Protoplasma
DOI 10.1007/s00709-016-1038-z

REVIEW ARTICLE

Intercellular crosstalk in human malignant melanoma

Barbora Dvořánková [1,2] · Pavol Szabo [1,2] · Ondřej Kodet [1,2,3] · Hynek Strnad [4] · Michal Kolář [4] · Lukáš Lacina [1,3] · Eliška Krejčí [1] · Ondřej Naňka [1] · Aleksi Šedo [5] · Karel Smetana Jr. [1,2]

Received: 5 September 2016 / Accepted: 19 October 2016

Abstract Incidence of malignant melanoma is increasing globally. While the initial stages of tumors can be easily treated by a simple surgery, the therapy of advanced stages is rather limited. Melanoma cells spread rapidly through the body of a patient to form multiple metastases. Consequently, the survival rate is poor. Therefore, emphasis in melanoma research is given on early diagnosis and development of novel and more potent therapeutic options. The malignant melanoma is arising from melanocytes, cells protecting mitotically active keratinocytes against damage caused by UV light irradiation. The melanocytes originate in the neural crest and consequently migrate to the epidermis. The relationship between the melanoma cells, the melanocytes, and neural crest stem cells manifests when the melanoma cells are implanted to an early embryo: they use similar migratory routes as the normal neural crest cells. Moreover, malignant potential of these melanoma cells is overdriven in this experimental model, probably due to microenvironmental reprogramming. This observation demonstrates the crucial role of the microenvironment in melanoma biology. Indeed, malignant tumors in general represent complex ecosystems, where multiple cell types influence the growth of genetically mutated cancer cells. This concept is directly applicable to the malignant melanoma. Our review article focuses on possible strategies to modify the intercellular crosstalk in melanoma that can be employed for therapeutic purposes.

Keywords Melanocyte · Melanoma cells · Melanoma ecosystem · Cancer-associated fibroblast · Keratinocyte · Cytokine

Handling Editor:Pavel Draber

✉ Karel Smetana, Jr.
karel.smetana@lf1.cuni.cz

[1] Institute of Anatomy, Charles University, 1st Faculty of Medicine, U Nemocnice 3, 128 00 Prague, Czech Republic

[2] BIOCEV, Průmyslová 595, 252 50 Vestec, Czech Republic

[3] Department of Dermatology and Venerology, Charles University, 1st Faculty of Medicine and General University Hospital in Prague, U Nemocnice 2, 128 08 Prague, Czech Republic

[4] Institute of Molecular Genetics, Academy of Sciences of the Czech Republic, Vídeňská 1083, 142 20 Prague, Czech Republic

[5] Institute of Biochemistry and Experimental Oncology, Charles University, 1st Faculty of Medicine, U Nemocnice 5, 128 53 Prague, Czech Republic

Increase of melanoma incidence

The incidence of malignant melanoma is growing worldwide. This phenomenon can be exemplified on the data from the Czech Republic, where the melanoma incidence increased almost four times over the last 35 years (Global Burden of Disease Cancer Collaboration et al. 2015; ÚZIS 2011). Fortunately, the melanoma-related mortality is not increasing so rapidly as efficient screening programs and public awareness allow dermatologists to identify and treat early stages of tumors (Higgins et al. 2015). The explanation of this global trend includes, but is not limited to, changes of the climate and more extensive UV light exposition (Nishisgori 2015). The initial stages of melanoma are quite easily curable by surgical resection. The advanced stages are traditionally treated by chemotherapy and/or immunotherapy. Several novel therapies for melanoma emerged over the last decade. These remarkable drugs allow the oncologist to target specifically mutated melanoma cells, block important signaling pathways in them or unblock immune checkpoints, and trigger

Published online: 03 November 2016

immune attack against them. However, the combination of several drugs is necessary for disease management in some cases. Unfortunately, efficiency of this combined therapy is still limited, accompanied by numerous complications, and it also represents financial burden on healthcare systems even in developed countries (Harries et al. 2016). In this light, search for more efficient therapeutic modalities is still essential.

Melanocytes, their function, and origin

In the basic concept of epidermal biology, normal melanocytes are located in the basal layer of the epidermis. The epidermal melanocytes produce pigment called melanin. Melanin packages, melanosomes, are exocytosed and immediately internalized by keratinocytes. Once ingested by keratinocyte, melanosomes protect the keratinocyte nucleus against UV light-induced DNA damage (Colombo et al. 2011). Next to this, melanocytes can be present also in the dermis and other extracutaneous tissues as uvea and many other atypical organs or tissues. Melanocytes in these locations can be relevant for the process of melanoma formation in less typical sites.

Developmentally, epidermal keratinocytes originate from the surface embryonic ectoderm and melanocytes develop from the neural crest. The neural crest is formed from the cellular layer connecting the neural plate with the surface ectoderm (the prospective epidermis). During the process of the neural tube formation, the neural folds elevate and fuse and cells at the lateral border of the neuroectoderm begin to dissociate from their neighbors. This population, the neural crest, will undergo and epithelial-to mesenchymal transition as it leaves the neuroectoderm by active migration and displacement to enter the underlying mesoderm. The melanocytes migrate by dorsal pathway through the dermis, where they will enter the ectoderm through holes in basal lamina to form melanocytes in the skin and hair follicles (Gilbert 2000; Hall 2008). The designation of the neural crest cells as precursors for distinct cell types is under precise genetic as well as epigenetic control (Zhang et al. 2014).

Hair follicle as a cradle for the stem cells

Hair follicle represents a unique human organ that goes postnatally through multiple cycles of repeated morphogenesis (Lee and Tumbar 2012). It is able to undergo cyclic regeneration and regression. A very important role in this process is assigned to a population of epidermal stem cells (Lavker et al. 2003) and their crosstalk with mesenchymal dermal papilla cells. The epidermal stem cells seem to be, at least, tripotent. They can give rise to cells of the hair follicle, epidermal keratinocytes, and cells of sebaceous glands. However, the bulge region also contains highly multipotent stem cells of the neural crest origin (Sieber-Blum and Grim 2004; Sieber-Blum et al. 2004). These cells, also called melanocyte stem cells, are highly multipotent, and they can in vitro differentiate to the same lineages as the neural crest cells (Sieber-Blum and Grim 2004; Sieber-Blum et al. 2004). They are the most superficially located and hence easily accessible multipotent stem cells in the human body (Krejčí and Grim 2010). This makes them suitable, e.g., for tissue engineering (Sieber-Blum et al. 2006). This coexistence of two distinct pools of highly potent stem cells in a limited compartment is physiologically unique and raises questions about their possible collaborative interactions. Transcription factor NFIB was proposed as an important molecule orchestrating the interaction between the two stem cell pools (Chang et al. 2013). This fact can be highly inspirative for further research on employment of these stem cell types in regenerative medicine.

