The significance and evaluation of the expression of PRAME in melanocytic lesions.

Karolina Wojcik, Dr Guy Orchard, Dr Nastassia Nardini, Dr Eduardo Calonje

St. John's Institute of Dermatology, Guy's and St. Thomas NHS Foundation Trust/Viapath Analytics/United Kingdom, SE1 7EH

The University of Greenwich, Biomed Online, Faculty of Engineering and Science, ME4 4TB

Keywords

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Introduction

According to Cancer Research UK statistics (2018) malignant melanoma is the fifth most common cancer in the UK, and each year there are around 16,000 people who are diagnosed with this deadly type of skin cancer.

Cutaneous melanocytic lesions can be divided into benign, and atypical melanocytic naevi, and the most aggressive type of skin malignancy, malignant melanoma (MM).

The study aims to evaluate the staining pattern of PRAME expression in melanocytic tumours and highlights PRAME expression in primary and metastatic melanomas as well as other melanocytic lesions.

This research looks at the potential to explore the PRAME expression in melanocytic nevi, which could prove an invaluable input to distinguish the benign/ atypical naevi from melanomas.

PRAME is a melanoma-associated antigen that was first identified through analysis of the specificity of T cell clones in a patient with metastatic melanoma.

The employment of PRAME antibody in a diagnostic laboratory can prove invaluable addition in the clinical setting distinguishing benign and malignant melanocytic lesions.

Materials and Methods

A total of 308 melanocytic tumours were examined for immunohistochemical expression of PRAME, including 157 malignant melanomas and 151 melanocytic naevi. All IHC staining was performed on a Roche BenchMark Ultra fully automated immunostaining platform using commercially available antibody to PRAME (ABCAM) with a optimum dilution factor of 1:2000.

Each case has also been stained with Haematoxylin and Eosin (H&E). The negative control was also run along the slides to determine the specificity of an antibody. No staining was observed when the primary antibody was omitted.

The scoring involved : Zero indicates no staining at all(negative), staining of 1% to 50% of tumour cells score as 1+, labelling of 51% or more of tumour cells score as 2+.

The Chi-Square statistical analysis was performed using GraphPad Prism software, P-value was generated to determine the sensitivity (labelling tumour cells) and specificity (distinction of naevi from melanoma).

Results

The nuclear labelling for PRAME was recorded in 77.7% of primary melanomas. The primary melanoma PRAME IHC expression was found in 80% of all melanoma in situ. According to melanoma histological subtypes, PRAME was expressed in 83.9% of superficial spreading melanoma 100% of Lentigo maligna melanoma, Acral lentiginous melanoma (ALM) and Nodular melanoma.

Invasive melanoma reported 82.6% PRAME expression levels; naevoid melanoma expressed PRAME in 57.1% of cases, other types of melanoma stained positive for PRAME in 50% of cases.

The PRAME IHC expression was found to be lowest for Desmoplastic (including Spindle cell Melanoma) - 42.9% of cases (Chart 1)

The Chi-square test of anti-PRAME IHC positive expression in all malignant melanoma subtypes studied here, revealed the p- value of 0.0294, which is statistically significant.

Chart 1- PRAME expression in Malignant Melanomas subtypes

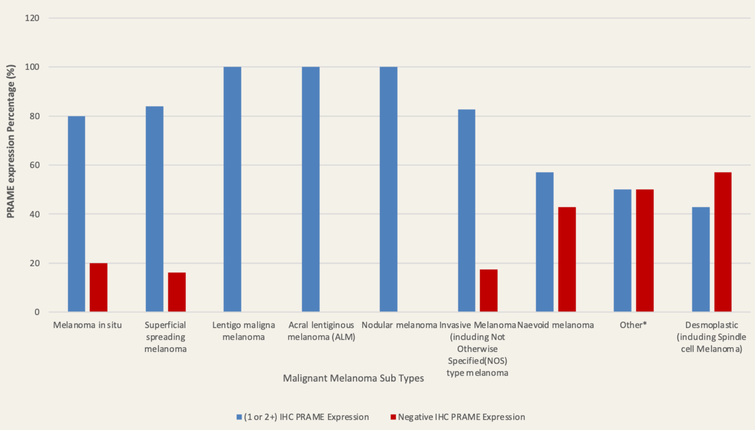


Figure 1A

Figure 1B

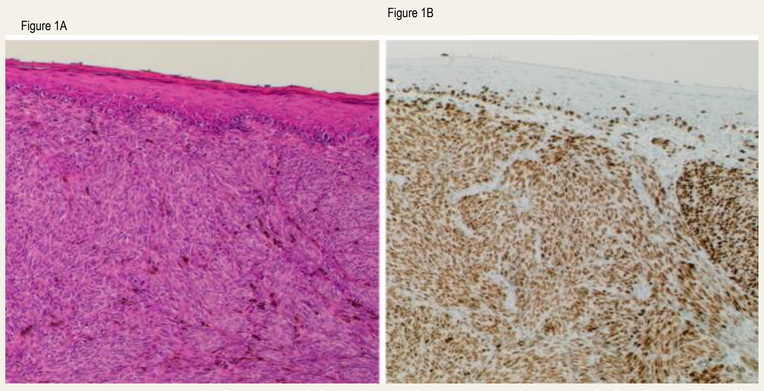


Figure 1(A-B) Invasive Melanoma NOS- Fig.1A H&E staining shows epithelioid/spindle cell tumour Fig.1B demonstrates diffusely and equally immunopositivity for anti-PRAME (2+) nuclear staining both in situ and invasive melanoma. (Magnification x100)

