A Practical Approach to the Diagnosis of Melanocytic Lesions

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Melanocytic lesions are a common component of the everyday workload for a surgical pathologist. Although distinction between nevi and melanoma can usually be easily achieved using well-established morphologic criteria,1 a significant minority of these specimens can cause diagnostic difficulties. Often these cases fall into one of several well-defined scenarios in which the diagnostic difficulties are well recognized and have caused consternation for many generations of pathologists.2 In this review we have attempted to outline a very practical approach to the assessment of melanocytic lesions. We begin with a brief outline of the steps in the histologic interpretation of these lesions, which is best approached in a methodical manner, progressing from low-power examination to higher-power scrutiny, while at the same time taking into account any clues that are provided by the clinical history. We then outline some of the better-recognized scenarios in which the standard morphologic rules are broken, resulting in potential diagnostic traps. Following a discussion of several of the more controversial areas in this field, we provide a brief synopsis on the current state of ancillary techniques, including immunohistochemistry and molecular testing.

Context.—Melanocytic lesions are common in routine surgical pathology. Although the majority of these lesions can be confidently diagnosed using well-established morphologic criteria, there is a significant subset of lesions that can be diagnostically difficult. These can be a source of anxiety for patients, clinicians, and pathologists, and the potential consequences of a missed diagnosis of melanoma are serious.

Objective.—To provide a practical approach to the diagnosis of melanocytic lesions, including classic problem areas as well as suggestions for common challenges and appropriate incorporation of ancillary molecular techniques.

Finally, we discuss some of the practical issues faced in daily practice and describe our method of approaching them.

Data Sources.—Literature search using PubMed and Google Scholar, incorporating numerous search terms relevant to the particular section, combined with contemporaneous texts and lessons from personal experience.

Conclusions.—Although a subset of melanocytic lesions can be diagnostically challenging, the combination of a methodical approach to histologic assessment, knowledge of potential diagnostic pitfalls, opinions from trusted colleagues, and judicious use of ancillary techniques can help the pathologist navigate this difficult area.

CLUES FROM THE REQUEST FORM

Although the information provided on the clinical request form is typically minimal, important clues can still be gleaned from it. In particular, the age of the patient is a critical factor. The incidence of melanoma increases with age, with a median age of diagnosis in Australia of 65 years.3 Melanoma is very uncommon in patients younger than 10 years (with the exception of specific rare scenarios, such as lesions arising within large congenital melanocytic nevi), and mimics of melanoma, such as Spitz nevi, are much more frequent in this age group.4 The site of the lesion can also be of value. It is now well recognized that benign melanocytic nevi in certain areas of the body can display atypical morphologic features that might lead to a misdiagnosis of melanoma. The list of these special sites now includes acral locations, the genital skin, the milk line (from the axillae over the breast to the genitalia), the umbilicus, the flexural skin, the scalp, the ears, and the back and shoulder of elderly patients.5 If provided, the clinical impression of the lesion is also worth taking into consideration. A general rule of thumb is to be cautious about diagnosing melanoma when an experienced clinician has made a clinical diagnosis of a nevus.1 Conversely, however, it is not rare to counter a clinical diagnosis of melanoma if the histologic features are those of a benign lesion. If provided, the size of the lesion and information as to whether or not it has been sampled in its clinical entirety may have a significant impact on the approach to reporting the lesion.
CLUES FROM LOW-POWER EXAMINATION

The important low-power criteria can be summarized as size, symmetry, circumscription, and sun damage (with a small abuse of phonetics, we often advise trainees to remember the “4 S’s”). Small lesions (defined here as less than 4 mm in maximum dimension) are usually benign. Despite its simplicity, this rule of thumb is remarkably reproducible. Of course small melanomas do exist, and if the overwhelming balance of histologic features point toward malignancy that should be the diagnosis. However, mimics of melanoma (eg, pagetoid Spitz nevus, spindle cell nevus of Reed) should be carefully considered before a small lesion is diagnosed as melanoma. It is important to note that, depending on the plane of sectioning, we may not always be viewing the largest dimension. Ideally, the size of the lesion should be provided in the clinical information, but in the absence of this, the macroscopic description can be useful. A small acral lesion with a predominantly nested architecture is particularly reassuring, as the lentiginous melanomas that typically arise in these areas tend to be of at least moderate size before nest formation becomes a feature. Within the broad category of lesion size, depth might also be considered. Although congenital nevi, deep penetrating nevi, cellular blue nevi, and some Spitz nevi may extend deeply into the dermis, lesions that extend deeply but do not fall into any of these categories should be viewed with some suspicion.

Symmetry is also a feature of benign lesions. The most obvious aspect of symmetry is a comparison of each half of the lesion, divided by an imaginary vertical line placed at the midpoint. However, the assessment of symmetry in melanocytic lesions encompasses other aspects. The growth pattern, cellular density, and cytologic features should all be similar across equivalent horizontal levels of the lesion, with exception allowed for populations within adventitial dermis. Junctional nests should be of roughly equivalent size and be evenly distributed. The pigment distribution should be symmetrical, as should accompanying nonmelanocytic components such as inflammatory infiltrates and epidermal changes. Attention to these more subtle aspects of symmetry is often the first clue to a melanoma. Closely related to the concept of symmetry is the presence of good circumscription, which is perhaps best expressed in practical terms as readily identifiable edges to the intraepidermal component. The most convincing form of a well-circumscribed lesion is one that ends with an identical nest at both lateral borders. Junctional or compound melanocytic lesions arising in sun-damaged skin (as indicated by the presence of dermal elastosis) should be regarded with care, particularly if the junctional component is prominent. One observation that may be of use in this setting is the “umbrella sign,” whereby the elastosis is diminished under nevi because of their long-standing presence acting as a solar shield for the underlying dermis. Conversely, invasive melanomas tend to push down the elastotic layer.

CLUES FROM MEDIUM TO HIGH-POWER EXAMINATION

At medium-power examination, the growth pattern of the lesional melanocytes can be interrogated. In a nevus, junctional melanocytes are arranged in a predominantly nested pattern, with the nests being of even size and regular distribution. Conversely, in a melanoma the growth will present a more disorderly appearance. The nests may appear enlarged, uneven, and irregularly distributed, perhaps with some located at the sides of rete ridges or over the tops of dermal papillae. There may be areas of predominantly single-cell, lentiginous (ie, similar to the pattern seen in lentigo) growth, with extension down the epithelium of adnexal structures. There may be areas of confluence where large nests appear to grow into one another. This can be difficult to distinguish from the bridging between adjacent rete ridges that is associated with dysplastic nevi, but the confluent growth of a melanoma often extends over the tops of the dermal papillae, rather than solely connecting the bases of adjacent rete ridges. The presence of melanocytes scattered within higher levels of the epidermis (and occasionally follicular epithelium) in a pagetoid pattern (ie, resembling the pattern seen in Paget disease) is a classic clue for melanoma. This finding is of particular concern if it occurs across the breadth of the lesion, if the cells are cytologically atypical, or if there is evidence of proliferative activity within the pagetoid cells. For it to be useful as a criterion, it is important to be precise as to what constitutes true pagetoid scatter. The cells in question should be identifiable as melanocytes, they should not be in continuity with the junctional component, and they should be located above a line parallel to the skin surface located at the basal epidermal layer overlying the most superficial dermal papilla. In practice, some authors require melanocytes to be located at least as high as the granular layer before using this criterion to justify a diagnosis of melanoma. Transepidermal elimination of melanocytes has a similar appearance to pagetoid spread, but in this instance the melanocytes are seen as large aggregates within the epidermis, apparently in the process of being eliminated at the surface. This relatively uncommon finding can be seen in both benign and malignant lesions and as such has no particular discriminatory value.

In practice, it is sometimes useful to draw an imaginary line across the tops of the dermal papillae. A lesion in which there is no action above this line is likely to be benign, whereas a lesion with significant growth above this line should be viewed with concern. If a dermal component is present, it may also show an irregular disordered growth pattern. Expansile nodules that compress or distort surrounding structures are suspicious for melanoma, as is the thinning of the epidermis secondary to melanocyte growth, which has been referred to as epidermal consumption. High-power examination allows for the assessment of the cytologic features of the lesional cells, as well as the presence of mitotic figures. Benign lesions classically demonstrate a cytologic alteration of melanocytes at the deeper aspects of the lesion, whereby they display a more spindled, “neural” appearance with dispersal into the surrounding collagen. This phenomenon is usually colloquially referred to as maturation, although of course technically this is incorrect (mature melanocytes synthesize melanin and deliver it to keratinocytes via dendritic processes). Most melanomas will display some degree of cytologic atypia, manifested by enlarged, pleomorphic nuclei with a coarse, clumped chromatin pattern and large, prominent nucleoli. A unique cytologic quality that has been described in association with melanoma is the so-called pulvocytome, characterized by voluminous cytoplasm with a fine, dusty melanin pigment. Some melanomas on acral sites, sun-exposed skin, or mucous membranes are charac-
terized by a dendritic morphology with an irregular, hyperchromatic nucleus and long dendritic processes extending into the upper spinous layer. Mitoses can be found in up to 20% of benign nevi. They tend to be found more often in the upper aspects of the dermis and more frequently in nevi from younger patients, in lesions with a polypoid architecture, and in pregnant patients. In benign lesions they are present in small numbers. Mitoses occurring in numbers rendering them easy to identify, or in clusters, are more worrying for melanoma. The combination of mitotic figures and nuclear pleomorphism should be of particular concern.

