Acta Derm Venereol 2016; 96: 426–427

CORRESPONDENCE

A Place for *BRAFV600E* Mutation-specific Immunohistochemistry Alongside Cell-free DNA Mutation Detection in Melanoma

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Accepted Nov 12, 2015; Epub ahead of print Nov 16, 2015

Sir,

We read with interest the article by Ashida et al. (1), describing a patient with metastatic melanoma who presented with a *BRAFV600E* mutation diagnosed only in circulating cell-free DNA with no detected mutation in the primary formalin-fixed and paraffin-embedded (FFPE) tumour. Molecular analysis of FFPE melanoma samples, primary or metastatic, is the main method of diagnosing a *BRAFV600* mutation, enabling anti-BRAF targeted therapy. Nevertheless, detection of *BRAF* mutations in circulating cell-free DNA offers a good opportunity to progress the monitoring and diagnosis of patients with metastatic melanoma, especially in cases with challenging metastatic sites with surgical issues that impair tumour sampling (2, 3). However, this new method requires heavily-specialized molecular genetics platforms. As an alternative, mutation-specific immunohistochemistry, a widespread and rapid technique available in most pathology laboratories, allows the detection of BRAFV600E mutated protein with excellent sensitivity and specificity, especially in challenging melanoma samples containing few tumour cells (4–7) (see Fig. 1 for an example from our laboratory). We hypothesize that anti-BRAFV600E immunohistochemistry, performed on the primary melanoma, could have enabled a conclusion to be reached at the time of diagnosis in the case of a *BRAFV600E* mutated melanoma reported by Ashida et al. (1), without requiring cell-free tumour DNA analysis.

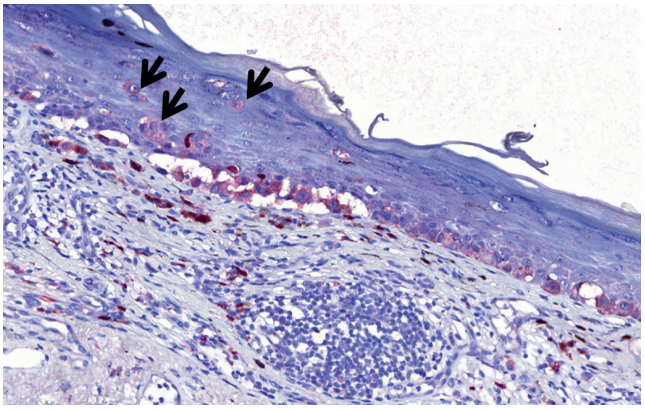

Fig. 1. Cell-poor melanoma sample stained with anti-BRAFV600E immunohistochemistry. The lateral component of this *BRAFV600E*-mutated superficial spreading malignant melanoma showed immunostaining of junctional and trans-epidermis pagetoid ascending (*arrows*) melanoma cells. The high number of non-tumour cells prevented detection of the *BRAFV600E* mutation through molecular pyrosequencing analysis (haematoxylin counter-staining, original magnification ×25).

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 doi: 10.2340/00015555-2286
 ISSN 0001-5555

Reply to the Correspondence by A. Uguen et al.

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We thank Uguen et al. for their comments and would like to take this opportunity to reply.

Detection of *BRAF* mutations is important for melanoma treatment. As noted by Uguen et al., mutation-specific immunohistochemistry is beneficial in detecting tumour cells with $BRAF^{V600E}$ mutation. We did not intend to exclude the possibility that immunohistochemistry would allow the detection of a specific mutation in the primary lesion in our case. On the other hand, analytical methods for circulating cell-free DNA usually require highly specialized platforms. However, castPCR (Competitive Allele-Specific TaqMan polymerase chain reaction) does not require advanced procedures or specific instruments other than a real-time PCR system. In fact, castPCR reagent kits are available commercially. We consider castPCR to be a readily accessible technique, which could help in identifying melanoma patients who would benefit from targeted anti-BRAF therapy.