Hofert

Access this and all other Annual Reviews journals via your institution at www.annualreviews.org.

ANNUAL REVIEWS | Connect With Our Experts

Tel: 800.523.8635 (US/CAN) | Tel: 650.493.4400 | Fax: 650.424.0910 | Email: service@annualreviews.org