**MALIGNANT FEATURES IN BENIGN LESIONS**

One of the main reasons that the assessment of melanocytic lesions is so difficult is that most of the criteria listed above are not absolute. Features that are traditionally associated with a malignant diagnosis can also be seen occasionally in the context of a nevus. Some of these scenarios are reproducible enough to be recognized as potential pitfalls, which the astute pathologist can avoid. Some of the better described of these are outlined here and in the following section.

**Disordered Junctional Growth in a Traumatized or Recurrent Nevus**

Regrowth of benign melanocytic proliferations (including dysplastic nevi and Spitz nevi) after excision (variably termed recurrent or persistent nevus) or following traumatization/excoriation can show a spectrum of histologic changes that overlap with those of melanoma. Nevi tend to clinically recur within a relatively shorter period of time (weeks to months), compared with recurrent melanomas (months to years). Similar appearances have also been reported in lesions without a history of surgery or recent trauma, described in this context as sclerosing nevus (although the authors who coined this term pointed out that they suspected at least some of the lesions had in fact been subject to some minor, unnoticed trauma or recurrent irritation). Analogous changes have been reported in the context of blistering disorders, as well as other inflammatory conditions including Stevens-Johnson syndrome and lichen sclerosus. Recognition of the background changes of lichen sclerosus is the key to avoiding the latter pitfall, and we agree with the advice that a search for these changes should be a reflex response when considering a diagnosis of melanoma on genital skin (especially in younger patients).

This group of lesions is characterized by a disordered junctional arrangement of melanocytic nests and single cells, with variably sized, irregular nests that often appear large and/or elongated. An element of cellular discohesion as well as enlargement of the individual melanocytes can accompany these changes. Pagetoid upward migration of melanocytes may be present. These changes, which have been referred to as pseudomelanoma, may be purely junctional (mimicking melanoma in situ) or extend to the superficial dermal component (mimicking invasive melanoma) (Figure 1, A and B). The presence of underlying fibrosis may be confused with regression within a melanoma. Often the atypical proliferation overlies an area of granulation tissue formation or scarring in the upper dermis, with residual banal nevus visible beneath. Identification of this classic trilayered appearance is very helpful in recognizing this phenomenon. In addition, the atypical proliferation should be limited to the area of damage, and the lesion as a whole should appear well circumscribed. In cases where the scarring is less obvious, subtle clues include surface hyperkeratosis or parakeratosis, effacement of rete ridges, and loss of elastin fibers within the dermis. A history of prior surgery or other traumatization is very helpful, and review of the previous biopsy specimen can be invaluable if it confirms the presence of a banal nevus. However, given that the histologic appearances of this phenomenon can be essentially indistinguishable from melanoma, if there remains doubt, complete excision of the lesion with a clear margin of normal skin is the most appropriate course.

**Mitoses in Nevi Removed During Pregnancy**

Dermal mitotic figures both are more common and are present in greater numbers in nevi removed from pregnant patients when compared with nevi from nonpregnant patients. In this setting, some authors have also described rounded clusters of large epithelioid melanocytes with prominent nucleoli, termed superficial micronodules of pregnancy. Some groups have also reported a degree of mild junctional atypia in nevi of pregnancy, including a lentiginous growth pattern, irregular or confluent nests, and cellular discohesion. Despite earlier suggestions to the contrary, studies have not demonstrated any significant effect of pregnancy on the size of nevi. A clinical history of pregnancy is clearly of value in establishing whether the presence of dermal mitoses can be attributed to this. Often this will require a phone call to the referring clinician, although clues may be found in the age (and sex!) of the patient and the presence of recent pregnancy-related screening tests in the pathology records. Although pregnancy may explain the presence of a few dermal mitotic figures, it should not be invoked as a reason to ignore other histologic features of malignancy. As well as nevi, melanomas also occur in pregnant patients, especially in an era where patients are not infrequently older than in previous generations.

**Pagetoid Spitz Nevi**

Some examples of early Spitz nevus can show a predominantly single-cell growth pattern limited to the epidermis, often with a significant element of suprabasal scatter mimicking the pagetoid spread of melanoma. As is typical of Spitz nevi, the lesional cells in these cases are usually large epithelioid or spindled with prominent nucleoli, which can add to their deceptive nature. The key to avoiding misdiagnosis is attention to the other aspects of the lesion, which should be small and symmetrical, with even hyperplasia of the lesional epidermis. One important clue is that the melanocytic proliferation and the epidermal hyperplasia should end at the same point. These lesions typically occur in younger patients (with an apparent predilection for the extremities of females).

Other examples of Spitz nevi may also display some element of suprabasal scatter of lesional melanocytes, although it may not be as prominent as that seen in the early pagetoid Spitz nevus described above. Similarly, other benign melanocytic proliferations can also show this feature, including traumatized/irritated nevi (see above), nevi recently exposed to strong ultraviolet light, congenital nevi (see below), pigmented spindle cell nevi (Reed nevi), and nevi of special sites (see below). In these benign lesions, the pagetoid cells are typically limited to the central
portions, often with associated nest formation. The presence of haphazard, asymmetrical, or peripheral pagetoid scatter is a clue for melanoma, as is any superimposed cytologic atypia or proliferative activity.7,8,10

Combined Nevi

Combined nevi are composed of 2 or more morphologically distinct populations of nevoid cells.38,39 This creates an impression of asymmetry, and thus these lesions are prone to misdiagnosis as melanoma. Although theoretically any combination of nevus subtypes is possible, there are several pairings that tend to be more commonly recognized. Oddly, given our emerging understanding of their differing molecular pathways, the most prevalent among these is the combination of conventional nevus and blue nevus (“true blue” nevus).39 characterized by a population of dendritic melanocytes admixed with the more conventional nevoid cells. The dendritic component is often more pigmented than the conventional component, and may be accompanied by prominent melanophages. Combinations of conventional and Spitz nevi can be particularly challenging because of the presence of a population of larger epithelioid cells mimicking melanoma.39 The spitzoid component of these lesions often displays glassy cytoplasm and eccentric nuclei, and may be accompanied by a lymphocytic infiltrate similar to that seen in halo nevi. Many of these lesions harbor a specific genomic signature characterized by loss of BAP-1 as well as a V600E BRAF mutation (otherwise uncommon in spitzoid lesions).40

Combinations of conventional and deep penetrating nevus-type cells also result in a population of larger melanocytes, which in this instance may extend deeply into the dermis or even the subcutis (often following the path of hair follicles, which can be a clue to the correct diagnosis) (Figure 2, A and B). Parenthetically, these lesions also appear to represent evolution of a distinct subpopulation of cells with a specific genetic abnormality within a conventional nevus.41 Many authors include within this category at least some lesions previously designated as clonal nevi, inverted type A nevi, atypical dermal nodule in benign melanocytic nevus, and melanocytic nevus with focal atypical epithelioid cell component.39 Another combination incorporating a population of larger, often heavily pigment−ed melanocytes is that of conventional nevus and pigmented epithelioid melanocytoma (see below). Although generally they are probably benign, the biological nature of these lesions is not yet fully characterized, and some examples have demonstrated metastatic potential. Thus, this diagnosis warrants some form of cautionary comment within the report, at least at this time. Although most combined nevi encountered in routine practice will include a component of conventional nevi, combinations of other subtypes can also occur. One example of this is the combination of Spitz and blue nevi (“BLITZ nevus”).42

An awareness of the entity of combined nevus is the first step in avoiding a misdiagnosis. They tend to be encountered more often in samples from younger patients.39 Although overall the lesion may appear asymmetrical, when the components are considered individually an element of symmetry becomes apparent. Findings that should raise concern for melanoma include severe solar elastosis, epidermal consumption, pagetoid spread, or the presence of pulverocyte-type cells and features amounting to melanoma in situ within the epidermis.1 Rare mitotic figures may be found in components of a combined nevus and do not necessarily indicate malignancy, although their presence should prompt a more careful consideration of the other features of the lesion.39 It is worth noting that melanoma developing in the dermal component of a nevus without an overlying component of melanoma in situ is rare in adult patients.39