Chart 2- PRAME expression in Melanocytic Naevi

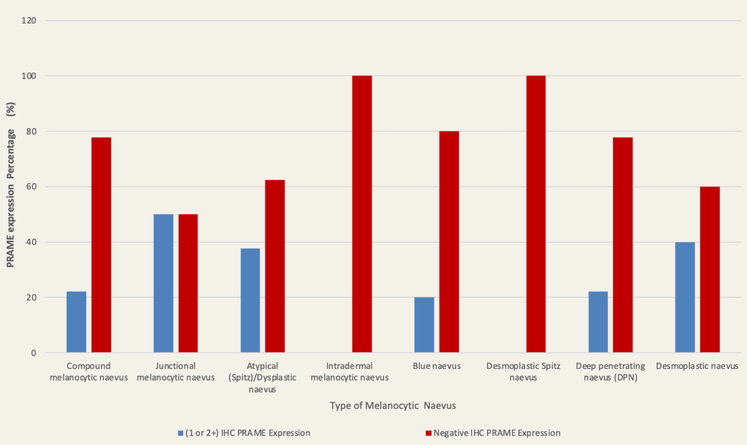


Figure 2A

Figure 2B

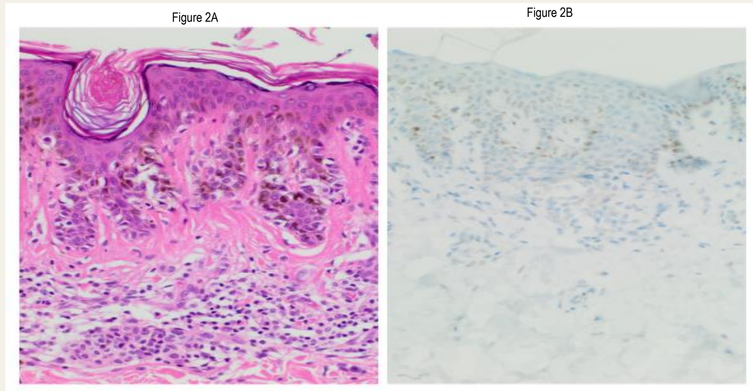
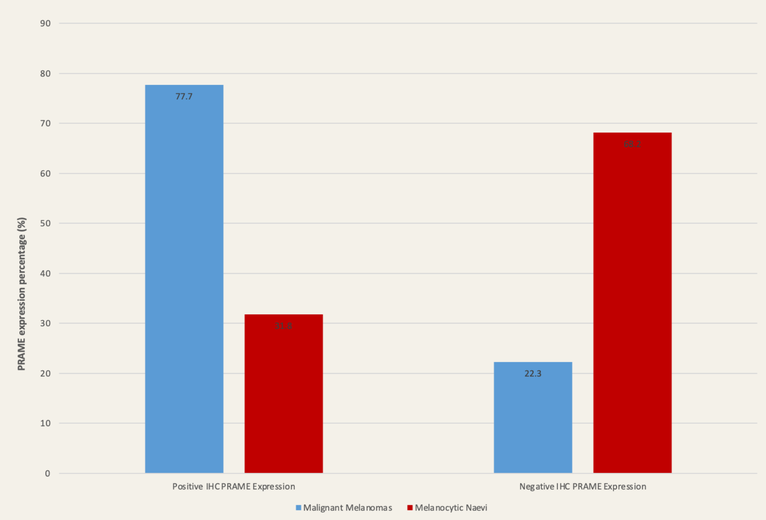


Figure 2(A-B)-Dysplastic melanocytic naevus- (Fig.2A) H&E staining reveals lightly asymmetrical, compound, non-ulcerated melanocytic naevus with a dysplastic (Fig.2B) shows an absence of anti-PRAME expression. (Magnification x100)

Chart 3: PRAME IHC expression percentage in Malignant Melanomas and Melanocytic Naevi



