но в виде единичных зеленоватых вкраплений (Matheson, Kaufman, 2017).

Таким образом, при воздействии дакарбазином на клетки меланомы B16 в клеточном цикле увеличивается доля G_0-положительных клеток, а также происходит снижение доли клеток в фазах G_1 и G_2. С учетом сохраняющейся способности покоящихся G_0-положительных клеток к пролиферации, феномен перехода в G_0, в равной степени как и сама популяция таких клеток, может быть целенаправленным объектом в рамках противоопухолевой терапии. Помимо этого, ранее мы наблюдали схожие изменения в клетках меланомы после воздействия таргетным препарата вемурафениб, ингибитора белка BRAF (Николаева, 2020), что может указывать на универсальность подобных изменений вне зависимости от действующего лекарственного средства, и подчеркивает необходимость разработки целенаправленных стратегий в отношении G_0-положительных клеток.

БЛАГОДАРНОСТИ

Результаты работы методом проточной цитометрии получены в Центре коллективного пользования Красноярского научного центра СО РАН.

ФИНАНСИРОВАНИЕ РАБОТЫ

Работа выполнена при финансовой поддержке Российского научного фонда (проект № 19-15-00110, https://rscf.ru/project/19-15-00110/).

СОБЛЮДЕНИЕ ЭТИЧЕСКИХ СТАНДАРТОВ

Экспериментов с участием животных или людей авторы не проводили.

КОНФЛИКТ ИНТЕРЕСОВ

Авторы заявляют об отсутствии конфликта интересов.

СПИСОК ЛИТЕРАТУРЫ

Зинченко И.С., Палкина Н.В., Рукша Т.Г. 2022. Изменение профиля микроРНК в клетках меланомы, резистентных к действию дакарбазина. Цитология. Т. 64. № 1. С. 26. (*Zinchenko I.S., Palkina N.V., Ruksha T.G.* 2022.Changes in miRNA profile in melanoma cells resistant to dacarbazine. Tsitologiya. V. 64. № 1. S. 26.)

Николаева Е.Д., Дубовцева И.Ю., Белоногов Р.Н., Наркевич А.Н., Мошев А.В., Савченко А.А., Рукша Т.Г. 2020. Вемурафениб индуцирует повышение уровня дормантных (Ki-67-негативных) клеток при BRAF-негативном статусе меланомы. Цитология. V. 62. № 11. С. 793. (*Nikolaeva E.D., Dubovtseva I.Yu., Belonogov R.N., Narkevich A.N., Moshev A.V., Savchenko A.A., Ruksha T.G.* 2020. Vemurafenib induces an increase in dormant (Ki-67-negative) cells in BRAF-negative melanoma. Tsitologiya. V.62. № 11. P. 793.)

Al-Qatati A., Aliwaini S. 2017. Combined pitavastatin and dacarbazine treatment activates apoptosis and autophagy resulting in synergistic cytotoxicity in melanoma cells. Oncol. Letters. V. 14. P. 7993.

Avci N.G., Ebrahimzadeh-Pustchi S., Akay Y.M., Esquenazi Y., Tandon N., Zhu J.J., Akay M. 2020. NF-κB inhibitor with Temozolomide results in significant apoptosis in glioblastoma via the NF-κB(p65) and actin cytoskeleton regulatory pathways. Sci. Rep. V. 10. P. 13352.

Beaumont K.A., Hill D.S., Daignault S.M., Lui G., Sharp D.M., Gabrielli B., Weninger W., Haass N.K. 2016. Cell cycle phase-specific drug resistance as an escape mechanism of melanoma cells. J. Invest. Dermatol. V. 136. P. 1479.

Cappel S.D., Mark K.G., Garbett D., Pack L.R., Rape M., Meyer T. 2018. EMI1 switches from being a substrate to an inhibitor of APC/C^{CDH1} to start the cell cycle. Nature. V. 7709. P. 313

Chapman P.B., Hauschild A., Robert C., Haanen J.B., Ascierto P., Larkin J., Dummer R., Garbe C., Testori A., Maio M., Hogg D., Lorigan P., Lebbe C., Jouary T., Schadendorf D. et al. 2011. Improved survival with vemurafenib in melanoma with BRAF V600E mutation. New England J. Med. V. 364. P. 2507.