Atypical Features in Congenital Nevi

Congenital nevi (a category that for practical reasons typically incorporates lesions that develop in the first few months after birth as well as those present at birth) occur in approximately 2% of the infant population.43,44 Although historically all such nevi were thought to carry a risk of malignant transformation, more recent studies45,46 have revealed that small to medium-sized lesions have no increased risk. However, large lesions (usually defined as greater than 5% of body area in infants/preteens or 20 cm or more in teenagers and adults) do carry an increased risk that is several hundred times that of the background population.45,46 Histologic features that can be associated with congenital nevi include extension into the deeper portion of the dermis and into the subcutaneous tissue; splaying of the collagen bundles of the reticular dermis and growth as cords

Figure 1. This lesion displays the classic trilayered appearance of a traumatized/recurrent nevus. An atypical junctional proliferation overlies an area of dermal scarring, below which is a bland intradermal nevus. In many cases, review of any previous specimen is critical to this diagnosis (hematoxylin-eosin, original magnifications ×20 [A] and ×100 [B]).
of single cells; and extension around and within adnexal structures, vessel walls, arrector pili, and nerves. It should be noted, however, that these criteria are not specific and are relatively insensitive.57 Atypical histologic features in congenital lesions from pediatric patients (Figure 3, A) are well recognized and are a potential pitfall. The atypia usually takes the form of architectural disorder (particularly of the junctional component), pagetoid spread, and cytologic atypia (Figure 3, B).58–61 A recent study61 documented at least one of these concerning changes in 77% of congenital nevi from young children, none of which underwent malignant transformation after a mean follow-up of approximately 7 years. Notably, mitotic activity was a relatively rare finding, even in the lesions showing other atypical features. Avoiding this pitfall relies largely on attention to the context of a lesion in a young patient, which might also show histologic features consistent with a congenital lesion. Other histologic clues to a benign lesion include nuclei remaining small and monotonous; atypia limited to the epidermis and superficial dermis, with the deeper portions of the lesion showing a bland appearance; the atypical junctional component not extending laterally beyond the intradermal component; and pagetoid extension tending to overlie nests.1 It is worth noting that most metastasizing melanomas arising within congenital nevi in childhood arise in the dermis and extend into the deep dermis or subcutis, in contrast to the largely superficial location of the benign atypical changes discussed here. In older patients, this rule does not apply; in these instances, a melanoma arising in a congenital nevus will typically demonstrate a conventional in situ component.1,4 Thus, in adolescents (and certainly in adults) a much higher degree of suspicion is warranted when faced with an atypical junctional melanocytic proliferation within a congenital nevus.

The presence of proliferative nodules is the other histologic feature encountered in congenital nevi that can cause concern for melanoma. These are seen in up to 5% of congenital nevi55,56,61 and simulate melanoma by presenting a nodular area of increased cellularity within the dermal component of the lesion. Factors favoring a benign diagnosis include symmetry of the nodule when considered individually, merging with the cells of the adjacent nevus at the periphery of the nodule, a low mitotic rate, lack of necrosis, and lack of diffuse high-grade cytologic atypia.55,56,61 Some authors62 have used the term atypical proliferative nodule for lesions displaying sharp demarcation, expansile growth, effacement of the epidermis, nuclear pleomorphism, or mitotic activity. Nonetheless proliferative nodules, including those designated as atypical, have not been significantly associated with any adverse outcomes.58,63–64 The distinction of these benign proliferations from a melanoma arising within a congenital nevus is notoriously difficult. Necrosis, ulceration, and increased mitotic figures, particularly atypical mitoses, have been suggested as criteria for melanoma.4,65 Given the difficulties in morphologic distinction, this may be a scenario where ancillary molecular testing will play an increasing role (see below).

Nevi From Special Sites

For a number of years it has become increasingly recognized that nevi occurring in certain areas of the body may show histologic features that overlap with those of melanoma. The number of these special sites seems to be continually rising. For this review we will focus on the most established areas, with a brief description of the histologic features that might be encountered as well as clues to avoid misdiagnosis. For a more in-depth analysis the reader is referred to published reviews on this topic.5,66,67 Perhaps the most classic example of this phenomenon is nevi occurring on acral sites (Figure 4, A). Although many of these appear like any other nevus, approximately half are characterized by atypical features within the intraepidermal component, including a lentiginous growth pattern, poor circumscription, irregular distribution of nests, and suprabasal scatter of melanocytes (Figure 4, B).66–69 Spitz nevi on volar sites can appear particularly concerning because of the uneasy combination of suprabasal scatter and large, atypical melanocytes.70 Similar features have also been reported in lesions from the ankle, particularly in female patients.71 These features tend to be more prominent in younger age groups, with the melanocytic proliferation residing mostly within the dermis after about 50 years of age.3,72 It is worth noting that in this anatomical location the histologic features can be greatly influenced by the plane of sectioning.
with regard to the dermatoglyphs. If sectioning has occurred parallel to these structures, the lesions tend to appear more disorganized, with apparently more haphazard suprabasal spread and poor circumscription. However, if the lesion is sectioned perpendicular to the dermatoglyphs, the lesions appear more circumscribed, with a more orderly arrangement of suprabasal scatter and columns of melanin pigment in the stratum corneum arranged over the tops of nests located primarily at the tips of the sulci. When this pattern is observed, it is a reassuring finding supporting the diagnosis of a nevus. Other clues to a benign diagnosis include small size (especially if the growth pattern is predominantly nested), young patient age, and the presence of a basal intradermal component. Concerning features include a large lesion with a predominantly lentiginous growth pattern, a significant junctional component in a lesion from an elderly patient, widespread cytologic atypia, thickened dendrites reaching into higher levels of the epidermis, and the presence of a dermal inflammatory infiltrate.

A proportion of nevi occurring on the genitalia of young females (the occurrence of similar lesions in males is not as well documented) can show worrisome histologic features, and the recognition of this variant is particularly important to avoid unnecessarily large excisions in this area. These lesions often present a nodular appearance at low-power examination (Figure 5, A) and are characterized by an atypical junctional component showing variation in melanocyte nest size, shape, and distribution (Figure 5, B). The nests may grow together to form large confluent structures or plaquelike arrays, and there is often an element of discohesion between the cells within the atypical nests. The combination of these factors may lead to clefting separation at the epidermal-dermal junction. The lesional melanocytes may be enlarged with an epithelioid appearance, and focal pagetoid spread or involvement of adnexal structures may be evident. The atypical features may extend into the dermis, but tend to be limited to the superficial component on the lesion. Mitotic figures may be present. Clues to the benign nature of the lesion include the young age of the patient, overall small size and symmetrical appearance, lack of a significant junctional component extending laterally away from the intradermal component, and preservation of cytologic maturation at the deeper aspects of the lesion. The possibility of a dysplastic nevus may also be considered, although classically these tend to show a more lentiginous growth pattern, with elongation of the rete ridges and fibroplasia of the papillary dermis. It is helpful to note that genital melanoma (especially if located on mucosal surfaces) is typically a disease of elderly patients and is often characterized by a lentiginous growth pattern with cells showing angulated, hyperchromatic nuclei and minimal cytoplasm. A good rule of thumb is that a genital melanocytic proliferation sent from the obstetric clinic (at which these lesions frequently come to medical attention) is very likely to represent a nevus unless there are absolutely compelling reasons to think otherwise.

Lesions from the scalp (particularly in adolescents) or anywhere along the milk line (typically encompassing axillae, breast, and genital areas) can show atypical features that often overlap with those described for dysplastic nevi and are similar to atypical genital nevi. This can include junctional shouldering and large confluent nests, which may show an element of cellular discohesion. There may be adnexal involvement and a degree of cytologic atypia. Nevi from the ear can also appear atypical, with poor circumscription and junctional shouldering, and may be composed of larger pigmented cells with a spitzoid appearance. The difficulty with these lesions may be enhanced by a concerning background of dermal solar elastosis and/or limited biopsy material because of anatomical difficulties with this site. Nevi over the umbilicus may lack maturation because of the fibrous nature of the underlying tissue. Nevi from flexural sites may have a papillated growth pattern and may also display a discohesive pattern of growth.