Grafting of melanoma cells to embryo

As depicted in seminal transplantation experiments in 1970s, cells of teratocarcinoma with the normal number of chromosomes injected to a mouse blastocyst are able to participate in formation of normal tissues. The resulting animals are a mosaic of normal cells and the cells originating from the teratocarcinoma (Mintz and Illmensee 1975). Based on this concept, Pierce postulated idea that embryonic microenvironment can determine cancer cell properties (Pierce 1983). This conclusion seems to be relevant also in melanoma, as the melanoma cells injected to an embryo migrate by the same routes as the neural crest cells (Kulesa et al. 2006; Hendrix et al. 2007; Kasemeier-Kulesa et al. 2008; (Bailey et al. 2012; Díez-Torre et al. 2009); Kulesa et al. 2013) (Fig. 1). This migration in the embryonic environment is accompanied by significant reduction of their malignant potential (Lee et al. 2005). However, it is necessary to note that the timescale of these animal models is significantly shorter than the human lifespan. These findings are further supported by the observation that the melanoma initiating (stem) cells exhibit CD271, a receptor for a growth factor important for the neural crest cells (Boiko et al. 2010). Human embryonic stem cells also seem to reduce malignant properties of the melanoma cells by production of gremlin (Kim et al. 2011). These observations indicate uniformly that the embryonic microenvironment can influence behavior of cancer cells (Abbott et al. 2008). This fact might be relevant for the identification of novel therapeutic targets in melanoma. The neural crest origin of the melanocytes also suggests explanation of the enormous invasivity of the cells from advanced melanomas because the neural crest cells migrate to practically all tissues of the body.

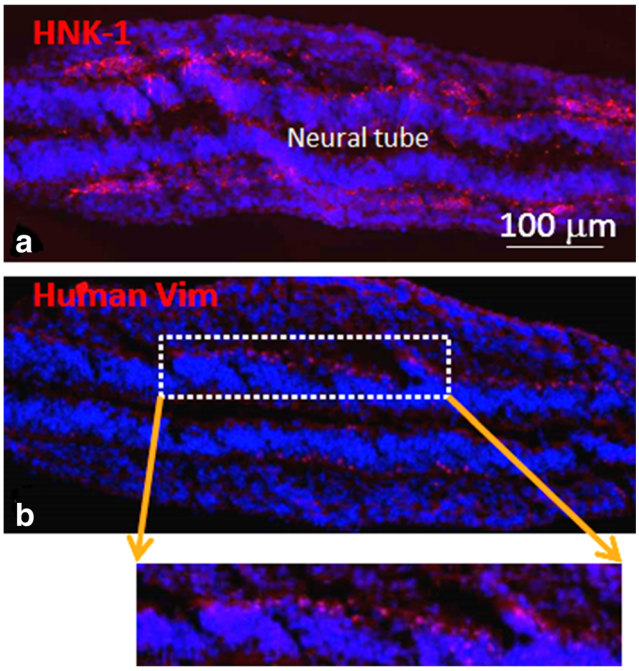

Fig. 1 **a** HNK-1-positive cells of the neural crest are located on both sides of the neural tube of a chicken embryo. **b** Human melanoma cells of the BLM cell line injected to the neural tube region, which exhibits human vimentin, are present in the same localization as the neural crest cells. Nuclei are counterstained with DAPI. Frozen longitudinal section through chicken embryo parallel with dorsal surface. *Bar* is 100 μm

Al least in part, cells in malignant melanoma also exhibit features of stemness; they specifically resemble in certain aspects neural crest derived stem cells. This may substantiate reasons for frequent melanoma treatment failures or recurrent disease, because it is exceedingly difficult to completely eradicate these cells (El-Khattouti et al. 2014, 2015). However, successful targeting of the principal pathways responsible for stemness maintenance in melanoma cells would represent an important novel strategy of this tumor treatment (Alamodi et al. 2016).

The cancer microenvironment—lesson from the cancer ecology

Last decades brought dramatic increase of available data about adult tissue stem cells. However, the data focused predominantly on their potential in regenerative medicine and their practical utilization is still far behind general expectation. However, better understanding to normal cell differentiation brought benefits in conjunction with the cancer stem cell theory (Hamburger and Salmon 1977). By definition, the cancer stem cells are in many aspects similar to the normal stem cells. This similarity has direct implications for tumor therapy, as it explains the resistance of the cancer stem cells to xenobiotics including chemotherapy. Next to that, regenerative potential of the normal cells has a good parallel in their cancerous counterparts with consequences for the residual disease (Motlík et al. 2007; Carnero et al. 2016). Despite sharing important features, the cancer stem cells do not necessarily originate from the tissue stem cells. They can arise by the process of cell dedifferentiation, which could be initiated by the accumulation of multiple genetic mutations acquired in course of cancer development (Woodward and Hill 2016).

A specific tissue microenvironment is critically important for stemness maintenance of the normal tissue stem cells as it seems to control the activity of the stem cells (Choi et al. 2015, 2015). This specific microenvironment is traditionally called the niche or the stroma. Surprisingly, our definition of and understanding to this crucial factor are still rather poor. The most substantial data are available for the hematopoietic stem cell niche in the bone marrow (Birbrair and Frenette 2016). However, this is not the most relevant model for biology of solid tumors. Hair follicle as a joint niche for both the epidermal stem cells and the neural crest-originated stem cells may be a more relevant model for the malignant melanoma. Next to the cells present in the niche, we have to acknowledge also the role of their products such as extracellular matrix, cytokines, growth factors, and extracellularly released enzymes. All these components must be considered as indispensable components of this specific microenvironment (Lane et al. 2014; Choi et al. 2015). Indeed, malignant cells do not form tumors by themselves: cancer-associated fibroblasts (CAFs), infiltrating immune cells, and blood/lymphatic endothelial cells as well as their extracellular products are participating together in the process of cancer formation and progression (Lacina et al. 2015). Mutual and multilateral interactions of individual cell types are extremely complex and resemble natural ecosystems (Kareva 2011).

