Standard step sectioning of skin biopsy specimens diagnosed as superficial basal cell carcinoma frequently yields deeper and more aggressive subtypes

To the Editor: Correct diagnosis of superficial basal cell carcinoma (BCC) is essential because of the increase of nonsurgical treatments for this subtype. Histologic confirmation by punch biopsy specimen for BCC diagnosis and its subtype is recommended. Although studies have shown the discordance between punch biopsy specimens and excision, there is a lack of research on the sampling error within punch biopsy specimens.[1,2] Moreover, a standardized method for histologic sectioning punch biopsy specimens is currently missing. The current histologic examination protocol of a 3-mm punch biopsy specimen, suspicious for BCC, at the Radboud University Medical Center (Nijmegen, The Netherlands) consists of evaluation of 2 to 3 hematoxylin-eosin-stained tissue sections (4 μm) obtained from 1 level. If a BCC is detected in these sections, no additional levels will be cut. If not, the punch biopsy specimen is cut at 4 additional levels and evaluated.

We compared the accuracy of histologic examination of only 1 level with a more extensive step-section method. In addition, we investigated whether tumor thickness (>0.4 mm), ulceration, and adnexal extension are determinants of treatment failure or recurrence in superficial BCCs.[3,4] This retrospective study was approved by the institutional review board of the Radboud University Medical Center, Nijmegen, The Netherlands.

In all, 116 superficial BCC punch biopsy specimens, obtained in 2014 to 2015, were cut in 4 additional levels at an interval of 200 μm. After every 200 μm, 10 sections (4 μm) were cut, of which the middle 2 sections were hematoxylin-eosin stained and evaluated. Finally, 5 levels per punch biopsy specimen were histopathologically evaluated (Fig 1). Tumor thickness was evaluated by measuring from the granular layer of the epidermis down to the deepest point of invasion (Breslow depth). Included patients were treated with imiquimod cream (n = 23), 5-fluorouracil (n = 19), excision (n = 50), or methylaminolevulinate photodynamic therapy (MAL-PDT) (n = 24) with a follow-up time until January 29, 2016 (Supplemental Table I).

In 22.4% (n = 26) a more aggressive BCC subtype was found in the additional hematoxylin-eosin-stained sections (Table I, Supplemental Table II, and Supplemental Fig 1). There were 2 clinical recurrences (5-fluorouracil, n = 1, and MAL-PDT, n = 1) and 2 treatment failures (MAL-PDT). In the treatment failure/recurrence group there was 1 underdiagnosed BCC (25%); this was the only superficial BCC thicker than 0.4 mm in this group, whereas none showed presence of ulceration or adnexal extension