Clues to the benign nature of these lesions include recognition of the site; a younger age group; maturation of the dermal component; low or absent mitotic rate, with any mitoses limited to superficial portion; and pagetoid spread limited to the central portions of the lesion. As with all nevi...
displaying atypical features, if there is doubt, re-excision of the site to ensure complete removal of the lesion is a reasonable option, even if the favored diagnosis is that of a benign nevus.

**Deep Penetrating Nevi**

Deep penetrating nevi are a variant that tends to occur in younger age groups (within the first 3 decades) on the head/neck, trunk, or upper extremities. Histologically they are characterized by a symmetrical, well-circumscribed, often wedge-shaped profile. The lesional cells are large, with a spindled to epithelioid morphology, and arranged in fascicles and nests. The lesion is often characterized by a plexiform growth pattern, with melanocytes following neurovascular bundles or adnexal structures and relatively deep extension into the reticular dermis or even subcutis. The cells maintain their large size throughout the depth of the lesion and lack the maturation associated with nevi, potentially leading to misdiagnosis as melanoma. An awareness of the entity, as well as attention to the reassuring low-power silhouette, should help avoid this trap.

Recent work has demonstrated that deep penetrating nevi are molecularly distinct from both conventional and Spitz nevi, being characterized by combined activating mutations in both the MAPK and β-catenin signaling pathways. Features of deep penetrating nevus may be also be seen as a component of combined nevi, perhaps reflecting evolution by the acquisition of β-catenin mutations within a subclone of conventional nevus cells.

The landscape of these lesions is somewhat complicated by a group of atypical deep penetrating nevus-like lesions, which may show asymmetrical cellular and/or expansile foci, cytologic atypia, or increased mitotic activity. Within this group of lesions there is a propensity for spread to local lymph nodes, demonstrated by sentinel node positivity in...
approximately a third, apparently without further systemic spread or death. These lesions could be categorized under the evolving concept of borderline tumors (discussed in greater detail below). However, also included in the group of atypical lesions are smaller numbers of tumors that have demonstrated more aggressive behavior, characterized by local recurrence and distant metastatic spread resulting in the death of the patient. These lesions have been termed plexiform melanoma. Although the morphologic features are relatively loosely defined, on a molecular basis they show chromosomal aberrations typical of melanoma (see below). Importantly, cases showing both a morphologic and molecular progression from deep penetrating nevus to melanoma have been documented, indicating that as a group these lesions could be conceptualized as a precursor lesion with the potential to acquire further mutations resulting in the transformation to a potentially fatal melanoma. This fact alone probably justifies the complete removal of all deep penetrating nevi.

**Benign Features in Malignant Lesions**

**Metastatic Melanoma**

Cutaneous deposits of metastatic melanoma are typically small, circumscribed, and symmetrical, thus potentially mimicking benign nevi. Metastatic lesions with a blue nevus–like morphology are well described (Figure 6, A through D), and more recently small, symmetrical deposits with a florid lymphocytic inflammatory infiltrate mimicking a halo nevus have been reported in the setting of immune checkpoint inhibitor therapy. In addition, a nevoid subtype of metastatic melanoma has been described, which can show features suggestive of maturation. A history of melanoma provides an important clue to the correct identification of these lesions.
Diagnosis in these cases. In addition, metastatic melanoma will typically have a degree of cytologic atypia (the cells may be monomorphous but atypical) and mitotic activity that is incongruous with a benign diagnosis.89 The presence of lymphatic invasion1 or angiotropism92 might also provide a clue to the metastatic nature of the lesion.

Desmoplastic Melanoma

Desmoplastic melanomas, especially in their pure form, are characterized histologically by a moderately cellular spindle cell proliferation within the dermis (Figure 7, A). The lesional cells are typically fibroblast-like (Figure 7, B) and can be mistaken for a benign spindle cell lesion (eg, scar, neurofibroma, dermatofibroma), a conventional blue nevus, or desmoplastic forms of nevi.1,2,93–95 The demographics can be helpful in the identification of desmoplastic nevi, as these lesions tend to occur on the limbs of young patients,95,96 compared with the typical setting of desmoplastic melanoma, which occurs in an older age group, particularly in sun-damaged skin of the head and neck.97 In addition, desmoplastic nevi are typically small with a wedge-shaped silhouette, with a diminution in cellularity towards the deeper aspects of the lesion. The lesional cells will tend to be larger, with an epithelioid or spindled morphology, abundant cytoplasm, and sharply demarcated cytoplasmic borders.96 An intraepidermal component can be present in both desmoplastic nevi and desmoplastic melanoma, but can be helpful if it is clearly malignant (often showing morphologic features of lentigo maligna) (Figure 7, C). The presence of clusters of lymphocytes within the dermis is a low-power (though not completely sensitive or specific) clue to desmoplastic melanoma.2,11,98 It should be noted that the cells of a desmoplastic melanoma will typically be negative for Melan-A and HMB45, in contrast to the cells of an overlying intraepidermal component.1 However, most desmoplastic melanomas will label with antibodies against S100, Sox10, p75, WT1, and nestin.99 Although these stains can be useful in identifying a subtle lesion, scattered cells within scar tissue can also show positive labeling with several of these markers100–102; thus, some caution is required in interpretation. Nonetheless, we advocate a low threshold for immunohistochemical evaluation of any unusual dermal spindle cell proliferation, particularly in sun-damaged skin of an elderly patient.

Nevoid Melanoma

As the name implies, this subset of melanomas is characterized by a constellation of histologic features that mimic nevi. The category is perhaps best summed up by the definition of McKee: “a melanoma which you diagnosed as a nevus and wished you hadn’t.”92 They may be broadly categorized into 3 morphologic groupings. The first is characterized by nested growth of melanocytes predominantly within the epidermis, mimicking a junctional nevus (Figure 8, A).103,104 However, these lesions tend to be larger than typical junctional nevi (greater than 6 mm) and usually occur in older patients. Morphologic clues include unusually large and irregularly shaped nests, which often have a layer of keratinocytes between them and the basement membrane (Figure 8, B),103 indicating that they are not truly junctional like the nests of a nevus. This layer can be highlighted by keratin immunohistochemical stains in difficult cases. Higher-power examination may also reveal a degree of cytologic atypia. It is worth noting that some of these cases may reveal focal invasive melanoma on examination of multiple levels.103 The other 2 categories are dominated by an intradermal component, described as papillated or nodular depending on the overall architecture (Figure 9, A).105 These lesions can occur in younger
patients,105,106 and thus recognition of them as malignant requires attention to subtle morphologic clues. These include a dermal growth pattern that is hypercellular and sheetlike rather than nested107 or the presence of large irregular nests with little intervening collagen,2 particularly if these occur at the deeper aspect of the lesion. If present, an atypical junctional component can be helpful. Subtle cytologic atypia is present at high-power examination, and there may be dermal mitotic figures (Figure 9, B).105 The combination of these 2 features should be of particular concern. Other clues include hyperpigmentation in the deeper aspects and necrosis of single melanocytes. In the deeper portions of the dermal component, the amount of cytoplasm may decrease, giving the false impression of maturation. However, the overall cellularity does not decrease and the subtle atypia remains.1

It should be stressed that the diagnosis of a nevoid melanoma rests on the constellation of features, with no one feature being diagnostic. Thus, the finding of an occasional mitotic figure in the dermis of an otherwise banal nevus does not mean the lesion is a melanoma. However, it should prompt a more careful examination of the lesion over multiple levels and consideration of ancillary studies if there is significant concern (Figure 9, C).2,105

**Spitzoid Melanoma**

Spitzoid melanomas are malignant lesions showing features that resemble Spitz nevi. These lesions can be remarkably symmetrical and circumscribed on low-power examination and are composed of large epithelioid or spindled melanocytes. The distinction of Spitz nevi from spitzoid melanomas is covered in greater detail below.
PROBLEMATIC LESIONS

Dysplastic Nevus Versus Melanoma

There are many controversial topics in melanocytic pathology, but perhaps the most commonly encountered in routine practice is the dysplastic nevus. The lesion currently known as a dysplastic nevus was delineated by Clark and colleagues108 as a type of large, atypical nevus that they encountered in patients who had an inherited tendency to develop melanoma. They proposed that lesions with this morphology may represent an intermediate stage between an ordinary nevus and melanoma. The nevi that Clark et al.108 described are characterized clinically by larger lesions (greater than 5 mm) that have a flat component and display an irregular border and variability in coloration. Histologically, these lesions show both architectural and cytologic atypia.108,109 In terms of architecture, the junctional component often extends beyond any intradermal component (“junctional shouldering”), and the melanocyte nests vary in terms of their size and distribution and may form “bridges” across adjacent rete ridges. Often there are areas where lentiginous single-cell growth predominates, particularly at the sides of the rete ridges. Within the papillary dermis there is often a lymphocytic infiltrate, as well as a relatively characteristic stromal reaction comprising either lamellar fibroplasia, characterized by horizontal collagen with clefts parallel to the epidermis, or concentric fibrosis, where the collagen bundles orient around the bases of the rete ridges. Cytologic atypia is described as random; in other words, atypical nuclei are scattered throughout the lesion, as opposed to the diffuse atypia more typical of melanoma.

