

Limitations of the Current Equine Melanocytic Tumour Classification

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Introduction

Equine melanocytic tumours (EMT) occur frequently in older grey horses but can be encountered in a horse of any age or coat colour. They are typically black masses occurring on the ventral side of the tail and in the perianal area, but the masses can be variably pigmented, even amelanotic, and can be located anywhere on the body. EMTs can, and often do, progress into generalised disease through metastases. It is estimated that 2-34 % of total neoplasms and 6-15 % of cutaneous neoplasms in horses are EMTs (1).

The process of greying happens with age and is associated with higher incidence of EMTs as well as vitiligo-like depigmentation in horses. Pleiberg et al. revealed that the Grey phenotype is caused by a 4.6-kb duplication in intron 6 of *syntaxin-17* (2). Recent publication by Nowacka-Wozzuk et al. contradicts this finding by proposing that the mutation is triplication rather than duplication based on their results (3).

The currently used histopathological classification scheme was proposed by Valentine in her retrospective study on 53 horses. Based on the results of the study, four types of EMTs were established: melanocytic nevus (MN), dermal melanoma/melanomatosis (DM/DMT) and anaplastic malignant melanoma (AMM). Dermal melanoma and dermal melanomatosis are histologically identical but present clinically as two different syndromes (4). Figure 1 shows typical examples of MN, DM/DMT and AMM. However, recent studies suggest that this classification scheme does not sufficiently reflect the biological behaviour of these tumours (5, 6).

This study aimed to classify EMTs diagnosed by routine HE staining with the aid of immunohistochemistry (IHC).

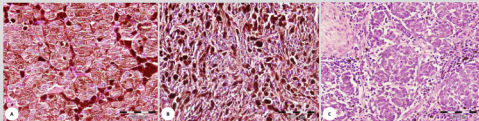


Figure 1: Typical examples of MN, DM/DMT and AMM. (A) Melanocytic nevus, case No. 5, HE, 400x. (B) Dermal melanomatosis, case No. 21, HE, 400x. (C) Anaplastic malignant melanoma, case No. 14, HE, 400x.

Materials and Methods

The samples selected for this study were either biopsies submitted to the Department of Pathological Morphology and Parasitology VETUNI Brno through the Equine Clinic VETUNI Brno or field veterinarians on behalf of the owners or necropsies from horses that were examined post-mortem at our department.

The collected samples were fixed in 10% neutral buffered formalin, paraffin embedded, cut and stained with HE and by immunohistochemistry for PNL 2, S100 protein, vimentin, Ki-67 and RACK1. The highly pigmented lesions were bleached with hydrogen peroxide before staining.

Results

The EMTs from thirty horses were examined, the cases are summarized in the Table 1. Most of the affected horses were grey, only three had other coat colours (black, perline, chestnut) and one horse had unknown coat colour. Based on the criteria specified by Valentine in her study (see Table 2), twelve EMTs were classified as DM/DMT, eleven as MN and two as AMM. Five cases could not be classified. They originated from three horses older than 10 years with a single or multiple tumours which showed characteristics of both MN and DM/DMT (see Figure 2) and two horses in the early stage of the disease.

Acknowledgements

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Results

Case	Breed	Sex	Colour	Age	Location	Diagnosis	Classification (Valentine, 1995)
1	Shank Warmblood	Male	Grey	14	Limb	Skin melanoma	MN
2	Czech Warmblood	Male	Grey	2	Head	Skin melanoma	MN
3	Andalus	Male	Grey	29	Perineum, ventral side of the abdomen, perianal region	Skin melanoma	DMT
4	Warmblood	Male	Grey	17	Ventral side of the tail	Unclassified	Early-stage lesion
5	Coldblood	Gender	Grey	4	Scrotum, thigh, withers, torso, snout, shoulder, neck	Skin melanoma	MN
6	British Coldblood	Male	Grey	16	Perianal area, vulva, upper eyelid	Skin melanoma	DMT
7	Welsh Part-Bred	Male	Unknown	1.0	Base of the tail	Skin melanoma	MN
8	Shank Warmblood	Gender	Grey	21	Ventral side of the tail, perianal region, inside of the abdominal wall, ventral side of the abdomen	Melanoma, malignant melanoma	DMT
9	Andalus	Male	Grey	27	Ventral side of the tail, perianal region, ventral side of the bladder	Skin melanoma	DMT
10	Portuguese Warmblood	Male	Grey	20	Ventral side of the tail, perianal region, vulva, mammary gland	Melanoma, malignant melanoma	DMT
11	English Thoroughbred	Male	Grey	18	Back of the tail	Skin melanoma	DMT
12	Czech Warmblood	Male	Grey	12	Perianal area	Skin melanoma	MN
13	Czech Warmblood	Gender	Grey	14	Anterior side of the eye, forehead	Skin melanoma	MN
14	Friesian	Gender	Black	14	Perineum	Unclassified	AMM
15	English Thoroughbred	Gender	Grey	13	Tail, perianal region, ventral side of the abdomen, pelvic cavity, lower limbs, ventral region	Melanoma, malignant melanoma	DMT
16	English Thoroughbred	Gender	Grey	13	Ventral side of the tail	Skin melanoma	DMT
17	Shagya Arabian	Male	Grey	8	Limb, chest, neck, head	Skin melanoma	MN
18	Holsteiner-mixed	Male	Grey	16	Tail, perianal area, eyelid, at the base of the ear	Skin melanoma	DMT
19	Welsh Cob	Male	Perline	9	Ventral side of the tail	Skin melanoma	MN
20	Kiabauer	Male	Grey	17	Ventral side of the tail, vulva	Skin melanoma	DMT
21	Andalus	Gender	Grey	13	Ventral side of the tail, perianal region, vulva, neck, perianal region	Skin melanoma	DMT
22	Shagya Arabian	Male	Grey	19	Perineum	Unclassified	Early-stage lesion
23	Shank Warmblood	Male	Grey	6	Caudal part of the equine groin	Skin melanoma	MN
24	Shank Sport Pony	Gender	Grey	10	Ventral side of the tail, scrotum, prepuce, test, thigh, abdomen, neck, mandibular region	Skin melanoma	DMT
25	English Thoroughbred	Male	Grey	7	Ventral side of the tail	Skin melanoma	DMT
26	Holsteiner	Male	Grey	11	Ventral side of the tail	Skin melanoma	DMT
27	Holsteiner	Male	Grey	20	Ventral side of the tail, perianal region, perineum, vulva, mammary gland, muscle of the right hindquarter, vulva, perianal region, scapula, LM	Melanoma, malignant melanoma	DMT
28	Oldenburg	Male	Grey	11	Shoulders	Skin melanoma	MN
29	Czech Warmblood	Male	Chestnut	14	Chestnut	Skin melanoma	MN
30	Holsteiner	Gender	Grey	18	Ventral side of the tail, prepuce, spleen	Anaplastic malignant melanoma, early-stage lesion	AMM