Great similarity between granulation tissue of the wound and the cancer stroma was highlighted many times (Dvorak 1986; Smetana et al. 2015). In both situations, characteristic presence of highly specialized mesenchymal cells expressing smooth muscle actin, myofibroblasts, was noted (Krejčí et al. 2016). In wound, these cells participate in the wound contraction and thus minimize the area which should be reepithelized. The myofibroblasts are most likely formed from the local mesenchyme (Jarkovska et al. 2014; Dvořánková et al. 2015), probably due to increased activity of TGF-β and an endogenous lectin galectin-1 (Dvořánková et al. 2011; Valach et al. 2012). Other sources of myofibroblasts were also proposed (Lacina et al. 2015). In tumor, CAFs also express smooth muscle actin, which makes them similar to the myofibroblasts (Lacina et al. 2015; Smetana et al. 2015). Their contractile ability is not emphasized but they are highly bioactive. They are able to modulate the biological properties of many types of tumors by production of multiple growth factors, chemokines, and cytokines (Lacina et al. 2015). Moreover, CAFs from epidermal carcinomas are able to influence the phenotype of normal human keratinocytes to be similar to the cancer cells or the epidermal stem cells. CAFs can also induce epithelial-to-

mesenchymal transition (Strnad et al. 2010; Kolář et al. 2012). This phenomenon is somewhat similar to the induction of hair follicles in glabrous skin by transplantation of dermal papilla mesenchymal cells (Jahoda et al. 1984). Moreover, normal human neonatal fibroblasts isolated from the wound exert activity comparable to CAFs in in vitro models (Mateu et al. 2016). This underscores the striking similarity between wound healing and cancer as noted by Dvorak in 1986: Tumors–wounds that do not heal.

Factors such as IL-6, IL-8, CXCL-1, BMP-4, IGF-2, FGF-7, and TGF-β3 were discovered to participate in the crosstalk between CAFs and the cancer cells (Lacina et al. 2015; Smetana et al. 2015; Krejčí et al. 2016). The remarkable number of these growth factors, chemokines, and cytokines is involved in immune reaction and thus they can participate in formation of inflammation and support the micromillieu that stimulates oncogenesis.

CAFs from basal cell carcinoma drive normal fibroblasts to acquire properties of the mesenchymal stem cells including their differentiation plasticity (Szabo et al. 2011). This observation suggests that the recruitment of the mesenchymal cells to the cancer stroma can be both of the frontal type (the cancer cells signal to the surrounding mesenchyme), but can also propagate further in the lateral direction (the mesenchyme signals to the mesenchyme).

Intercellular interactions in malignant melanoma

The function of the melanocytes in the epidermis is dependent on mutual contacts with keratinocytes. The number of these intercellular contacts is significantly reduced during the melanocyte malignization (Haass and Herlyn 2005). A switch of expression of E-cadherin to N-cadherin as well as significant reduction of connexin 43 expression was observed in the keratinocytes surrounding the malignant cells. This is a strong evidence of alterations in intercellular crosstalk (Haass et al 2004). Advanced nodular melanoma is overlayered by the epidermis that exhibits, peripherally from the tumor, signs of hyperplasia, paradoxically without accumulation of proliferating cells (McCarty et al. 2003; (Drunkenmölle et al. 2005; Kodet et al. 2015). Moreover, this epidermis also suprabasally and aberrantly expresses a marker of proliferating basal cells, keratin 14, even in the distance of 15 mm from the tumor margin. The mutual interaction between the neighboring tissues, i.e., the melanoma and the epidermis, is also supported by the presence of the pluripotency marker Nanog in their nuclei (Fig. 2). Normal human keratinocytes cocultured with the melanoma cells express markers of low differentiation status such as keratins 8, 14, and 19. Majority of the epithelial cells paradoxically express mesenchymal vimentin. Its expression can be interpreted as a marker of epithelial-to-mesenchymal transition. Unlike normal human melanocytes,

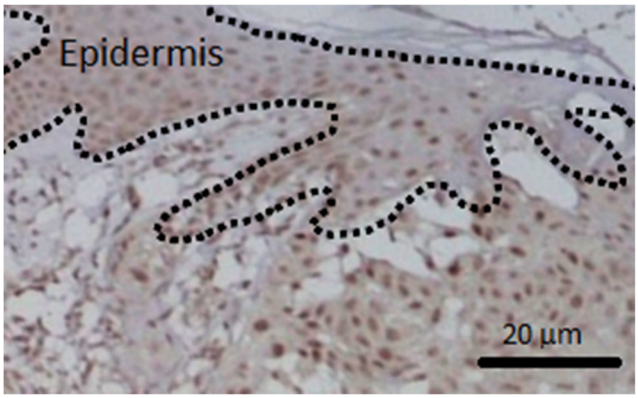

Fig. 2 Nuclei of epidermal keratinocytes in the vicinity of nodular melanoma as well as nuclei of cells of malignant melanoma are positive for the presence of the pluripotency marker Nanog. *Bar* is 20 µm

the neural crest-originated stem cells isolated from the hair follicle exhibit the same activity (Kodet et al. 2015).

Production of FGF-2, CXCL-1, IL-8, and VEGF-A by the melanoma/neural crest-originated stem cells seem to be responsible for the control of the phenotype of the cocultured keratinocytes (Kodet et al. 2015). It is necessary to emphasize here that these results harmonize with the abovementioned hypothesis on collaboration between both stem cell types colocalized in the bulge region of the hair follicle (Chang et al. 2013).

