ESMO Research Fellowship

(November 2019 – October 2020)

Mariela Borisova Vasileva-Slaveva

FINAL REPORT

Host Institute: **EXTRO-Lab, Department of Therapeutic Radiology and Oncology, Medical University of Innsbruck**
Mentor: **Prof. Ira-Ida Skvortsova, MD, PhD**
Project title: **Breast cancer metastasis and lipid metabolism implication**

Home Institute: **Alexandrovska University Hospital Sofia, Bulgaria**

Introduction

Obesity, defined as a BMI ≥30kg/m^2, is currently diagnosed in more than 13% of adult population(1). It has strong impact on health since people with BMI between 30-35 kg/m^2 have reduced median overall survival (OS) with 2-4 years (2). In breast cancer (BC) patients obesity is negative prognostic factors for survival(3) and the strongest negative impact is observed in women with hormone receptor positive cancer (4) compared to almost no effect in the other subtypes (5). Obesity has been related to worse outcomes in all treatment modalities – surgery, radiotherapy and systemic therapy and the cosmetics.(6) Higher BC recurrence rate after radiotherapy has been found in women with increased BMI.(7) Since cancer progression is a main cause of death in patients with solid tumors up to 15 years after diagnosis (8, 9), investigating the obesity related pro-metastatic and pro-survival pathways diminishing radiation response of breast carcinoma cells is essential.

Obesity is associated with alteration in adipose tissue and lipid metabolism.(10) Changes in lipid metabolism and, consequently, lipid composition could modulate therapy response of cancer cells.(11) Many lipids have been found to be increased in cancer, as for example sphingolipid 1-phosphate in BC. Studies also suggested that the choline-containing lipids and phospholipids in cancer cells could increase during the metastatic process.(12)

Adipocytes in BC microenvironment can communicate with cancer cells and this crosstalk leads to phenotypical and functional changes of both cell types (13). Adipokines, secreted by adipocytes, promote breast cancer initiation, proliferation, invasion, can have anti-tumorigenic effect (14) or can modulate response to treatment.(15) Additionally, the co-culture of mature adipocytes and breast cancer cells results in heterogeneous and probably breast cancer cell type specific communications(16).

Dipeptidyl peptidase DPPIV/CD26 is an adipokine - type II transmembrane multifunctional glycoprotein involved in various biological and pathologic processes. DPPIV degrades incretins such as glucagon like peptide 1 (GLP-1) and glucose-dependent insulinotropic polypeptide,(17) leading to reduced insulin secretion and it is a target for treatment of diabetes mellitus. Its expression is positively correlated with BMI (18). In cultured cells, DPPIV knockdown induced epithelial to mesenchymal transition (EMT) and cell migration and inhibition of DPPIV increases the metastatic potential of breast cancer (19). Currently, there are several DPPIV inhibitors available, which have different binding modes in the DPPIV active center. Among them Teneligliptin has the highest inhibitory function toward DPPIV (18). It also has antioxidant properties and ameliorates oxidative stress (20).

Rationale and Aim

We aimed to elucidate whether affected lipid metabolism can contribute to therapy response in breast cancer patients.

Experimental design

We investigated the lipid metabolism and radio sensitivity of three types of breast cancer cell lines: MDA – MB-231 (triple-negative, TN), T47D (hormone receptor positive, HR+) and AU565 (Her2-positive, HER2+) in correlation with their invasive capacities.

BC cells (MDA – MB-231; T47D and AU565 cell lines) were exposed to repetitive migration through an uncoated 8 µm-pore membrane in order to obtain invasive (INV) BC cells. Both parental cells and derived INV cells are accessed for their:

- INV potential - CytoSelect Invasion Assay (Collagen I or Laminin);
- Proteomics and lipidomics – nano-LC-mass spectrometry (Omics Technologies Inc., USA)
- Metabolic activities – Agilent Seahorse Metabolic Analyzer XFp
- Metastatic abilities - in vivo experiments using nude mice;
- Radiation response - exposure to photon-based ionizing radiation and assessment of their clonogenic survival (Figure 1);