Table 1: A brief overview of the cases.

Location in the skin	Melanocytic nevus			Dermal melanoma/melanomatosis			Anaplastic malignant melanoma		
	Superficial masses in dermoepithelial junction, frequent epithelial involvement.	Deep dermal location.		Deep dermal location.			Anaplastic malignant melanoma		
Morphological characteristics of the neoplastic cells	Cluster nests of relatively large, frequently pigmented, pleomorphic pigmented epithelial to spindle-shaped tumor cells with epithelioid nuclei. Occasional binucleate cells. Variable cytoplasmic pigmentation and intracellular bridges.	Small, monotonous, indistinct, round, or spindle-shaped tumor cells with indistinct borders and dense cytoplasmic pigmentation, no mitoses.		Sheets of extremely pleomorphic epithelioid cells, with irregular cell borders, nuclear pleomorphism, and numerous mitoses.			Sheets of extremely pleomorphic epithelioid cells, with irregular cell borders, nuclear pleomorphism, and numerous mitoses.		
Characteristics of the tumours	Generally solitary, circumscribed, superficial lesions.	DM: one or two discrete masses; DM/DMT: frequently confluent lesions.		DM: one or two discrete masses; DM/DMT: frequently confluent lesions.			DM: one or two discrete masses; DM/DMT: frequently confluent lesions.		
Tumour location on the body	Most often occur on the neck, trunk and limbs but can occur in more typical locations.	Most frequently involve the ventral tail, perianal region, perineum, mammary glands, vulva, scrotum, perineum or perianal gland regions. High metastatic rate for DM/DMT.		Most frequently involve the ventral tail, perianal region, perineum, mammary glands, vulva, scrotum, perineum or perianal gland regions. High metastatic rate for DM/DMT.			Anywhere on the body, locally aggressive, early metastases.		
Age and coat color	Young horses of all coat colors.	DM: generally mature, but not aged, grey horses; DM/DMT: aged grey horses, usually over 10 years of age.		DM: generally mature, but not aged, grey horses; DM/DMT: aged grey horses, usually over 10 years of age.			Older, predominantly non-grey horses.		

Table 2: Histopathological classification criteria (Valentine, 1995).

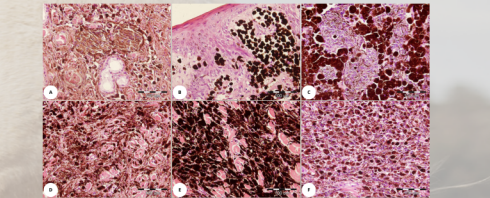


Figure 2: The unclassified cases. (A, B, C) Characteristics typical of MN. (D, E, F) Characteristics typical for DM/DMT. (A, D) Case No. 13, HE, 400x; (B, E) Case No. 18, HE, 400x; (C, F) Case No. 24, HE, 400x.

Conclusions

Nine cases (30 % of the total cases) showed characteristics typical for both MN and DM/DMT but in six cases the characteristics for one type were significantly predominant. Remaining three cases (10 % of the total cases) showed the characteristics for both MN and DM/DMT to similar extent and therefore could not be classified. In our opinion, these findings may suggest the presence of a histological continuum between the two types. Our results indicate the need for improving/extending the current classification.