The histological reporting of melanoma

Eduardo Calonje

Abstract

The incidence of malignant melanoma has increased steadily over the past 30 years and this type of malignancy is the leading cause of death from cutaneous malignant disease. Cutaneous malignancies, including melanoma, can be detected at a very early stage and a cure is possible with prompt detection and treatment. In recent years, and mainly because of increased awareness of the early detection of melanoma, histopathologists have been exposed more and more to melanocytic lesions. Therefore, it is essential that histopathologists are able to provide a report to the clinician that conveys relevant information in a concise and precise manner. This paper provides a set of guidelines aimed at helping histopathologists with the gross and microscopic description and diagnosis of malignant melanoma.

Information required in the request form

(1) Name of the patient.
(2) Date of birth.
(3) Hospital number.
(4) Name of surgeon/clinician and address to which the report should be sent.
(5) Clinical details including sex, site and accurate description of lesion (outline, colour, measurement).
(6) Primary, recurrent, or metastatic lesion.
(7) Patient with or without atypical mole syndrome.
(8) Family history of skin cancer, particularly malignant melanoma.
(9) Type of biopsy (shave, punch, simple excision, wide excision).
(10) Date of biopsy.

All specimens should be adequately fixed in neutral buffered formalin and fixation time should be around 24 hours. Small biopsies can be allowed less fixation time if results are needed urgently. Frozen sections or cytology should not be requested in the diagnosis of primary or recurrent cutaneous malignant melanoma. Suspicious melanocytic lesions should be removed by an excisional biopsy with narrow margins including subcutaneous fat. Small biopsies such as a punch are only justified in exceptional circumstances, as a diagnostic tool for very large lesions or lesions in difficult locations (subungual). Incisional biopsies do not seem to increase the risk of recurrence or metastasis. However, incisional biopsies not only make the evaluation of important prognostic factors (particularly depth of invasion) difficult, but are often non-representative and might lead to errors in diagnosis.

Gross description

(1) Type of biopsy.
(2) Dimensions of the specimen in millimetres (length, breadth, thickness).
(3) If the specimen is marked with stitches, describe their position in detail.
(4) Length and breadth of pigmented lesion in millimetres.
(5) Symmetry, regularity, colour, and outline of pigmented lesion.
(6) Presence or absence of nodule and measurement of the latter.
(7) Presence or absence of ulceration.
(8) Associated lesions (scar, areas of depigmentation).
(9) Measurement of distance between edge of lesion and nearest excision margin.
(10) If possible, photograph the macroscopic specimen.

All specimens from excisional biopsies should be sectioned transversely in a "sliced bread" pattern (fig 1A). The first section should be taken from the centre of the lesion and this should be followed by sectioning the whole specimen every 2–3 mm. All of the specimen should be processed. The use of cruciate section sampling is not recommended because this only allows for the evaluation of part of the specimen in some of the blocks. The latter makes assessment of the architecture difficult (fig 1B). Punch biopsies including small lesions can be embedded intact or cut through the middle and both halves embedded and processed.

For many years, it has been recommended that re-excision specimens should be processed completely, with cutting of blocks every 2–3 mm. However, if a lesion has been excised completely, and there are no macroscopic abnormalities other than a scar, processing of the whole specimen does not seem to offer any additional information and increases the costs for the laboratory. Therefore, it seems more logical to take a single representative section from the centre of the specimen. If the original lesion was excised incompletely, or there is macroscopic evidence of a residual lesion, it is recommended that blocks every 2–3 mm through the whole scar are cut and embedded.

**Microscopic description**

(1) Histogenetic subtype: superficial spreading melanoma; lentigo maligna melanoma; acral lentiginous melanoma; nodular melanoma; desmoplastic; or unclassifiable.

**Histogenetic Subtype**

Traditionally, every effort is made to try to classify a given melanoma into one of several well defined histogenetic subtypes (superficial spreading, lentigo maligna, acral lentiginous, nodular). Each of these types defines a pattern of radial growth phase (see below) and, except for nodular melanoma, all have a biphasic growth pattern. By definition, nodular melanoma is always in vertical growth phase and lacks a recognisable radial growth phase. It has increasingly been recognised that classification of melanomas into histogenetic subtypes has no significant prognostic relevance. However, its use is still recommended because these subtypes define well known clinicopathological entities. Assigning a given lesion into a particular subtype should be based on strict histological criteria and not on isolated attributes such as location. Not uncommonly, a melanoma cannot be classified into a specific histogenetic subtype and these lesions should be put into a separate category (unclassified). Any histogenetic type of melanoma can develop a prominent desmoplastic component, but this is most commonly seen in lentigo maligna melanoma and acral lentiginous melanoma. A melanoma should only be classified as desmoplastic if this component predominates. In most cases, desmoplastic and neurotropic melanomas are closely associated. It has been suggested that desmoplastic melanomas have a higher recurrence rate, a lower incidence of lymph node metastasis, and a better prognosis than conventional melanomas. However, this has been challenged recently in a large series showing that there is no difference between the recurrence and survival rate of desmoplastic melanoma and other cutaneous melanomas.
that ulceration is not the result of trauma. This is not always possible and might require close clinicopathological correlation.