The existence of lesions with this morphology is not controversial; they are routinely encountered in everyday practice in the setting of patients with multiple lesions and strong familial melanoma predisposition, but more commonly as (apparently) sporadic lesions. Significant debate has occurred around concepts of these lesions as markers of risk and/or precursors of melanoma, as implied by the term dysplastic. Some have worked around this by using the term Clark nevus10 to acknowledge the entity without accepting the concept of it as a premalignant lesion. Others111 have suggested more cumbersome descriptive terminology (“nevus with architectural disorder, with or without cytologic atypia”), which, not surprisingly, has not been widely accepted. The observation that some dysplastic nevi–like histologic features can be observed in a broad range of otherwise banal nevi112 also contributes to the controversy. Lesions labeled as mildly dysplastic nevi in many practices, particularly small lesions, in fact represent very low-risk proliferations, with a negligible chance of progression to melanoma and little implication for individual or familial risk. In our view, many of these small lesions would be better classified as lentiginous junctional nevus. It is not our intention to expand further on this ongoing debate in this review, as there are innumerable pages to which the interested reader can refer.7,113,114 Rather, here we will concentrate on the practical issue of separating a dysplastic nevus from a melanoma.

If one considers the clinical and histologic features of dysplastic nevus as outlined above, it becomes apparent that there is potential overlap with features of melanoma. The presence of significant pagetoid spread of melanocytes (particularly at the edges of the lesion), diffuse cytologic atypia, and mitotic figures in the intradermal component are important clues to melanoma.1 The bridging of rete ridges in dysplastic nevi should be distinguished from more significant confluent growth involving the undersurface of the epidermis between the ridges.1 The diagnosis of dysplastic nevus should be made with caution if there is a background of solar elastosis, particularly on the face.1 Lesions with a dysplastic morphology in these areas show great overlap with a group of lesions encompassing the “lentiginous dysplastic nevus of the elderly”112,113 and “lentiginous melanoma,”114 and in our view all dysplastic nevi on sun-damaged skin should be completely excised with a margin of normal tissue. It should also be noted that dysplastic nevi can show superimposed features of regression and/or traumatization, further adding to the difficulty of the distinction from melanoma.1 Lesions showing features of both Spitz nevus and dysplastic nevi (“Sparks nevi”) have also been described, and can be particularly concerning because of the combination of architectural atypia with large cells.115 Lack of widespread pagetoid spread involving all levels of the epidermis, dermal maturation, and lack of diffuse nuclear atypia help to distinguish these lesions from spitzoid melanoma. Finally, melanoma (particularly melanoma in situ) can be seen in association with dysplastic nevi; indeed, there is increasing evidence that the morphologic borderline is characterized by cumulative acquisition of the molecular attributes of frank melanoma.120 Clues to evolving melanoma include asymmetry, both of the junctional component and of any associated lymphocytic infiltrate; confluence of junctional nests; and widespread pagetoid spread.1 With this said, it is unsurprising that there is very significant interobserver variability in interpreting lesions in this group.127 Complete excision is prudent in all lesions where there is doubt, and it is important for the pathologist, the clinician, and the patient to recognize that the outcome is always excellent in completely excised lesions with borderline or equivocal changes of melanoma in situ, and almost always excellent in lesions where the differential diagnosis lies between a severely dysplastic nevus and a thin invasive melanoma.128,129

Malignant/Atypical Blue Nevus Spectrum

This group of melanocytic lesions is notoriously difficult. The combination of relative rarity and a lack of consensus regarding diagnostic terms and histologic criteria has led to a situation where lesions within this spectrum are subject to marked interobserver variability even among experts in the field.130 Although we do not advocate use of the oxymoron term malignant blue nevus in practice, we have used it here to encompass 2 separate entities: a de novo melanoma that adopts a blue nevus–like morphology (blue nevus–like melanoma) and a melanoma arising within a preexisting blue nevus. In both scenarios the blue nevus component (whether a precursor lesion or simply a facsimile) can be either a common blue nevus, with spindled or dendritic melanocytes, or a cellular blue nevus, with more ovoid cells occurring in nests or fascicles.131 Although as a group these are rare lesions, they are important, as they tend to present at an advanced stage, with concomitant high rates of recurrence and/or metastasis.132,133 Melanoma can develop within a blue nevus (either common or cellular), just as it can in conventional nevi.131 One might also include in this category melanomas arising in areas of dermal melanocytosis (eg, nevus of Ota).133 These lesions are characterized by a melanoma in conjunction with evidence of a precursor blue nevus, which may be best visualized at the periphery of the lesion. Most cases

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seem to arise within cellular blue nevi, particularly larger lesions on the scalp. The development of melanoma within these lesions is heralded by the development of nodules or plaques within a long-standing lesion or other forms of sudden clinical evolution such as ulceration. The tumor is typically large (>1–2 cm)30 and composed of asymmetrical nodules of highly atypical epithelioid or spindled melanocytes that compress or displace surrounding structures. The tumors may show an infiltrative pattern and tend to be readily recognizable as melanoma at low-power examination.131 There may be areas of necrosis, and the presence of mitoses, particularly in numbers greater than 2/mm², is very helpful in confirming the diagnosis.130,131 If the lesion has truly arisen within a blue nevus there should be no component of conventional melanoma in situ. Interestingly, in view of the molecular similarities between blue nevi and uveal melanoma, loss of BAP1 staining in this context appears to be a strong marker of malignancy.134

Other melanomas can arise de novo with a morphology that resembles blue nevus, recapitulating the expansile extension into the subcutaneous tissue that is often seen with cellular blue nevi.33 Blue nevus-like melanoma will often have a junctional component, as opposed to conventional blue nevi, which are intradermal lesions only.1 In a melanoma there will be diffuse cytologic atypia with enlarged nuclei displaying hyperchromasia and a coarse chromatin pattern. Mitotic figures are an important clue to melanoma in this context.

Also falling within the differential diagnosis for this group of lesions is the so-called atypical (cellular) blue nevus. This term is used to describe lesions that have clinical features consonant with a blue nevus and histologic features that include some atypical findings, but not enough to render a definite diagnosis of malignancy.130,135,136 The atypia may be architectural or cytologic, and may take the form of scattered atypical cells without overt expansile nodules, low-grade rather than high-grade atypia, occasional mitotic figures (generally not more than 2/mm²), or only limited foci of necrosis.138 These lesions tend to be larger and more cellular than the typical blue nevus. It is worth noting that there are documented examples of lesions designated as atypical blue nevus that have gone on to metastasize and ultimately proved fatal.139 Although there is some genomic evidence suggesting that these lesions may be a separate entity from melanoma arising within a blue nevus, until they are better characterized in terms of diagnostic criteria and biological behavior it is sound practice to regard all such lesions with a healthy dose of caution, with complete excision and close clinical monitoring regarded as mandatory.1

Two final practical points regarding this group of lesions are worth mentioning. First, it should be remembered that deposits of metastatic melanoma can be exceedingly good mimics of blue nevi; thus, any blue nevus diagnosed in a patient with a history of melanoma is probably worth a second look prior to sign-out. It is also worth noting that there are reports of otherwise benign cellular blue nevus deposits occurring in lymph nodes, further complicating the landscape when it comes to determining what constitutes malignancy when it comes to lesions of this type.130,139