The reciprocal influence of keratinocytes on the behavior of the melanoma cells is continuously discussed. Keratinocytes control adhesion and migration of the melanoma cells on laminin in vitro (Chung et al. 2011). This process may affect melanoma metastasation. The role of keratinocytes seems to be context dependent and plays an important role in melanoma invasion in a reconstructed skin model (Van Kilsdonk et al 2010).

As mentioned in the previous chapter, fibroblasts and namely CAFs significantly influence biological properties of various types of tumors. Conversely, platelet derived growth factor CC (PDGF-CC), which is produced in melanoma, stimulates recruitment of fibroblasts to the tumor and drives their conversion into CAFs (Anderberg et al. 2009). CAFs located in melanoma (M-CAFs) frequently express podoplanin, similarly to other types of tumors. Its expression correlates with aggressive behavior of the tumor cells (Kan et al. 2014). In models where M-CAFs and melanoma cells are in a direct contact, their crosstalk seems to be dependent on the Notch1 signaling pathway (Shao et al. 2015). Melanoma cells of advanced tumors growing in effusion fluid in pleural cavity, where there are virtually no interacting fibroblasts, usually lack their typical phenotype, which is otherwise shared by cells of the primary tumor and organ metastases. Both normal dermal fibroblasts as well as M-CAFs significantly influence phenotype of ascitic melanoma cells in coculture, where the melanoma cells acquire phenotypic

properties of their primary tumors. This fibroblast activity is further strengthened by conditioned medium from embryonic stem cells (Kodet et al. 2013). Inflammatory cytokines and chemokines, such as IL-6 and IL-8, also participate in the CAFs crosstalk with the cancer cells in a paracrine manner even without direct cellular contact (Kolář et al. 2012). These factors are elevated in the sera of patients suffering from advanced melanoma (Kucera et al. 2014; Sanmamed et al. 2014). Conditioned media from M-CAFs and from M-CAFs cocultured with the melanoma cells have no effect on the proliferation of the melanoma cells but they significantly stimulate invasivity of the melanoma cells in collagen gel (Jobe et al. 2016). Application of antibodies blocking the activity of IL-6 and IL-8 fully inhibits the migration of the melanoma cells in the collagen gel (Jobe et al. 2016). M-CAFs are also able to influence phenotype of normal human keratinocytes where they induce expression of a marker of proliferating basal layer keratinocytes, keratin type 14. Moreover, the keratinocytes stimulated by M-CAFs express vimentin, a marker of epithelial-to-mesenchymal transition (Kučera et al. 2015). These observations indicate that the keratinocytes in the vicinity of the malignant melanoma are under the influence of the melanoma cells and that M-CAFs and these keratinocytes are able to affect the migratory activity of the melanoma cells.

An interesting question arises, whether the activity of CAFs is tumor type specific. M-CAFs are not able to stimulate proliferation of glioblastoma cells more extensively than normal dermal fibroblasts. However, M-CAFs significantly stimulate glioblastoma invasivity (Trylcova et al. 2015). Cells similar to CAFs were also discovered in samples of human glioblastoma (Clavreul et al. 2014; Trylcova et al. 2015; Busek et al. 2016), which is interesting with respect to the glioblastoma histogenesis. The CAFs of glioblastoma probably originate from the mesenchymal stem cells that actively migrate to the glioblastoma. This phenomenon seems to harbor certain therapeutic potential as they can also bring some therapeutic cargo to the tumor (Pacioni et al. 2015). Next to glioblastoma, M-CAFs are able to significantly influence phenotype of breast cancer cells (Dvořánková et al. 2012). Observation that activity of M-CAFs is not tumor type specific can be important. This suggests a more general mechanism of their bioactivity upon different types of tumors. Therefore, targeting of this mechanism could have wider application in different types of tumors.

Dealing with resistance to Vemurafenib treatment by melanoma may be a prominent example. Vemurafenib is a powerful therapeutic agent for the treatment of the melanomas harboring B-Raf V600E mutation. Unfortunately, the therapy must be frequently terminated due to acquired resistance to Vemurafenib with consequent rapid disease progression. It is likely that this therapeutic resistance is driven by the stroma with high CAFs activation (Whipple and Brinckerhoff 2014). Therapeutical suppression of CAFs activity can thus help to delay or completely abrogate the acquired resistance to B-Raf inhibitor therapy. Synthetic polyamines seem to be a promising option in this context (Mifková et al. 2014).

Stromal immune reaction is evident in many tumors and displays a multifaceted interaction of immune system and tumor. Infiltration of malignant melanoma by leukocytes is a frequent phenomenon with complicated interpretation. Interestingly, the prognosis of a patient depends on the predominant type of leukocytes. For example, high peritumoral incidence of T and B lymphocytes and dendritic cells represents a good prognostic marker. On the other hand, predominance of granulocytes suggests poor survival prognosis (Ladányi 2015). Myeloid-derived suppressor cells present in melanoma have an immunosuppressive effect and influence negatively survival of melanoma patients. Application of Ipilimumab, CTLA4 targeted antibody, significantly improves survival of some patients and increases their quality of life (Umansky et al. 2016). The PD-1 (programmed cell death—1) receptor is expressed on the surface of activated T cells. Its ligands, PD-L1/PD-L2 belong to the family of immune checkpoint proteins that act as co-inhibitory factors, which can halt or limit the development of the T cell response. The granulocyte macrophage colony stimulating factor (GM-CSF) application to melanoma patients also seems to have a promising therapeutic effect in some patients, but the results are quite heterogeneous (Hoeller et al. 2016).