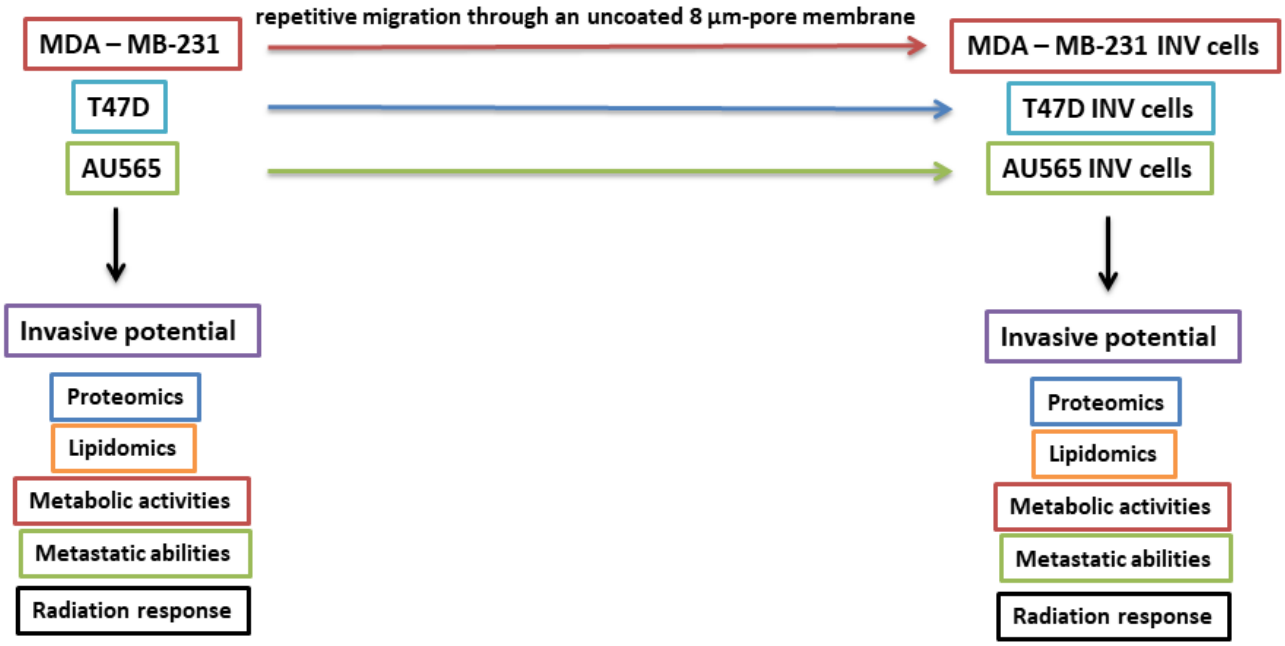

Figure 1. Schedule for assessment of the parental and the invasive cells

The response to radiotherapy was evaluated as we exposed parental and invasive BC cells to photon-based ionizing radiation and investigated how their clonogenic survival is affected. The parental and invasive MDA-MB-231 cells did not show differences in clonogenic survival; T47D-INV became markedly more resistant to ionizing radiation; Au565-INV cells became also more resistant, as the effect was less significant then the observed in T47D. We further applied this model of response to radiotherapy in the investigation of lipid profile of BC cells and their expression of adipokines.

1. Lipidomics - Lipid profile of the parental and invasive cell lines was analysed as each lipid class, identified from the samples analysis, was related to a category, class and, wherever possible, to a subclass according to the nomenclature of LIPID Metabolites and Pathways Strategy (LIPID MAPS). When a lipid was identified multiple times in the analysis of a certain cell line, the average value of the peak areas was calculated and used for further analysis.
2. Adipokines - The secretion of adipokines by the different BC cell lines was investigated with Proteomics Profiling Array (R&D Systems, Inc.).
3. DPPIV - The role of DPPIV /CD26 was tested by investigating its effect on BC cells viability, clonogenic survival and migratory abilities

Results, Conclusions and Future Perspectives

1. Lipidomics

We first investigated how the lipid profile of the cell lines is changed in the process of acquirement of increased invasive abilities. We analysed the common lipids found in the parental cell lines and their invasive counterparts. The highest percentage of lipids in common was found in MDA-MB-231 cells (74%), followed by Au 565 (68.3%) and T47D (55.9%).We also found some lipids, which were unique for a specific subtype as PIP3 species, which were found only in Au 565 parental and were not detected in Au 565 invasive.
We further investigated the common lipids between parental and invasive cell lines in order to find differences in their expression. Lipids, which were expressed more than 1.5 times in invasive cells compared to parental were considered for upregulated; lipids, which levels of expression in invasive cells was 2/3 of the observed in parental were considered as downregulated. We found 7 lipids species, which were downregulated in all invasive cell lines and 8 lipid species which were all upregulated.