**Atypical Spitz Nevi/Tumors and Spitzoid Melanoma**

Even in the first descriptions by Sophie Spitz,140 the histologic overlap between what we now refer to as Spitz nevi and melanoma was recognized, and lesions that combine a spitzoid morphology with atypical features are renowned as a source of diagnostic difficulty and substantial interobserver variability.143,142 Around the turn of the century there were a number of attempts to delineate criteria to separate Spitz nevi from melanoma.145–147 These were somewhat disparate, and a landmark study by Barnhill and colleagues141 in 1999 established that there was little consensus, even among experts. Furthermore, the poor discriminatory ability of the prevailing criteria was demonstrated in a striking fashion when 68% of the lesions in that study that had been proven to be malignant clinically were diagnosed as benign by the majority of participants. Around this time the term atypical Spitz tumor (as well as others including malignant Spitz nevus and metastasizing Spitz nevus) had entered the lexicon to label lesions with a spitzoid morphology for which a definite distinction between a nevus and a melanoma could not be made.35,148–152 Although there were no established criteria for this diagnosis, the lesions tended to be characterized by a large size, ulceration, significant depth, lack of maturation, prominent cellular density and/or pleomorphism, and increased numbers of mitoses.153 To underscore the difficulty pathologists faced with these lesions, reports emerged of lesions designated as Spitz nevus or atypical Spitz nevus that had subsequently metastasized.154–156 Also around this time it was becoming apparent that some spitzoid lesions seemed to spread to local lymph nodes without evidence of further distant metastases, at least on the follow-up available at the time of the reports.146,149 These concepts were opposed vociferously by some, perhaps most notably Mones and Ackerman,157 who argued that even a lymph node metastasis was still a metastasis and that any spitzoid lesion that had displayed an ability to metastasize should be labeled a melanoma and nothing else. Others158 countered that this approach was too simplistic for this group of lesions, instead advocating for a risk stratification of lesions into Spitz tumors without abnormality, atypical Spitz tumors, and spitzoid melanoma (with the replacement of nevus with tumor, designed to avoid designating any of these lesions as unequivocally benign). Further complicating the landscape is the tendency within some areas of the literature to separate spitzoid melanomas of childhood from the broader group of spitzoid melanomas,138 although specific criteria other than age (which itself has its own inherent issues around when childhood ends) for this subgrouping have not been well delineated.

Some authors advocated the use of sentinel lymph node biopsy as a diagnostic adjunct in difficult cases, with lesions reclassified as melanomas if the sentinel node was found to be positive. This was initially proposed to have the dual benefit of providing management appropriate for a melanoma (if that is what the lesion turned out to be) as well as being a diagnostic adjunct. Studies with this approach156,160–164 reported a sentinel node positivity rate of approximately 30% to 50% for lesions labeled as atypical Spitz tumors. In one study,160 lesions associated with nodal deposits tended to have a greater thickness, increased mitoses, deep mitoses, incomplete maturation, and expan-sile dermal nodules (although only tumor thickness showed a statistically significant association). Others argued against the legitimacy of this approach, citing uncertainty about the true biological meaning of small, subclinical deposits within lymph nodes in the setting of spitzoid lesions, especially in the context of the known propensity of benign nevus cells to be found within nodes. Further, it became apparent that as a group these lesions have a better

1. Harvey & Wood
prognosis than conventional melanoma, as despite the relative depth of the primary lesions and the high rate of positive sentinel nodes, the fatality rate is typically low.\textsuperscript{156,160–164,166,167} Although a number of explanations could be proposed to explain this (young age of the patients, potential misinterpretation of benign nodal nevi, therapeutic effect of sentinel node removal), the possibility that these lesions may represent a group of intermediate tumors (analogous to the pigmented epithelioid melanocytoma discussed below) was also raised.\textsuperscript{166}

More recent advances in the classification of spitzoid neoplasms have revolved around their molecular characteristics. Initially described was a group of lesions characterized by oncogenic mutation, with or without amplification, of the \textit{HRAS} gene on chromosome 11p, which typically show a predominantly intradermal growth pattern with desmoplasia.\textsuperscript{168} Subsequent studies have revealed that activation of kinase pathways seems to play a central role in the pathogenesis of lesions with a spitzoid morphology. This was confirmed in a landmark study by Wiesner et al\textsuperscript{169} in 2014, which demonstrated in-frame fusion transcripts resulting in the overexpression of kinase genes (including \textit{ROS1}, \textit{ALK}, \textit{NTRK1}, \textit{RET}, and \textit{BRAF}) in just more than 50% of spitzoid neoplasms. At least some of these molecular subtypes can be suspected morphologically: for example, lesions with a \textit{ALK} fusion tend to display a wedge-shaped configuration with a plexiform growth of spindled spitzoid melanocytes.\textsuperscript{170,171} Mutations in conventional melanoma-associated genes such as \textit{BRAF}, \textit{NRAS}, \textit{KIT}, \textit{GNAQ}, or \textit{GNA11} are uncommon in spitzoid lesions,\textsuperscript{169} with the notable exception of a morphologically distinct subset defined by \textit{BRAF} mutations combined with biallelic \textit{BAP1} loss. These lesions are characterized by large epithelioid melanocytes and have been described in syndromic and sporadic forms as well as representing a component of combined nevi.\textsuperscript{169,372,175} Whether these molecular variants correlate with the potential biological behavior of an individual lesion or with differing morphologic or molecular criteria for predicting malignancy remains to be fully determined.

With this long history of confusion and controversy as a backdrop, how should the pathologist deal with spitzoid lesions? The answer is with a healthy dose of caution. A classic Spitz nevus should be characterized by the following lesions? The answer is with a healthy dose of caution. A backdrop, how should the pathologist deal with spitzoid determined.

criteria for predicting malignancy remains to be fully correlate with the potential biological behavior of an melanoma (Figure 10, A and B). This term emerged

The Borderline Melanocytic Lesion

Despite our best efforts, it is well established that there are a group of melanocytic lesions that cannot be reproducibly classified into standard diagnostic categories, even after review by expert panels.\textsuperscript{130,141,142,174} Conceptually, these lesions can be broadly categorized into 2 groups, succinctly described by Zembowicz and Scolyer\textsuperscript{175} as indeterminate and intermediate lesions. Indeterminate lesions represent those lesions that, because of conflicting histologic criteria, have been difficult to classify as benign or malignant. Presumably they represent one or the other in terms of their true biological potential, but the constellation of histologic features has not allowed for a definitive distinction between these possibilities. Hopefully, further development of ancillary molecular techniques will help to reduce the size of this group. Alternatively, an evolving concept in melanocytic pathology is the intermediate lesion, representing a proposed category of tumors that show intermediate or low malignant potential.\textsuperscript{175} These tumors have a relatively good long-term prognosis, despite the frequent presence of at least some degree of metastatic spread. The most well-categorized example of this concept is the lesion referred to by some authors\textsuperscript{175} as pigmented epithelioid melanocytoma (Figure 10, A and B). This term emerged

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from the observation of histologic similarities between epithelioid blue nevus (either sporadic or associated with Carney syndrome) and a type of heavily pigmented melanocytic tumor referred to at that time as animal-type melanoma because of its resemblance to a lesion found in gray horses. At the time of this observation, many patients with these lesions were undergoing sentinel node biopsy, which revealed metastatic deposits in approximately half. Despite this, clinical follow-up had not identified any fatalities from the disease. This finding was confirmed by a subsequent study after a longer follow-up period, again showing a high rate of sentinel node metastasis (44%) with no disease-related deaths after a median follow-up period of approximately 5 years. Given these observations, it became clear that although pigmented epithelioid melanocytoma has metastatic potential, the clinical outcome is markedly different from that of conventional melanoma. Thus, it was proposed that it may represent a unique low-grade variant. Other types of difficult melanocytic lesions that may also represent low-grade neoplasms include some atypical Spitz tumors and atypical deep penetrating nevi. Apart from limited studies involving microRNAs, we are currently unaware of any molecular data clearly supporting this concept, although we expect that the upcoming decade will encompass numerous studies focusing on this element.

Discussions regarding various subtypes of borderline lesions aside, in a practical sense these lesions can be problematic on a number of levels. As their clinical behavior cannot be accurately predicted, there is a level of uncertainty regarding the outcome, which can create understandable frustration on behalf of both clinician and patient. As these lesions are rare, practitioners are often not familiar with them or with the nuances of interpretation and nomenclature, and are often puzzled as to why a more definitive diagnosis cannot be provided. Indeed, the lack of diagnostic certainty is often mistaken for a lack of diagnostic acumen. It may be tempting to avoid these issues by upgrading a lesion to a melanoma if there is any doubt, with the rationale that it will ensure adequate treatment and cannot be proven to be incorrect (a good outcome can be attributed to appropriate management). Indeed, there is evidence for overdiagnosis of melanoma in recent decades. This temptation should be avoided: an unwarranted diagnosis of melanoma may cause a number of difficulties for the patient, most of which are essentially hidden from the pathologist. These include significant influences on life decisions, difficulty obtaining insurance, unwarranted surgical procedures with the potential for adverse events, and unnecessary fear and anxiety.