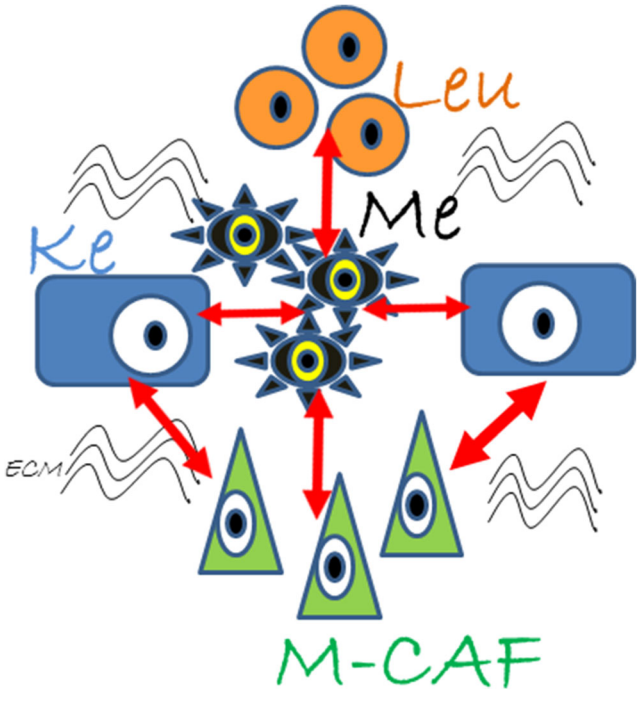

Fig. 3 Proposed model of the malignant melanoma ecosystem with marked interaction between the melanoma cells (Me), melanoma cancer-associated fibroblasts (M-CAF), keratinocytes (Ke), and infiltrating leukocytes (Leu). Extracellular matrix (ECM)

Conclusion

Similarly to other types of tumors, human malignant melanoma represents a complicated ecosystem, where keratinocytes, M-CAFs, and infiltrating leukocytes communicate with the melanoma cells (Fig. 3). This interplay is able to substantially influence the biological properties of the tumor, including its metastatic potential. Data from grafting of the melanoma cells to the early embryo indicate certain ability of the environment to attenuate their malignant potential. Targeted manipulation of the melanoma microenvironment thus seems to be a rising therapeutic perspective promising, for example, reversion of acquired therapeutic resistance to Vemurafenib in BRAF mutated tumors.

Acknowledgments This publication is a result of the project implementation: "The equipment for metabolomic and cell analyses," registration number CZ.1.05/2.1.00/19.0400, supported by Research and Development for Innovations Operational Program (RDIOP) co-financed by European regional development fund and the state budget of the Czech Republic. This study was also supported by the Grant Agency of the Czech Republic (project no. 16-05534S), Charles University (project of Specific University Research and PRVOUK-27 and UNCE 204013), Ministry of Education, Youth and Sports of CR within the National Sustainability Program II (project BIOCEV-FAR reg. no. LQ1604), and project BIOCEV (CZ.1.05/1.1.00/02.0109). Access to computing and storage facilities owned by parties and projects contributing to the National Grid Infrastructure MetaCentrum provided under the program "Projects of Projects of Large Research, Development, and Innovations Infrastructures" (CESNET LM2015042) is greatly appreciated.

Compliance of ethical standards

Conflict of interest The authors declare that they have no competing interests.

References

Abbott DE, Bailey CM, Postovit LM, Seftor EA, Margaryan N, Seftor RE, Hendrix MJ (2008) The epigenetic influence of tumor and embryonic microenvironments: how different are they? Cancer Microenviron 1:13–21. doi:10.1007/s12307-008-0004-5

Alamodi AA, Eshaq AM, Hassan SY, Al Hmada Y, El Jamal SM, Fothan AM, Arain OM, Hassan SL, Haikel Y, Megahed M, Hassan M (2016) Cancer stem cell as therapeutic target for melanoma treatment. Histol Histopathol 31(12):1291–1301. doi:10.14670/HH-11-791

Anderberg C, Li H, Fredriksson L, Andrae J, Betsholtz C, Li X, Eriksson U, Pietras K (2009) Paracrine signaling by platelet-derived growth factor-CC promotes tumor growth by recruitment of cancer-associated fibroblasts. Cancer Res 69:369–378. doi:10.1158/0008-5472.CAN-08-2724

Bailey CM, Morrison JA, Kulesa PM (2012) Melanoma revives an embryonic migration program to promote plasticity and invasion. Pigment Cell Melanoma Res 25:573–583. doi:10.1111/j.1755-148X.2012.01025.x

Birbrair A, Frenette PS (2016) Niche heterogeneity in the bone marrow. Ann N Y Acad Sci 1370:82–96. doi:10.1111/nyas.13016

Boiko AD, Razorenova OV, van de Rijn M, Swetter SM, Johnson DL, Ly DP, Butler PD, Yang GP, Joshua B, Kaplan MJ, Longaker MT, Weissman IL (2010) Human melanoma-initiating cells express neural crest nerve growth factor receptor CD271. Nature 466:133–137. doi:10.1038/nature09161

Busek P, Balaziova E, Matrasova I, Hilser M, Tomas R, Syrucek M, Zemanova Z, Krepela E, Belacek J, Sedo A (2016) Fibroblast activation protein alpha is expressed by transformed and stromal cells and is associated withmesenchymal features in glioblastoma. Tumour Biol. doi:10.1007/s13277-016-5274-9

Carnero A, Garcia-Mayea Y, Mir C, Lorente J, Rubio IT, LLeonart ME (2016) The cancer stem-cell signaling network and resistance to therapy. Cancer Treat Rev 49:25–36. doi:10.1016/j.ctrv.2016.07.001

Chang CY, Pasolli HA, Giannopoulou EG, Guasch G, Gronostajski RM, Elemento O, Fuchs E (2013) NFIB is a governor of epithelial-melanocyte stem cell behaviour in a shared niche. Nature 495:98–102. doi:10.1038/nature11847

Choi HR, Byun SY, Kwon SH, Park KC (2015) Niche interactions in epidermal stem cells. World J Stem Cells 26:495–501. doi:10.4252/wjsc.v7.i2.495

Chung H, Suh EK, Han IO, Oh ES (2011) Keratinocyte-derived laminin-332 promotes adhesion and migration in melanocytes and melanoma. J Biol Chem 286:13438–13447. doi:10.1074/jbc.M110.166751

Clavreul A, Guette C, Faguer R, Tétaud C, Boissard A, Lemaire L, Rousseau A, Avril T, Henry C, Coqueret O, Menei P (2014) Glioblastoma-associated stromal cells (GASCs) from histologically normal surgical margins have a myofibroblast phenotype and angiogenic properties. J Pathol 233:74–88. doi:10.1002/path.4332