Cheng L., Lopez-Beltran A., Massari F., MacLennan G.T., Montironi R. 2018. Molecular testing for BRAF mutations to inform melanoma treatment decisions: a move toward precision medicine. Mod. Pathol. V. 31. P. 24.

Domingues B., Lopes J.M., Soares P., Populo H. 2018. Melanoma treatment in review. Immunotargets Ther. V. 7. P. 35–49.

Fares J., Fares M.Y., Khachfe H.H., Salhab H.A., Fares Y. 2020. Molecular principles of metastasis: a hallmark of cancer revisited. Signal Transd. Targeted Ther. V. 5. P. 28.

Gyrylova S.N., Aksenenko M.B., Gavrilyuk D.V., Palkina N.V., Dyhno Y.A., Ruksha T.G., Artyukhov I.P. 2014. Melanoma incidence mortality rates and clinico-pathological types in the Siberian area of the Russian Federation. Asian Pac. J. Cancer Prev. V. 15. P. 2201.

Huncharek M., Caubet J.F., McGarry R. 2001. Single-agent DTIC versus combination chemotherapy with or without immunotherapy in metastatic melanoma: a meta-analysis of 3273 patients from 20 randomized trials. Melanoma Res. V. 11. P. 75.

Huang X., He J.X., Zhang H.T., Sun K., Yang J., Wang H.J., Zhang H.X., Guo Z.Z., Zha Z.G., Zhou C.R. 2017. Effect of dacarbazine on CD44 in live melanoma cells as measured by atomic force microscopy-based nanoscopy. Int. J. Nanomed. V. 12. P. 8867.

Hill D.S., Lovat P.E., Haass N.K. 2014. Induction of endoplasmic reticulum stress as a strategy for melanoma therapy: is there a future? Melanoma Manag. V. 2. P. 127.

Joshi K.S., Rathos M.J., Mahajan P., Wagh V., Shenoy S., Bhatia D., Sharma S. 2007. P276-00, a novel cyclin-dependent inhibitor induces G_1–G_2 arrest, shows antitumor activity on cisplatin-resistant cells and significant in vivo efficacy in tumor models. Mol. Cancer Ther. V. 6. P. 926.

Li W., Sanki A., Karim R.Z., Thompson J.F., Lee C.S., Zhuang L.Q., Zhuang L., McCarthy S.W., Scolyer R.A. 2006. The role of cell cycle regulatory proteins in the pathogenesis of melanoma. Pathol. V. 38. P. 287.

Lui P., Cashin R., Machado M., Hemels M., Corey-Lisle P.K., Einarson T.R. 2007. Treatments for metastatic melanoma:

synthesis of evidence from randomized trials. Cancer Treat. Rev. V. 33. P. 665.

Manke I.A., Nguyen A., Lim D., Stewart M.Q., Elia A.E., Yaffe M.B. 2005. MAPKAP kinase-2 is a cell cycle checkpoint kinase that regulates the G_2/M transition and S phase progression in response to UV irradiation. Mol. Cell. V. 17. P. 37.

Mattia G., Puglisi R., Ascione B., Malorni W., Carè A., Matarrese P. 2018. Cell death-based treatments of melanoma: conventional treatments and new therapeutic strategies. Cell Death Dis. V. 9. P. 112.

Matheson T.D., Kaufman P.D. 2017. The p150N domain of chromatin assembly factor-1 regulates Ki-67 accumulation on the mitotic perichromosomal layer. Mol. Biol. Cell. V. 28. P. 21.

McConnell A.M., Zon L.I. 2021. Dissecting melanocytes to predict melanoma. Nat. Cell Biol. V. 23. P. 930.

Mhaidat N.M., Zhang X.D., Jiang C.C., Hersey P. 2007. Docetaxel-induced apoptosis of human melanoma is mediated by activation of c-Jun NH2-terminal kinase and inhibited by the mitogen-activated protein kinase extracellular signal-regulated kinase 1/2 pathway. Clin. Cancer Res. V. 13. P. 1308.