Effective reporting of borderline melanocytic lesions requires consideration of all of the above issues. Given the diversity of lesions that might fall into this category, it is difficult to provide concrete recommendations that would apply to all situations. Our general approach is similar to that described by others. While outlining the difficulties of the lesion, we include our favored diagnosis as well as a brief synopsis of our reasoning. If we have sought further opinions from our colleagues (which we would do routinely for difficult melanocytic lesions), the outcome of these is also included. For most cases we will also include a recommendation for further management, commensurate with our level of anxiety regarding the lesion. We reiterate the advice of others that all borderline melanocytic lesions should be completely excised, and thus we will recommend a wider excision of all lesions that are incompletely or only marginally excised. For wholly intraepidermal lesions, as well as the majority of thin lesions (Breslow thickness of 1 mm or less), this approach will ensure appropriate treatment regardless of the true biological nature of the lesion. Recommendations regarding sentinel node biopsy are more problematic, being clouded both by uncertainty regarding the significance of a positive finding in a subset of lesions (such as pigmented epithelioid melanocytomas; see above) and by ongoing controversies regarding the role of sentinel node sampling in melanocytic lesions.

**Lentigo Maligna Versus Actinic Melanocytic Hyperplasia Versus Cicatricial Melanocytic Hyperplasia**

Lentigo maligna is a form of melanoma in situ characterized in its early stages by increased numbers of melanocytes predominantly arranged as single units, with minimal suprabasal spread and occasional nest formation. The adnexal epithelium may also be involved. The lesional cells may show atypical, hyperchromatic nuclei, but this can...
be subtle. A single-cell junctional melanocytic hyperplasia can also be seen in chronically sun-damaged skin and overlying areas of dermal scarring (cicatricial melanocytic hyperplasia). As lentigo maligna is a lesion of chronically sun-damaged skin and as it often requires multiple excisions to obtain histologic clearance, these 3 entities often pose a differential diagnostic challenge. A particular challenge involves the assessment of lentigo maligna margins, where the surrounding skin will often show actinic melanocytic hyperplasia. Essentially the distinction relies on an assessment of the density of melanocytes as well as nuclear enlargement and hyperchromasia, both of which should be greater in the lentigo maligna compared with the surrounding skin. Of course, although this is easy to say, the distinction can be exceedingly difficult in practice. In a similar fashion, it can be difficult to distinguish residual lentigo maligna from the cicatricial melanocytic hyperplasia resulting from the scarring process. Again, this relies upon a judgement regarding the density and cytologic features of the proliferation. In addition, limitation of the changes to scarred areas supports a diagnosis of cicatricial hyperplasia, whereas subtle nest formation, confluence, and suprabasal spread support residual melanoma. The assessment of subtle lesions in this spectrum is greatly assisted by reference to the clinical appearance and history: if there is a clinically apparent pigmented macule without other obvious histologic explanation, or a history of lentigo maligna extending to the margins of a prior biopsy, the level of suspicion can be much higher than when an incidental diagnosis (eg, in the setting of re-excision of nonmelanoma skin cancer) is being contemplated.

**IMMUNOHISTOCHEMISTRY**

In our practice immunohistochemistry plays a relatively small role in the assessment of primary melanocytic lesions. Although it is relatively easy to demonstrate immunohistochemical differences between obvious nevi and obvious melanomas, the contribution of these markers in truly ambiguous cases is limited. Although melanocytic markers can occasionally be helpful in appreciating the distribution of cells in subtle lesions, the routine ordering of multiple markers of melanocytic differentiation in challenging melanocytic proliferations (which we see very frequently in external cases) is at best unhelpful, and at worst may waste tissue on which more fruitful investigation could be undertaken. In contrast, deeper levels are occasionally valuable when faced with a challenging lesion. Before ordering deeper levels and/or immunohistochemistry, it is prudent to consider whether material should be conserved for ancillary studies by retention of ribbons, retention of material on charged slides for fluorescence in situ hybridization (FISH) testing, or retention of thick (in our practice 5 × 20 μm) sections on slides for microdissection and array-based comparative genomic hybridization (aCGH). It is better to have such material available and not require it than to be in the opposite position.

The presence of a decreasing gradient in expression of HMB45 is often quoted as support for a benign diagnosis, reflecting the maturation of melanocytes. However, there are many nevi that lack this gradient (blue nevi, deep penetrating nevi, Spitz nevi), and we do not regularly perform this stain for this purpose. The use of MIB1 to assess proliferative activity can occasionally be useful, particularly in spitzoid and other thick lesions, and we will invariably combine this with a Melan-A stain to help ensure that we are only assessing proliferative activity in melanocytes. An emerging antibody that may prove useful detects 5-hydroxymethylcytosine. Loss of 5-hydroxymethylcytosine is an epigenetic marker associated with melanoma. Although the role of this stain remains to be established, initial studies have shown promising results.

Immunohistochemistry can be useful to identify or delineate subtle melanocytic proliferations such as desmoplastic melanoma, lentigo maligna, and subtle invasion in areas of fibrosis or inflammation underlying melanoma in situ. For this purpose we tend to use Sox-10, as it has excellent sensitivity for melanocytes in these situations, although it is important to be aware of the labeling of scattered cells within scar tissue in the context of consideration of desmoplastic melanoma or subtle invasion. Markers of endothelial cells, such as D2-40, can be used (with or without double labeling with a melanocytic marker) to confirm the presence of lymphatic invasion, and will also detect it at a higher rate than morphologic assessment alone (although whether this is significant with regard to prognostication remains uncertain).

Immunohistochemistry can also be useful as a surrogate marker of key mutations, which in some situations can be diagnostically useful. Foci showing loss of immunohistochemical staining for p16 may result from biallelic loss of chromosome 9p21, an aberration that would support a diagnosis of melanoma (Figure 11, A and B). Some caution should be exercised in assessing these stains, and it should be noted that there are other mechanisms by which immunoreactivity could be lost; thus, negative staining does not necessarily correspond to homozygous 9p21 deletion. Alternatively, strong patchwork staining for p16 throughout the lesion is good evidence for a lack of this molecular aberration. A specific antibody is also available for detecting the presence of the BRAF V600E mutation, which may occasionally be useful in the assessment of spitzoid lesions, which (with the exception of BAP1-deficient lesions) seldom show this mutation. The combination of a BRAF mutation and immunohistochemical loss of p16 staining in a putative spitzoid lesion is concerning for melanoma. Although the combination of BAP1 loss and BRAF V600E mutation is typically (but not always) a feature of clinically benign lesions, BAP1 loss in the setting of a blue nevus-like lesion driven by G-protein abnormalities is very concerning.

**ANCILLARY MOLECULAR TESTING**

The many pitfalls in the morphologic assessment of melanocytic lesions discussed above provide a sobering reminder that although morphologic criteria are relatively well established, in practice they are not always reliable. This is reflected in the many studies demonstrating only moderate interobserver agreement for the diagnosis of melanocytic lesions. Recent years have seen the advent of ancillary molecular tests able to identify chromosomal aberrations commonly associated with melanoma, providing pathologists with another tool to assist with difficult lesions. Seminal studies using comparative genomic hybridization demonstrated that both gains and losses of genetic material are common in melanomas but rare in nevi (with the exception of single specific abnormalities in a subset of Spitz nevi). Subsequent work has confirmed that distinct genetic alterations are found in different clinicopathologic groups of melanomas, and
as our understanding of these differences improves it is feasible that a molecular classification of melanomas may replace the current morphologic system.\(^{207}\)

The potential for cytogenetic aberrations to provide a diagnostic tool for separating melanoma from benign proliferations led to the development of a FISH probe set targeting the more common chromosomal copy number changes identified in melanoma.\(^{208}\) Fluorescence in situ hybridization probes targeting 13 of these regions were developed and tested against a cohort of nevi and melanomas, eventually yielding a final probe set targeting 6p25 (RREB1), 6q23 (MYB), and 11q13 (CCND1) (as well as a probe against centromere 6). This probe set achieved a sensitivity of 86.7% and a specificity of 95.4% for the separation of melanoma from nevi.\(^{208}\) Fluorescence in situ hybridization testing in this manner has been demonstrated to be useful in many of the difficult scenarios that we have previously discussed, including the separation of blue nevi from blue nevus–like metastases,\(^{209}\) nevoid melanoma from nevi,\(^{210}\) desmoplastic melanoma from desmoplastic nevi,\(^{211}\) and cellular blue nevi from blue nevus–like melanoma,\(^{212}\) and in the diagnosis of superficial lesions with suprabasal spread of melanocytes.\(^{213}\) Despite these encouraging results, FISH testing has limitations,\(^{214,215}\) and it has been shown to be most useful when used in combination with expert morphologic evaluation.\(^{216}\) In particular, it may not be as sensitive in detecting aberrations in spitzoid neoplasms,\(^{216-218}\) although ironically this may be the most common scenario where it is requested.\(^{219}\) The addition of probes directed at 8q24 (c-MYC) and 9p21 (CDKN2A, to detect homozygous loss) has been shown to improve sensitivity in this context.\(^{208,221}\) Going forward, the FISH test may play an increasing role in prognostication, particularly for spitzoid lesions, with evidence that certain aberrations (homozygous loss of 9p21, gains in 8q24) are associated with more aggressive clinical behavior.\(^{222-224}\)