**Clark’s Levels of Invasion**

There are five categories of invasion: (1) Level I. The melanoma cells are confined to the epidermis. (2) Level II. Refers to invasion of the papillary dermis by melanoma cells. However, these cells do not fill and expand the papillary dermis. Most tumours classified as level II are in radial growth phase. (3) Level III. Tumour cells expand and fill the papillary dermis. These melanomas are almost always in vertical growth phase. (4) Level IV. Melanoma cells infiltrate the reticular dermis. (5) Level V. Tumour cells infiltrate the subcutaneous tissue. It has been clearly demonstrated that increasing levels of invasion correlate with a decrease in survival. However, although this is true when this feature is analysed as a single variable, the same result has not been obtained in most multivariate analyses.

**Tumour Thickness**

The depth of invasion is measured in millimetres from the top of the granular cell layer to the deepest melanoma cell. Measurement can be difficult in cases with ulceration or prominent epidermal hyperplasia. In the former situation, the thickness of the tumour should be measured from the base of the ulcer to the deepest melanoma cell. When epidermal hyperplasia is present, the depth of invasion might be overestimated, and this should be pointed out in the histological description. Involvement of adnexal structures by melanoma cells should not be included in the measurement. Tumour thickness is the single most important prognostic indicator in melanoma. However, a small number of patients with thin melanoma die of disease and some patients with thick melanoma survive.

**Mitotic Rate**

There is no standard method for counting mitoses and, until recently, the mitotic rate was expressed as the number of mitoses/10 high power fields. The size of the field varies in different microscopes and this method is therefore inaccurate. In recent years, the preferred method is to report the mitotic count/mm². When a mitotic count is performed, sampling error should be avoided. Therefore, fields with the highest number of mitotic figures should be identified before the count is done. The mitotic rate correlates inversely with survival and it has been found to provide independent predictive prognostic information.

**Host Immune Responses**

This attribute is difficult to evaluate and to standardise. It has been proposed that the presence of a host immune response should be divided into infiltrative (tumour infiltrating lymphocytes) or non-infiltrative. This is because the presence of tumour infiltrating lymphocytes is an important independent pre-
dictive prognostic factor in melanoma—the presence of such lymphocytes correlates with improved survival.

REGRESSION
This feature is often seen in melanoma and it is characterised in its early stages by a prominent mononuclear cell infiltrate. This is followed by the destruction of tumour cells and melanophages, vascular proliferation, and fibrosis. The association between the presence of regression and prognosis is controversial. Although the reports are contradictory, it seems that the rate of metastasis is higher in thin melanomas with extensive regression.19

MICROSATELLITES
These are defined as the presence of aggregates of melanoma cells away from the main tumour. These aggregates have been defined arbitrarily as being \( \geq 0.05 \) mm in diameter. Serial sections should always be examined to make sure that there are no connections to the main tumour. Microsatellites probably represent local metastases. It is controversial whether the presence of microsatellites is an independent prognostic factor.11 However, they do appear to be associated with an increase risk of local recurrence.

VASCULAR INVASION
This feature is not an independent factor in predicting prognosis in patients with melanoma.11 Vascular invasion can be difficult to identify on haematoxylin and eosin stained sections and false positives are often a problem. With the use of immunohistochemistry, it is possible to increase the detection of vascular invasion, but this is an expensive procedure that might require evaluation of multiple sections and offers little advantage.

PERINEURAL INVASION
Infiltration of nerves by tumour cells should be recorded. A melanoma should only be classified as neurotropic if this is the dominant feature.

PREDOMINANT CELL TYPE
This feature is described mainly for the sake of completeness, but there is some suggestion that melanomas with a pure spindle cell morphology have a better prognosis.2 However, this has not been validated adequately.

ASSOCIATION WITH A COEXISTENT BENIGN MELANOCYTIC LESION
Recording this finding can provide useful information for epidemiological studies. Often, it is very difficult to decide whether there is a residual benign component in a given lesion.

ASSESSMENT OF EXCISION MARGINS
Measurement of the distance from both lateral and the deep excision margins should be provided in millimetres.

Special techniques
Immunohistochemical stains including S-100, HMB-45, and MART-1 are useful in the differential diagnosis between melanocytic and non-melanocytic tumours. They should not, however, be used to distinguish between benign and malignant melanocytic lesions. The importance of calculating the proliferation index with antibodies such as MIB-1 has not been adequately assessed and it should therefore not be used in the routine reporting of melanocytic lesions.

Since the introduction of immunohistochemistry, electron microscopy has been relegated as an aid in the differential diagnosis of poorly differentiated melanocytic tumours. At present, electron microscopy has no role in the routine reporting of melanocytic lesions.

Other techniques that have been used in the evaluation of melanocytic lesions include DNA flow cytometry and cytogenetics. DNA ploidy11 has little value in the evaluation of melanocytic lesions because many melanomas have a diploid DNA content. Cytogenetics is still an experimental technique and its value as an aid in the diagnosis of melanocytic tumours remains to be proved because of the heterogeneity of these lesions.