Colombo S, Berlin I, Delmas V, Larue L (2011) Classical and nonclassical melanocytes in vertebrates. In: Borovanský J, Riley PA (eds) Melanins and melanosomes. Biosynthesis, biogenesis, physiological and pathological functions. Wiley-Blackwell, Weinhem, pp 21–61

Díez-Torre A, Andrade R, Eguizábal C, López E, Arluzea J, Silió M, Aréchaga J (2009) Reprogramming of melanoma cells by embryonic microenvironments. Int J Dev Biol 53:1563–1568. doi:10.1387/ijdb.093021ad

Drunkenmölle E, Marsch WC, Lübbe D, Helmbold P (2005) Paratumoral epidermal hyperplasia: a novel prognostic factor in thick primary melanoma of the skin? Am J Dermatopathol 27:482–488

Dvorak HF (1986) Tumors: wounds that do not heal. Similarities between tumor stroma generation and wound healing. N Engl J Med 315:1650–1659

Dvořánková B, Szabo P, Lacina L, Gal P, Uhrova J, Zima T, Kaltner H, André S, Gabius HJ, Sykova E, Smetana K Jr (2011) Human galectins induce conversion of dermal fibroblasts into myofibroblasts and production of extracellular matrix: potential application in tissue engineering and wound repair. Cells Tissues Organs 194:469–480. doi:10.1159/000324864

Dvořánková B, Szabo P, Lacina L, Kodet O, Matoušková E, Smetana K Jr (2012) Fibroblasts prepared from different types of malignant tumors stimulate expression of luminal marker keratin 8 in the EM-G3 breast cancer cell line. Histochem Cell Biol 137:679–685. doi:10.1007/s00418-012-0918-3

Dvořánková B, Smetana K Jr, Říhová B, Kučera J, Mateu R, Szabo P (2015) Cancer-associated fibroblasts are not formed from cancer cells by epithelial-to-mesenchymal transition in nu/nu mice. Histochem Cell Biol 143:463–469. doi:10.1007/s00418-014-1293-z

El-Khattouti A, Selimovic D, Haïkel Y, Megahed M, Gomez CR, Hassan M (2014) Identification and analysis of CD133(+) melanoma stem-like cells conferring resistance to taxol: an insight into the mechanisms of their resistance and response. Cancer Lett 343:123–133. doi:10.1016/j.canlet.2013.09.024

El-Khattouti A, Sheehan NT, Monico J, Drummond HA, Haikel Y, Brodell RT, Megahed M, Hassan M (2015) $CD133^{+}$ melanoma subpopulation acquired resistance to caffeic acid phenethyl ester-

induced apoptosis is attributed to the elevated expression of ABCB5: significance for melanoma treatment. Cancer Lett 357: 83–104. doi:10.1016/j.canlet.2014.10.043

Gilbert SF (2000) The central nervous system and the epidermis. Developmental Biology. Sinauer Associates, Sunderland, pp 379–410

Global Burden of Disease Cancer Collaboration et al (2015) The global burden of cancer 2013. JAMA Oncol 1:505–527. doi:10.1001/jamaoncol.2015.0735

Haass NK, Herlyn M (2005) Normal human melanocyte homeostasis as a paradigm for understanding melanoma. J Investig Dermatol Symp Proc 10:153–163

Haass NK, Smalley KS, Herlyn M (2004) The role of altered cell-cell communication in melanoma progression. J Mol Histol 35:309–318

Hall BK (2008) The neural crest and neural crest cells: discovery and significance for theories of embryonic organization. J Biosci 33: 781–793

Hamburger AW, Salmon SE (1977) Primary bioassay of human tumor stem cells. Science 197:461–463

Harries M, Malvehy J, Lebbe C, Heron L, Amelio J, Szabo Z, Schadendorf D (2016) Treatment patterns of advanced malignant melanoma (stage III-IV)—a review of current standards in Europe. Eur J Cancer 60:179–189. doi:10.1016/j.ejca.2016.01.011

Hendrix MJ, Seftor EA, Seftor RE, Kasemeier-Kulesa J, Kulesa PM, Postovit LM (2007) Reprogramming metastatic tumour cells with embryonic microenvironments. Nat Rev Cancer 7:246–255

Higgins HW 2nd, Lee KC, Galan A, Leffell DJ (2015) Melanoma in situ: Part I. Epidemiology, screening, and clinical features. J Am Acad Dermatol 73:181–190. doi:10.1016/j.jaad.2015.04.014

Hoeller C, Michielin O, Ascierto PA, Szabo Z, Blank CU (2016) Systematic review of the use of granulocyte-macrophage colony-stimulating factor in patients with advanced melanoma. Cancer Immunol Immunother 65:1015–1034. doi:10.1007/s00262-016-1860-3

Jahoda CA, Horne KA, Oliver RF (1984) Induction of hair growth by implantation of cultured dermal papilla cells. Nature 311:560–562

Jarkovska K, Dvorankova B, Halada P, Kodet O, Szabo P, Gadher SJ, Motlik J, Kovarova H, Smetana K Jr (2014) Revelation of fibroblast protein commonalities and differences and their possible roles in wound healing and tumourigenesis using co-culture models of cells. Biol Cell 106(7):203–218. doi:10.1111/boc.201400014

Jobe NP, Rösel D, Dvořánková B, Kodet O, Lacina L, Mateu R, Smetana K Jr, Brábek J (2016) Simultaneous blocking of IL-6 and IL-8 is sufficient to fully inhibit CAF-induced human melanoma cell invasiveness. Histochem Cell Biol 146:205–217. doi:10.1007/s00418-016-1433-8

Kan S, Konishi E, Arita T, Ikemoto C, Takenaka H, Yanagisawa A, Katoh N, Asai J (2014) Podoplanin expression in cancer-associated fibroblasts predicts aggressive behavior inmelanoma. J Cutan Patho 41: 561–567. doi:10.1111/cup.12322

Kareva I (2011) What can ecology teach us about cancer? Transl Oncol 4: 266–270

Kasemeier-Kulesa JC, Teddy JM, Postovit LM, Seftor EA, Seftor RE, Hendrix MJ, Kulesa PM (2008) Reprogramming multipotent tumor cells with the embryonic neural crest microenvironment. Dev Dyn 237:2657–2566. doi:10.1002/dvdy.21613

