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DIFFERENTIAL EXPRESSION OF NICOTINAMIDE N-METHYLTRANSFERASE IN CUTANEOUS KERATOACANTHOMA AND SQUAMOUS CELL CARCINOMA: AN IMMUNOHISTOCHEMICAL STUDY.

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Keywords:	keratoacanthoma, immunohistochemistry, Nicotinamide N-methyltransferase, non-melanoma skin cancer, squamous cell carcinoma, tumor biomarker

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9 **Key words:** keratoacanthoma; immunohistochemistry; Nicotinamide N-methyltransferase; non-
10 melanoma skin cancer; squamous cell carcinoma; tumor biomarker.
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18 Salvolini¹, A. Campanati², A. Offidani², M. Emanuelli^{1, 4}.
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22 *Davide Sartini and Veronica Pompei contributed equally to this work.
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51 The authors have no conflict of interest to declare.
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3 Nowadays, we are witnessing a worldwide increasing incidence of non-melanoma skin cancers
4 (NMSCs), whose major types are basal cell carcinoma (BCC) and squamous cell carcinoma (SCC).
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6 Although the latter is several times less common than the basal cell type, it accounts for the majority
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8 of NMSC-related metastatic disease and death. Head and neck SCCs, in particular, are more likely to
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10 metastasize^{1,2}.
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14 As concerns keratoacanthoma (KA), some authors consider it as a self-resolving benign lesion, while
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16 others think that it is a precursor or a well-differentiated variant of SCC^{2,3}. A prompt differential
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18 diagnosis between these two neoplasms represents a primary goal, since KA, unlike SCC, shows an
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20 excellent prognosis, thanks to its tendency to spontaneous involution. From a histopathological point
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22 of view, KA and well-differentiated SCC are difficult to distinguish. Nevertheless, it has been recently
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24 reported that they exhibit distinct gene expression profiles⁴.
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28 The aim of our study was to evaluate for the first time in cutaneous KA and SCC the expression of
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30 nicotinamide N-methyltransferase (NNMT), which was previously found to be upregulated in
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32 different solid tumors, including clear cell renal cell carcinoma (ccRCC), oral SCC and melanoma,
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34 in order to assess its diagnostic and prognostic potential⁵⁻⁹.
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38 This retrospective study was conducted on a total of forty-eight specimens collected from patients
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40 undergoing biopsy or excisional surgery between 2016 and 2018, in accordance with the principles
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42 of the Declaration of Helsinki. In particular, 20 KA cases (7 males and 13 females; mean age: 79.19
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44 years; age range: 56-98 years; mean lesion diameter: 1.25±0.62 cm; diameter range: 0.4-2.5 cm), 18
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46 SCC cases occurred on the trunk and extremities (9 males and 9 females; mean age: 75 years; age
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48 range: 54-94 years; mean lesion diameter: 0.89±0.33; diameter range: 0.5-1.6 cm), and 19 SCC cases
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50 from head and neck (17 males and 2 females; mean age: 75.35 years; age range: 57-94 years; mean
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52 lesion diameter: 1.35±0.83 cm; diameter range: 0.4-3 cm) were analyzed.
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56 NNMT expression was evaluated by means of immunohistochemistry as previously described⁹.
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59 All analyses were independently performed by two investigators blinded to the patient group using a
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Nikon Eclipse E600 light microscope equipped with a Nikon DS-Vi1 digital camera. Agreement

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2
3 between observers was always >95%. Stained cells were counted in at least ten fields per sample
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5 (field: 0.07 mm², magnification 400×) and quantified as a percentage of the total counted cells. Each
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7 experiment was carried out three times in duplicate. Differences between groups were determined
8
9 using the Kruskal-Wallis and Dunn's multiple comparison tests. Correlations with clinic-pathological
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11 parameters were assessed using the Spearman test. A p-value less than 0.05 was considered
12
13 statistically significant.
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16 No statistically significant relationship was found between protein level and age, sex and lesion size.
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18 Our results evidenced a significantly higher enzyme expression in KA, where it showed a strong
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20 cytoplasmic staining, compared to SCC. In particular, in head and neck SCC NNMT
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22 immunoeexpression was moderate/good and the percentage of positive cells was significantly lower
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24 than in those of the rest of the body. Moreover, in tumors of the trunk and extremities the
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26 immunostaining was focal and faint (Figures 1 and 2). It is noteworthy that tumors with a less
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28 favorable prognosis showed decreased NNMT levels, in agreement with what was previously
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30 observed in other cancer⁵⁻¹⁰.
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35 Taken together, these observations lead us to hypothesize the existence of an inverse correlation
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37 between NNMT expression and tumor progression, thus suggesting a potential role of NNMT as a
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39 novel biomarker that could be used in the future for the early diagnosis and prognosis of these
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41 neoplasms, as well as for the development of targeted therapeutic strategies.
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REFERENCES

