British Association of Dermatologists Project Grant Report
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I am extremely grateful to the BAD for awarding me a project grant prize, as it has allowed me to expand on my intercalated degree research project, and further my interest in dermatology and pathology.

My intercalated degree research project aimed to identify the reliability of mitotic counting in pT1 melanomas. This was of interest to me as only *one* dermal melanocyte mitoses is needed to upstage a pT1a melanoma to pT1b, which is clinically relevant as the management of pT1a and pT1b melanomas differ significantly. Patients with pT1a melanomas have a recommended follow-up duration of up to one year, versus five years if the melanoma is pT1b.[1] In addition, whereas guidelines state that a sentinel lymph node biopsy can be offered to patients with a pT1b melanoma, they do not state this for patients with pT1a melanomas.[1]

To investigate reliability of mitotic counting, five *additional* haematoxylin & eosin (H&E) sections were cut from twenty one previously diagnosed pT1a melanomas. Nine out of twenty one (43%, 95% CI 24-63%) melanomas were found to contain mitoses in the additional H&E sections. This led to the following question: were mitoses identified more because of variability in the distribution of mitoses in melanoma tissue, or because we were analysing melanomas which contained mitoses in the original H&E sections that had been missed?

To answer this, my supervisors and I analysed the original melanoma sections for mitoses during the summer. After excluding melanoma cases with dermal melanocyte mitoses in the original sections, seven out of eighteen (39%, 95% CI 20-61%) melanoma cases were still found to contain mitoses in the additional H&E sections, suggesting that staging of pT1 melanomas is significantly influenced by the distribution of mitoses in the melanoma tissue. Our results observed that this results in understaging of melanomas; some patients might not be receiving accurate prognostic information, offer for sentinel lymph node biopsy, as well as the recommendation durations suggested for follow-up. Thus, better methods of mitotic counting, which take into account the variability of distribution of mitoses, should be investigated to improve reliability of mitotic counting.

It was also evident during the intercalated degree project that there was subjectivity in distinguishing which cells were mitotic and melanocytic. Therefore, immunohistochemistry was performed on 3 sections per melanoma case to double-label mitotic (using phosphohistone H3 staining) and melanocytic (Melan-A staining) cells. We were surprised to find not a single immunohistochemical section contained mitoses, whereas six out of eighteen melanomas contained mitoses, in the three H&E sections that were selected for comparison (selection of H&E sections to be compared was done without prior knowledge of which sections contained mitoses). This corresponded to a statistically significant difference ($p=0.0191$, Fisher's exact test) between H&E and PHH3/Melan-A in the upstaging of pT1a melanomas. The results suggest that PHH3/Melan-A could potentially improve the accuracy of melanoma staging, by allowing more accurate recognition of melanocytes and mitoses. Therefore, long term studies should clarify whether PHH3/Melan-A (or indeed other combinations of stains which label mitoses and melanocytes) has a higher utility than H&E in melanoma staging, by comparing the prognostic value of different cut-offs of mitotic rate for H&E and PHH3/Melan-A stained melanoma sections.

Reference

1. Marsden JR, Newton-Bishop JA, Burrows L, et al. (2010) Revised U.K. guidelines for the management of cutaneous melanoma 2010. Br J Dermatol. **163**, 238–256