Olszewska-Słonina D.M., Styczyńisk J., Drewa T.A., Olszewski K.J., Czajkowski R. 2005. B16 and cloudman S91 mouse melanoma cells susceptibility to apoptosis after dacarbazine treatment. Acta Pol. Pharm. V. 62. P. 473.

Oliferenko S., Kaverina I., Small J.V., Huber L.A. 2000. Brief report hyaluronic acid (HA) binding to CD44 activates Rac1 and induces lamellipodia outgrowth. J. Cell Biol. V. 148. P. 1159.

Ossowski L., Aguirre-Ghiso J.A. 2010. Dormancy of metastatic melanoma. Pigment Cell Melanoma Res. V. 23. P. 41.

Pawlowska E., Szczepanska J., Szatkowska M., Blasiak J. 2018. An interplay between senescence, apoptosis and autophagy in glioblastoma multiforme-role in pathogenesis and therapeutic perspective. Int. J. Mol. Sci. V. 19. P. 889.

Reyes-Reyes E.M., Jin Z., Vaisberg A.J., Hammond G.B., Bates P.J. 2013. Physangulidine A, a withanolide from *Physalis angulata*, perturbs the cell cycle and induces cell death by apoptosis in prostate cancer cells. J. Nat. Prod. V. 1. P.2.

Ruksha T.G. 2019. MicroRNAs' control of cancer cell dormancy. Cell Div. V. 14. P. 11.

Risson E., Nobre A.R., Maguer-Satta V., Aguirre-Ghiso J.A. 2020. The current paradigm and challenges ahead for the dormancy of disseminated tumor cells. Nat. Cancer. V. 7. P. 672.

Ralhan R., Kaur J. 2007. Alkylating agents and cancer therapy. Expert Opin. Ther. Path. V. 17. P. 1061.

Shah M.A., Schwartz G.K. 2001. Cell cycle-mediated drug resistance: an emerging concept in cancer therapy. Clin. Cancer Res. V. 7. P. 2168.

Sheppard K.E., McArthur G.A. 2013. The cell-cycle regulator CDK4: an emerging therapeutic target in melanoma. Clin Cancer Res. V. 19. P. 5320.

Sun X., Kaufman P.D. 2018. Ki-67: more than a proliferation marker. Chromosoma. V. 127. P. 175.

Tyumentseva A., Averchuk A., Palkina N., Zinchenko I., Moshev A., Savchenko A., Ruksha T. 2021. Transcriptomic profiling revealed plexin A2 downregulation with migration and invasion alteration in dacarbazine-treated primary melanoma cells. Front. Oncol. V. 11. P. 732501.

Xu W., McArthur G. 2016. Cell cycle regulation and melanoma. Curr. Oncol. Rep. V. 18. P. 34.

Wang H., Feng W., Lu Y., Li H., Xiang W., Chen Z., He M., Zhao L., Sun X., Lei B., Qi S., Liu Y. 2016. Expression of dynein, cytoplasmic 2, heavy chain 1 (DHC2) associated with glioblastoma cell resistance to temozolomide. Sci. Rep. V. 6. P. 28948.

Cell Cycle Phase Distribution in B16 Melanoma Cells under Dacarbazine Treatment

E. Z. Lapkina[a], A. R. Esimbekova[a], V. D. Beleniuk[b], A. A. Savchenko[b], and T. G. Ruksha[a, *]

[a] *Voino-Yasenetsky Krasnoyarsk State Medical University, Krasnoyarsk, 660022 Russia*

[b] *Research Institute for Medical Problems in the North , Krasnoyarsk, 660022 Russia*

**e-mail: tatyana_ruksha@mail.ru*

Reversible transition to the resting phase (G_0) of the cell cycle is implicated in the development of cancer cells drug resistance. The effect of dacarbazine on B16 melanoma cells was used to study the distribution of phases of the cell cycle of melanoma cells. The ability of cells to enter into a G_0 phase of cell cycle was determined by immunocytochemistry and flow cytometry based on the negative staining of Ki-67 protein. The pool of G_0-positive cells was increased with subsequent a decrease in the proportion of cells in the G_1 and G_2 phases in the cell cycle in dacarbazine-treated B16 melanoma cells.

Keywords: melanoma, B16, dacarbazine, cell cycle, G_0 phase, Ki-67, cell dormancy, cell senescence