H&E staining and PRAME expression

Figure £A

Figure 3B

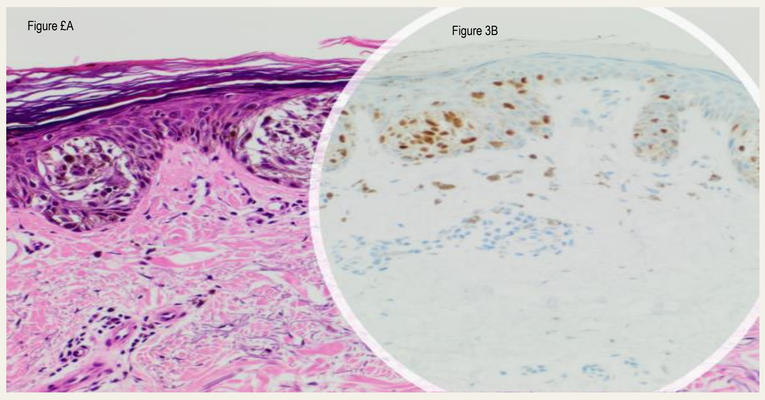


Figure 3(A-B)- (Fig. 3A) H&E staining demonstrating melanoma in situ Fig.3B shows nuclear labelling of tumour cells that are anti-PRAME positive(1+) in the epidermis component. (Magnification x100)

Figure 4A

Figure 4B

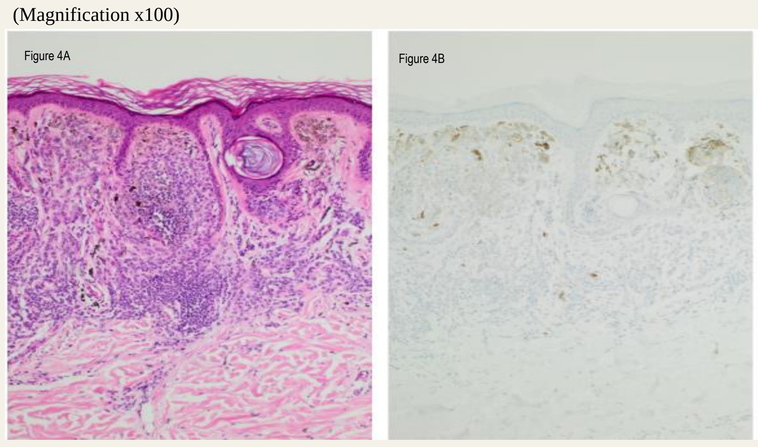


Figure 4(A-B)- (Fig. 4A) Compound melanocytic naevus- - (Fig 4A) H&E staining reveals a symmetrical and large, superficial, compound melanocytic naevus.

Fig. 8B shows lack of staining for anti-PRAME antibody. Component, however there is presence of a melanin pigment(brown deposits) (Magnification x100)

Discussion

The total of 157 cases of malignant melanoma that were studied the results demonstrate the 122 of these (77.7%) prove that PRAME is frequently expressed in malignant melanoma.

This correlates with the study conducted by (Lezcano et al., 2018) whereby diffuse nuclear immunoreactivity for PRAME was found in 87% of metastatic and 83.2% of primary melanomas.

Lezcano and colleagues (2018) emphasized the need for an alternative treatment for patients with unresectable melanoma stage IV in whom other treatments such as BRAF V600E inhibitors have failed and the clinical trials stage I involving the patients with PRAME-positive metastatic melanoma are currently underway (Clinicaltrials.gov, 2018).

The expression of anti-PRAME may demonstrate its valuable aspect in difficult cases of malignant melanoma as this research study outlined PRAME IHC expression in a variety of melanomas.

Out of 68.2% of all melanocytic naevi studied lacked any PRAME expression.

Majority of the PRAME positive (37.6%) cases in naevi encountered in this project, were, in fact, atypical Spitz naevi as they may possess a more prominent risk for malignant behaviour.

An investigation by Kim et al., 2015 described the difficulty in diagnosis Spitz (Spitzoid) melanoma and Spitz naevus. The presence of atypical Spitz naevus and its sharing histological hallmarks with melanoma, is associated with the higher risk of developing malignant melanoma.

During the evaluation and discussing of findings of PRAME expression in naevi, there has been a consensus that positive immunoreactivity of PRAME in such cases does not mean malignancy.

PRAME expression would be an auxiliary tool coupled with other clinical findings such as dermoscopy, histological features, clinical setting, genetic profile data, abnormal FISH findings that are present in metastatic melanoma but not naevi, to reach the diagnosis of melanoma.

PRAME is a member of the family of cancer-testis antigens (CTA), and an attractive target for immunotherapy.

Conclusion

PRAME is preferentially expressed in cutaneous malignant melanomas, as well as other tumours such as breast carcinoma, renal cell carcinoma, therefore normal tissue are not known to express it apart from testis, endometrium and adrenals.

Thus, by recognising and distinguishing the malignant melanomas from atypical naevi and subsequent diagnostic problems, utilization the anti-PRAME antibody adjunct to other markers, could establish the solution in diagnostically challenging cases.

Further studies are required to analyse the expression in different types of malignant tumours thoroughly such as lung, breast and colon cancers.

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