Kim MO, Kim SH, Oi N, Lee MH, Yu DH, Kim DJ, Cho EJ, Bode AM, Cho YY, Bowden TG, Dong Z (2011) Embryonic stem-cell-preconditioned microenvironment induces loss of cancer cell properties in human melanoma cells. Pigment Cell Melanoma Res 24: 922–931. doi:10.1111/j.1755-148X.2011.00891.x

Kodet O, Dvořánková B, Krejčí E, Szabo P, Dvořák P, Štork J, Krajsová I, Dundr P, Smetana K Jr, Lacina L (2013) Cultivation-dependent plasticity of melanoma phenotype. Tumour Biol 34:3345–3355. doi:10.1007/s13277-013-0905-x

Kodet O, Lacina L, Krejčí E, Dvořánková B, Grim M, Štork J, Kodetová D, Vlček Č, Šáchová J, Kolář M, Strnad H, Smetana K (2015) Melanoma cells influence the differentiation pattern of human epidermal keratinocytes. Mol Cancer 14:1. doi:10.1186/1476-4598-14-1

Kolář M, Szabo P, Dvořánková B, Lacina L, Gabius HJ, Strnad H, Sáchová J, Vlček C, Plzák J, Chovanec M, Cada Z, Betka J, Fík Z, Pačes J, Kovářová H, Motlík J, Jarkovská K, Smetana K Jr (2012) Upregulation of IL-6, IL-8 and CXCL-1 production in dermal fibroblasts by normal/malignant epithelial cells in vitro: immunohistochemical and transcriptomic analyses. Biol Cell 104:738–751. doi:10.1111/boc.201200018

Krejčí E, Grim M (2010) Isolation and characterization of neural crest stem cells from adult human hair follicles. Folia Biol 56:149–157

Krejčí E, Dvořánková B, Szabo P, Naňka O, Strnad H, Kode O, Lacina L, Kolář M, Smetana K Jr (2016) Fibroblasts as drivers of healing and cancer progression: from in vitro experiments to clinics. In: Taihao Q (ed) Molecular mechanisms of skin aging and age-related diseases. CRC Press, Boca Raton, pp 121–137

Kucera R, Treskova I, Vrzalova J, Svobodova S, Topolcan O, Fuchsova R, Rousarova M, Treska V, Kydlicek T (2014) Evaluation of IGF1 serum levels in malignant melanoma and healthy subjects. Anticancer Res 34:5217–5220

Kučera J, Dvořánková B, Smetana K Jr, Szabo P, Kodet O (2015) Fibroblasts isolated from the malignant melanoma influence phenotype of normal human keratinocytes. J Appl Biomed 13:195–198

Kulesa PM, Kasemeier-Kulesa JC, Teddy JM, Margaryan NV, Seftor EA, Seftor RE, Hendrix MJ (2006) Reprogramming metastatic melanoma cells to assume a neural crest cell-like phenotype in an embryonic microenvironment. Proc Natl Acad Sci U S A 103:3752–3757

Kulesa PM, Morrison JA, Bailey CM (2013) The neural crest and cancer: a developmental spin on melanoma. Cells Tissues Organs 198:12–21. doi:10.1159/000348418

Lacina L, Plzak J, Kodet O, Szabo P, Chovanec M, Dvorankova B, Smetana K Jr (2015) Cancer microenvironment: what can we learn from the stem cell niche. Int J Mol Sci 16(10):24094–24110. doi:10.3390/ijms161024094

Ladányi A (2015) Prognostic and predictive significance of immune cells infiltrating cutaneous melanoma. Pigment Cell Melanoma Res 28: 490–500. doi:10.1111/pcmr.12371

Lane SW, Williams DA, Watt FM (2014) Modulating the stem cell niche for tissue regeneration. Nat Biotechnol 32:795–803. doi:10.1038/nbt.2978

Lavker RM, Sun TT, Oshima H, Barrandon Y, Akiyama M, Ferraris C, Chevalier G, Favier B, Jahoda CA, Dhouailly D, Panteleyev AA, Christiano AM (2003) Hair follicle stem cells. J Investig Dermatol Symp Proc 8:28–38

Lee J, Tumbar T (2012) Hairy tale of signaling in hair follicle development and cycling. Semin Cell Dev Biol 23:906–916. doi:10.1016/j.semcdb.2012.08.003

Lee LM, Seftor EA, Bonde G, Cornell RA, Hendrix MJ (2005) The fate of human malignant melanoma cells transplanted into zebrafish embryos: assessment of migration and cell division in the absence of tumor formation. Dev Dyn 233:1560–1570

Mateu R, Živicová V, Drobná Krejčí E, Grim M, Strnad H, Vlček Č, Kolář M, Lacina L, Gál P, Borský J, Smetana K Jr, Dvořánková B (2016) Functional differences between neonatal and adult fibroblasts and keratinocytes: donor age affects epithelial-mesenchymal crosstalk in vitro. Int J Mol Med 38:1063–1074. doi:10.3892/ijmm.2016.2706

McCarty MF, Bielenberg DR, Nilsson MB, Gershenwald JE, Barnhill RL, Ahearne P, Bucana CD, Fidler IJ (2003) Epidermal hyperplasia overlying human melanoma correlates with tumour depth and angiogenesis. Melanoma Res 13:379–387

Mesa KR, Rompolas P, Greco V (2015) The dynamic duo: niche/stem cell interdependency. Stem Cell Reports 4:961–966. doi:10.1016/j.stemcr.2015.05.001

Mifková A, Kodet O, Szabo P, Kučera J, Dvořánková B, André S, Koripelly G, Gabius HJ, Lehn JM, Smetana K Jr (2014) Synthetic polyamine BPA-C8 inhibits TGF-β1-mediated conversion of human dermal fibroblast to myofibroblasts and establishment of

galectin-1-rich extracellular matrix in vitro. Chembiochem 15: 1465–1470. doi:10.1002/cbic.201402087