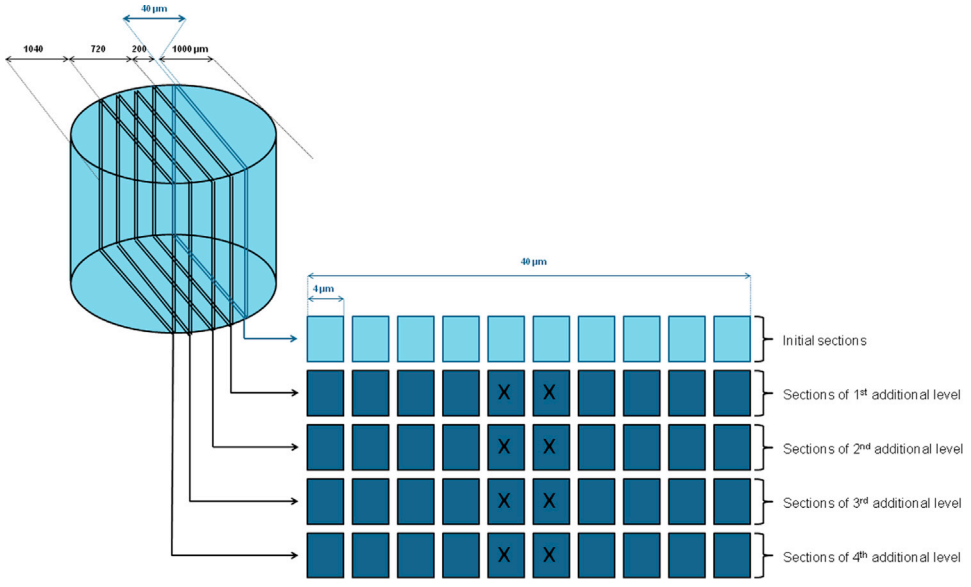

Fig 1. Histopathological examination method of a punch biopsy specimen suspicious for basal cell carcinoma. Schematic overview of the histopathological examination process. The dimensions of the punch biopsy specimen might be smaller than depicted in this figure because of shrinkage after formalin fixation and histologic processing, embedding, and mounting. **X**: These sections are stained with hematoxylin-eosin and used for histopathological examination. In the initial sections it is unknown which slices are stained and examined.

Table I. Histopathological diagnosis and clinical treatment failure or recurrence

Histopathological diagnosis of punch biopsy specimen, n (%)*								
sBCC	nBCC	iBCC	mnBCC	n/mnBCC	n/iBCC	n/mn/iBCC	Total	More aggressive subtype missed
90 (77.6)	16 (13.8)	1 (0.9)	1 (0.9)	4 (3.4)	1 (0.9)	3 (2.6)	116 (100.1†)	26 (22.4)

Clinical treatment failure‡ or recurrence§									
Patient	Treatment failure or recurrence	Location	Presence of clinical ulceration	Presence of adnexal extension	sBCC thickness, mm‖	Initial treatment	Histopathological diagnosis of second punch biopsy specimen¶	Histopathological diagnosis of excision specimen	Histopathological diagnosis of punch biopsy specimen#
1	Recurrence	Lower extremities	No	No	0.32	5-FU	sBCC	sBCC	sBCC
2	Recurrence	Lower extremities	No	No	0.20	MAL-PDT	sBCC	sBCC	sBCC
3	No response	Trunk	No	No	0.33	MAL-PDT	Not performed	sBCC	sBCC
4	Partial response	Head	No	No	0.74	MAL-PDT	n/mnBCC	n/mnBCC	sBCC

FU, Fluorouracil; *iBCC*, infiltrative basal cell carcinoma; *MAL-PDT*, methylaminolevulinate photodynamic therapy; *nBCC*, nodular basal cell carcinoma; *n/mnBCC*, micronodular basal cell carcinoma; *sBCC*, superficial basal cell carcinoma.

*Based on histopathological evaluation of 5 levels per punch biopsy specimen, all biopsy specimens based on 1 level evaluation were diagnosed as sBCC.

†Numbers do not add up to 100 because of percentages round off.

‡Clinical treatment failure: partial or no response after initial treatment.

§Presence of tumor tissue detected during follow-up after previous tumor clearance.

‖Largest measurement taken from either the initial or additional hematoxylin-eosin-stained sections.

¶Extra punch biopsy specimen taken after treatment failure/recurrence for diagnosis and determination of excision margin for treatment.

#Based on histopathological evaluation of 5 levels per punch biopsy specimen.

(Table I). We did not found an association of tumor thickness, ulceration, or adnexal extension with treatment failure/recurrence. The mean follow-up period was of 312 ± SD 194 days with a median of 301 days.

Hoogedoorn et al[5] found that more than 50% of the treatment failures of superficial BCC, after MAL-PDT, were a result of underdiagnosis of the primary punch biopsy specimen and that in approximately 50% of the recurrences a mixed type BCC was present. Their median follow-up was 2 years, in which they reported higher rates of treatment failures and recurrences. Our limited follow-up time might explain the current lack of association between presence of more aggressive BCC subtypes in punch biopsy specimens and treatment failure/recurrence.

This study shows that histologic examination of only 1 level from a punch biopsy specimen leads to underdiagnosis of more aggressive BCC subtypes in biopsy specimens diagnosed as superficial BCCs. We recommend step sectioning to reduce this risk and to prevent undertreatment.

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