1. Apalla Z, Nashan D, Weller RB, Castellsagué X. Skin Cancer: Epidemiology, Disease Burden, Pathophysiology, Diagnosis, and Therapeutic Approaches. *Dermatol Ther* 2017;**7**:5–19.
2. WHO Classification of Skin Tumours. In: *WHO Classification of Tumours* (Elder DE, Massi D, Scolyer RA, Willemze R, eds), 4th edn. 2018;11.
3. Mandrell JC, Santa Cruz D. Keratoacanthoma: hyperplasia, benign neoplasm, or a type of squamous cell carcinoma? *Semin Diagn Pathol* 2009;**26**:150-163.
4. Ra SH, Su A, Li X et al. Keratoacanthoma and squamous cell carcinoma are distinct from a molecular perspective. *Mod Pathol* 2015;**28**:799–806.
5. Sartini D, Muzzonigro G, Milanese G, Pierella F, Rossi V, Emanuelli M: Identification of nicotinamide N-methyltransferase as a novel tumor marker for renal clear cell carcinoma. *J Urol* 2006;**176**:2248-2254.
6. Sartini D, Santarelli A, Rossi V, Goteri G, Rubini C, Ciavarella D, Lo Muzio L, Emanuelli M. Nicotinamide N-methyltransferase upregulation inversely correlates with lymph node metastasis in oral squamous cell carcinoma. *Mol Med* 2007;**13**:415-421.
7. Emanuelli M, Santarelli A, Sartini D, Ciavarella D, Rossi V, Pozzi V, Rubini C, Lo Muzio L. Nicotinamide N-Methyltransferase upregulation correlates with tumour differentiation in oral squamous cell carcinoma. *Histol Histopathol* 2010;**25**:15-20.
8. Sartini D, Pozzi V, Renzi E, Morganti S, Rocchetti R, Rubini C, Santarelli A, Lo Muzio L, Emanuelli M. Analysis of tissue and salivary nicotinamide N-methyltransferase in oral squamous cell carcinoma: basis for the development of a noninvasive diagnostic test for early-stage disease. *Biol Chem* 2012;**393**:505-511.

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9. Ganzetti G, Sartini D, Campanati A, Rubini C, Molinelli E, Brisigotti V, Cecati M, Pozzi V, Campagna R, Offidani A, Emanuelli M. Nicotinamide N-methyltransferase: potential involvement in cutaneous malignant melanoma. *Melanoma Res* 2018;**28**:82-88.
10. Zhou W, Gui M, Zhu M, Long Z, Huang L, Zhou J, He L, Zhong K. Nicotinamide N-methyltransferase is overexpressed in prostate cancer and correlates with prolonged progression-free and overall survival times. *Oncol Lett* 2014;**8**:1175-1180.

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3 **LEGENDS**
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5 **Figure 1.** Immunohistochemical expression of NNMT in tumor tissue sections. a) Keratoacanthoma.
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7 b) Squamous Cell Carcinoma of the trunk and extremities. c) Head and neck Squamous Cell
8 Carcinoma (Immunoperoxidase, original magnification: x200).
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14 **Figure 2.** NNMT immunohistochemical expression in KA, SCC of the trunk and extremities, and
15 head and neck SCC. Values represent the mean percentage \pm standard deviation of stained cells with
16 respect to total counted cells. $p < 0.0001$.
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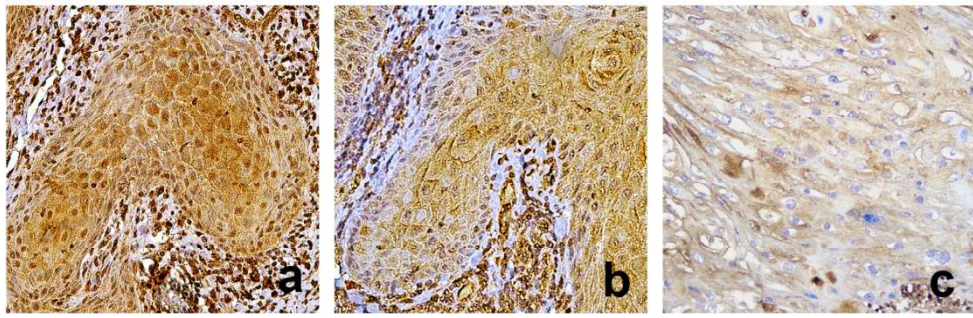


Figure 1. Immunohistochemical expression of NNMT in tumor tissue sections. a) Keratoacanthoma. b) Squamous Cell Carcinoma of the trunk and extremities. c) Head and neck Squamous Cell Carcinoma (Immunoperoxidase, original magnification: x200).

150x51mm (300 x 300 DPI)

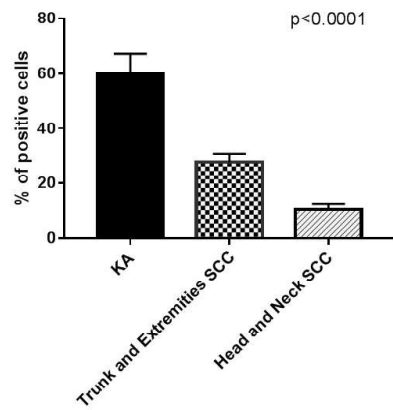


Figure 2. NNMT immunohistochemical expression in KA, SCC of the trunk and extremities, and head and neck SCC. Values represent the mean percentage \pm standard deviation of stained cells with respect to total counted cells. $p < 0.0001$.

338x190mm (96 x 96 DPI)

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 19
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23 POTENTIAL FINANCIAL CONFLICTS

- 24 4. I, my spouse, or one of my dependent children is an employee of a company whose product is being studied. **NO**
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 27 5. I, my spouse, or one of my dependent children has significant equity interest (>USD10,000) in the company that **NO**
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