Mintz B, Illmensee K (1975) Normal genetically mosaic mice produced from malignant teratocarcinoma cells. Proc Natl Acad Sci U S A 72: 3585–3589

Motlík J, Klíma J, Dvoránková B, Smetana K Jr (2007) Porcine epidermal stem cells as a biomedical model for wound healing and normal/malignant epithelial cell propagation. Theriogenology 67:105–111

Nishisgori C (2015) Current concept of photocarcinogenesis. Photochem Photobiol Sci 14:1713–1721. doi:10.1039/c5pp00185d

Pacioni S, D'Alessandris QG, Giannetti S, Morgante L, De Pascalis I, Coccè V, Bonomi A, Pascucci L, Alessandri G, Pessina A, Falchetti ML, Pallini R (2015) Mesenchymal stromal cells loaded with paclitaxel induce cytotoxic damage in glioblastoma brain xenografts. Stem Cell Res Ther 6:194. doi:10.1186/s13287-015-0185-z

Pierce GB (1983) The cancer cell and its control by the embryo. Rous-Whipple Award lecture. Am J Pathol 113:117–124

Sanmamed MF, Carranza-Rua O, Alfaro C, Oñate C, Martín-Algarra S, Perez G, Landazuri SF, Gonzalez A, Gross S, Rodriguez I, Muñoz-Calleja C, Rodríguez-Ruiz M, Sangro B, López-Picazo JM, Rizzo M, Mazzolini G, Pascual JI, Andueza MP, Perez-Gracia JL, Melero I, Sanmamed MF, Carranza-Rua O, Alfaro C, Oñate C, Martín-Algarra S, Perez G, Landazuri SF, Gonzalez A, Gross S, Rodriguez I, Muñoz-Calleja C, Rodríguez-Ruiz M, Sangro B, López-Picazo JM, Rizzo M, Mazzolini G, Pascual JI, Andueza MP, Perez-Gracia JL, Melero I (2014) Serum interleukin-8 reflects tumor burden and treatment response across malignancies of multiple tissue origins. Clin Cancer Res 20:5697–5707. doi:10.1158/1078

Shao H, Kong R, Ferrari ML, Radtke F, Capobianco AJ, Liu ZJ (2015) Notch1 pathway activity determines the regulatory role of cancer-associated fibroblasts in melanoma growth and invasion. PLoS One 10:e0142815. doi:10.1371/journal.pone.0142815

Sieber-Blum M, Grim M (2004) The adult hair follicle: cradle for pluripotent neural crest stem cells. Birth Defects Res C Embryo Today 72:162–172

Sieber-Blum M, Grim M, Hu YF, Szeder V (2004) Pluripotent neural crest stem cells in the adult hair follicle. Dev Dyn 231:258–269

Sieber-Blum M, Schnell L, Grim M, Hu YF, Schneider R, Schwab ME (2006) Characterization of epidermal neural crest stem cell (EPI-NCSC) grafts in the lesioned spinal cord. Mol Cell Neurosci 32:67–81

Smetana K Jr, Szabo P, Gal P, André S, Gabius HJ, Kodet O, Dvořánková B (2015) Emerging role of tissue lectins as microenvironmental effectors in tumors and wounds. Histol Histopathol 30:293–309

Strnad H, Lacina L, Kolár M, Cada Z, Vlcek C, Dvoránková B, Betka J, Plzák J, Chovanec M, Sáchová J, Valach J, Urbanová M, Smetana K Jr (2010) Head and neck squamous cancer stromal fibroblasts produce growth factors influencing phenotype of normal human keratinocytes. Histochem Cell Biol 133:201–211. doi:10.1007/s00418-009-0661-6

Szabo P, Kolář M, Dvořánková B, Lacina L, Štork J, Vlček Č, Strnad H, Tvrdek M, Smetana K Jr (2011) Mouse 3T3 fibroblasts under the influence of fibroblasts isolated from stroma of human basal cell carcinoma acquire properties of multipotent stem cells. Biol Cell 103:233–248. doi:10.1042/BC20100113

Trylcova J, Busek P, Smetana K Jr, Balaziova E, Dvorankova B, Mifkova A, Sedo A (2015) Effect of cancer-associated fibroblasts on the migration of glioma cells in vitro. Tumour Biol 36:5873–5879. doi:10.1007/s13277-015-3259-8

Umansky V, Utikal J, Gebhardt C (2016) Predictive immune markers in advanced melanoma patients treated with ipilimumab. Oncoimmunology 5:e1158901. doi:10.1080/2162402X.2016.1158901

ÚZIS (2011) Cancer incidence in the Czech Republic. NOR ČR, ÚZIS ČR, pp 1–263

Valach J, Fík Z, Strnad H, Chovanec M, Plzák J, Cada Z, Szabo P, Sáchová J, Hroudová M, Urbanová M, Steffl M, Pačes J, Mazánek J, Vlček C, Betka J, Kaltner H, André S, Gabius HJ, Kodet R, Smetana K Jr, Gál P, Kolář M (2012) Smooth muscle actin-expressing stromal fibroblasts in head and neck squamous cell carcinoma: increased expression of galectin-1 and induction of poor prognosis factors. Int J Cancer 131: 2499–2508. doi:10.1002/ijc.27550

Van Kilsdonk JW, Bergers M, Van Kempen LC, Schalkwijk J, Swart GW (2010) Keratinocytes drive melanoma invasion in a reconstructed skin model. Melanoma Res 20:372–380. doi:10.1097/CMR.0b013e32833d8d70

Whipple CA, Brinckerhoff CE (2014) BRAF(V600E) melanoma cells secrete factors that activate stromal fibroblasts and enhance tumourigenicity. Br J Cancer 111:1625–1633. doi:10.1038/bjc.2014.452

Woodward WA, Hill RP (2016) Cancer stem cells. Recent Results Cancer Res 198:25–44. doi:10.1007/978-3-662-49651-0_2

Zhang D, Ighaniyan S, Stathopoulos L, Rollo B, Landman K, Hutson J, Newgreen D (2014) The neural crest: a versatile organ system. Birth Defects Res C Embryo Today 102:275–298. doi:10.1002/bdrc.2108