The other technique in diagnostic use is aCGH. This technique essentially compares the sample genomic DNA with that of a (in our practice sex mismatched) reference, each differentially labeled with a different fluorescent marker, to detect the presence of copy number variations affecting large areas of chromosomes.\(^{203,225-229}\) Similar to FISH testing, aCGH has been shown to be useful in a number of troublesome settings within melanocytic pathology, including blue nevus–like lesions,\(^{230,231}\) proliferative nodules in congenital nevi,\(^{232}\) congenital melanomas,\(^{233}\) deep penetrating nevi,\(^{87}\) and acral proliferations.\(^{234}\) Fluorescence in situ hybridization and aCGH can be a complementary pair of techniques to have in one’s diagnostic armamentarium. Although aCGH provides greater coverage of the genome, it requires a sufficient amount of DNA to be successful, and thus, at least without whole-genome amplification, its use is limited to relatively thick lesions (a Breslow thickness of at least approximately 1 mm in our hands).\(^{226}\) Fluorescence in situ hybridization provides an alternative technique for thinner lesions and allows assessment of subpopulations within a heterogenous lesion.

**PRACTICAL PROBLEMS**

**Partial Biopsies**

Partial biopsies of pigmented lesions are a daily source of consternation for any pathologist who deals with significant numbers of skin biopsies, and this frustration is not without justification. Published clinical practice guidelines recommend excisional biopsy with narrow (2 mm) clinical margins as the standard approach for biopsy of clinically concerning pigmented lesions.\(^{235}\) The essential low-power assessment of histologic features such as size, symmetry, and circumscription of the lesion cannot be achieved on a partial sample (Figure 12), and heterogeneity within melanocytic lesions is well recognized, including areas of evolving melanoma in a nevus or foci of regression where diagnostic changes of melanoma are absent.\(^{126,236}\) Partial sampling, particularly punch biopsy sampling, has been clearly demonstrated to be associated with an increased risk for the misdiagnosis of melanoma.\(^{2}\) In one Australian study,\(^{237}\) the odds ratio for misdiagnosis was 16.6 times with punch biopsy sampling compared with excisional biopsies, and 20.4 times for a misdiagnosis with an adverse clinical outcome. It is also worth noting that partial biopsy of...
melanocytic lesions is a recurrent cause of litigation. An argument can be made for saucerization shave biopsy (aiming for complete excision of the lesion), particularly for lesions with a relatively low clinical suspicion of melanoma (aiming for complete excision of the lesion), particularly for lesions with a relatively low clinical suspicion of melanoma or deeper extension. However, use of this technique appears to be most successful for specialist practitioners. Of course, there are some circumstances in which excisional biopsy cannot be performed or is inappropriate, including broad lesions on the face or functionally sensitive sites (e.g., the soles of the foot). Punch biopsy or incisional biopsy to confirm a diagnosis prior to definitive management might be reasonable in these circumstances, but it is essential that this information be conveyed to the reporting pathologist.

Unfortunately, it is an all-too-common experience to be faced with a melanocytic proliferation that extends to the edges of the biopsy specimen, with no accompanying information as to whether it was intended to represent complete removal, and it is frequently difficult to identify on histologic features alone whether a lesion has been completely encompassed within the sample. Our approach in these situations is to append our reports with a comment tailored to the specific situation. If the histologic features in the biopsy are compatible with a nevus, we will indicate that, but we will also suggest that correlation with the clinical appearances would be of value to ensure that the biopsy appearances are representative. On the other hand, if there are any histologic features of concern (e.g., a prominent junctional component to a lesion in sun-damaged skin) we will at least be discussed with patients by an expert who is able to adequately explain the issues, including any influence the procedure may have on eligibility for clinical trials. To this end, we routinely append reports to nonspecialist practitioners for melanomas in the groups for which sentinel node biopsy has been advocated with a comment suggesting that the possibility of sentinel node biopsy should be discussed. The usual result of these comments is to prompt referral to either a specialist or a multidisciplinary team, which we regard as entirely appropriate.

Communication of Clinically Important Findings

The adequate communication of histopathologic findings is a critical component of clinical practice, and physician dissatisfaction with pathology laboratories is often related to poor communication, including the notification of significant abnormal results. There are published guidelines regarding the reporting of critical diagnoses, although by necessity these are relatively general in nature. In the specific setting of melanocytic lesions, a diagnosis of melanoma is clearly significant and requires prompt communication to the referring clinician to ensure timely and appropriate ongoing management. For similar reasons, any atypical melanocytic proliferation that requires further excision might also be considered in the same light. Although for dermatopathologists such cases represent a common event, this may not be the case for the referring clinician, who may expect more prompt notification. Thus, some thought should be given to the means by which this can be achieved, while at the same time avoiding overly onerous demands on pathologist time. The specific means will differ for each laboratory. In this context, it is also worth noting the demonstrated benefit of clinicopathologic correlation in a multidisciplinary setting, and thus, where facilities for multidisciplinary input are available, they should be used.

To improve communication, there have been recent efforts to establish a standardized classification scheme, linked to management recommendations, for melanocytic lesions, as is commonly used in other areas of surgery and cytopathology. Although we do not use this in our diagnostic reports at this time, such a framework can be very valuable in crystallizing concepts of risk and appropriate management for challenging lesions and in discussion with colleagues.

Sentinel Node Biopsy

In many countries, including our own, sentinel node biopsy is currently regarded as part of the appropriate management for melanomas of intermediate thickness (usually defined as a Breslow thickness of 1–4 mm), and possibly for a subset of high-risk thin melanomas. Often-cited reasons for this are the prognostic value of the sentinel node status, improvement in disease-free survival, and a possible improvement in overall survival for a subgroup of patients with clinically occult nodal disease. More recently, the emergence of effective adjuvant therapy and clinical trials of adjuvant therapy have increased the premium on identification of patients with sentinel node metastases, including in patients with thick melanoma. It should be noted, however, that not all authors agree with the routine performance of sentinel node biopsies, arguing that without evidence of an overall survival benefit it is unjustified, as well as raising questions regarding some of the aforementioned conclusions. Regardless of this ongoing debate, it seems reasonable that the procedure should at least be discussed with patients by an expert who is able to adequately explain the issues, including any influence the procedure may have on eligibility for clinical trials. To this end, we routinely append reports to nonspecialist practitioners for melanomas in the groups for which sentinel node biopsy has been advocated with a comment suggesting that the possibility of sentinel node biopsy should be discussed. The usual result of these comments is to prompt referral to either a specialist or a multidisciplinary team, which we regard as entirely appropriate.

CONCLUSIONS

Although usually straightforward, the assessment of melanocytic lesions can occasionally be exceedingly difficult. When combined with the relatively high stakes, this results in difficult melanocytic lesions making up the large bulk of cases seen in dermatopathology consultation practice. Here
we have tried to provide a practical approach to the assessment of these lesions, based on the available literature as well as our personal experience. We will conclude with 2 pieces of advice that can be applied across the spectrum of melanocytic lesions, and that, if followed, should go a long way towards minimizing adverse outcomes for our patients. The first is never to be hesitant to seek the opinion of a trusted colleague for any melanocytic lesion. Differences in opinion are to be expected in this group of lesions, but even the existence of contrasting opinions can be useful, as it confirms the lesion as difficult and will likely lead to appropriate management. This leads us to the final word of advice: if in doubt, cut it out. If a lesion is sufficiently clinically concerning to warrant biopsy, it should generally be removed in its clinical entirety. Despite the numerous reasons that make this desirable, every day brings multiple examples of partial sampling of such lesions. We have seen many cases in which adverse outcomes could have been prevented, or at least the patient reassured that appropriate management was undertaken, if complete excision had been performed. If the histologic features on a punch biopsy or other partial sample are anything other than clearly those of a banal nevus, excision to ensure the lesion has been removed in its clinical entirety. Despite the numerous reasons that make this desirable, every day brings multiple examples of partial sampling of such lesions. We have seen many cases in which adverse outcomes could have been prevented, or at least the patient reassured that appropriate management was undertaken, if complete excision had been performed. If the histologic features on a punch biopsy or other partial sample are anything other than clearly those of a banal nevus, excision to ensure the lesion has been completely removed provides the best available protection for the pathologist, the clinician, and, most importantly, the patient.

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