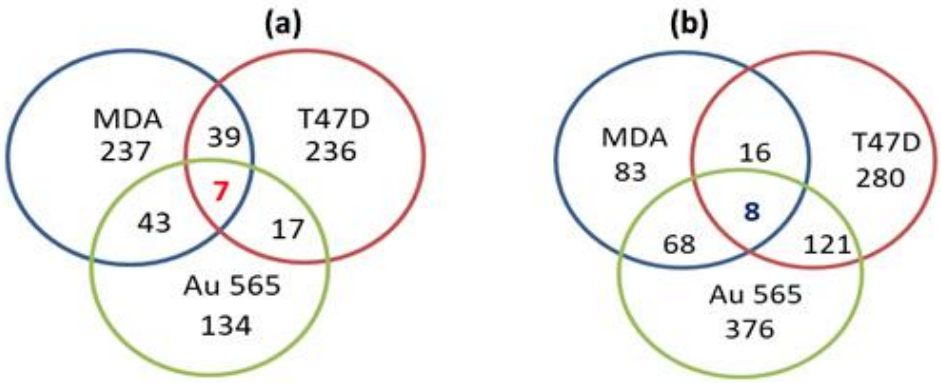

Figure 2. Lipid species in common between parental and invasive cells and (a) downregulated in invasive; (b) upregulated in invasive.

Taking into accountthe model of response to radiotherapy observed in the three cell lines we searched for a lipids class with the similar pattern of expression as the observed for the radiotherapy response. We investigated the ratio of lipid content from each main class in invasive to parental cells of each cell line. The results are shown on Table 1.

Table 1. Lipids expression in invasive to parental cell lines

Category	Main class	MDA INV to PAR	T47D INV to PAR	Au 565 INV to PAR
	Fatty Acids and Conjugates	*	0.48165744	0.883675955
Fatty Acyls (FA)	Fatty esters		2.837753047	4.512442898
	Diradylglycerols	1.158191	0.450112281	2.661971867
	Triradylglycerols	0.744085	1.115601537	0.509453434
Glicerolipids (GL)	Glycosyldiradylglycerols	0.563908	6.292520966	3.555041726
	Other Glycerophospholipids	0.210936	0.352815478	0.630401557
	Glycerophosphocholines	0.958423	1.439405398	1.231485791
	Glycerophosphoethanolamines	0.750787	0.771262522	0.957254397
	Glycerophosphoserines	0.622584	0.29316928	2.139268982
Glycero-phospholipids (GP)	Glycerophosphoglycerols	0.445577	2.312552594	1.299299303

	Glycerophosphoinositols	0.556589	1.087791949	1.422207647
	Glycerophosphates		0.164648961	0.223861002
	Glycerophosphoglycerophosphoglycerols	0.633888	6.636934643	0.301948049
	Sphingoid bases	2.107092	0.436607635	1.280312639
	Ceramides	1.337889	2.508846749	0.860139693
	Phosphosphingolipids	1.428675	0.902227851	1.487014593
	Neutral glycosphingolipids	0.962641	0.398644813	0.372531757
Sphingolipids (SP)	Acidic glycosphingolipids	0	1.176956815	8.172252677
Sterol Lipids (ST)	Sterols	0.546382		25.62431514

*When the certain lipid class is presented in only one of the cell lines –parental or invasive, the results are not shown

Ceramides were the class of lipids most responding to the pattern of expression that we are interested in. Additionally, these results are in line with the higher ceramides content in MDA-MB-231 and T47D cells with radioresistant phenotype (that have been repetitively irradiated at a total dose of 100 Gy) compared to parental, which are not included in the current study.

Higher level of ceramides in tumor tissue has been related to increase BC metastatic potential. (21) However, since ceramides include many species with different functions further analysis within this class of lipids is needed.

2. Adipokines

The secretion of adipokines by the different BC cell lines was investigated with Proteomics Profiling Array (R&D Systems, Inc.). We compared the levels of expression of adipokines in invasive cells to the expression in their parental counterparts and again looked for the pattern of expression similar to the pattern of radiotherapy response. The adipokines, shown in Table 2. are the ones most corresponding to the pattern of radioresistance of interest.

Table 2. Adipokines, expressed in the same manner with the pattern of the observed response to radiotherapy and the ratio of expression INV/parental cells

Adipokines	MDA-MB-231-INV	T47D-INV	Au565-INV
DPPIV/CD26	1.52	5.71	0.43
Nidogen-1/Entactin	1.68	4.55	0.77
Proprotein Convertase 9/PCSK9	0.90	2.27	0.73
LIF	1.17	2.11	0.79

3. DPPIV

We have decided to further evaluate the role of DPPIV /CD26 as possible key determinant of radiotherapy response in breast cancer cell lines.

First, we have detected that DPPIV /CD26 expression in the INV cells correlates with glucose uptake and total volume of intracellular lipids. Thus, invasive hormone receptor-positive breast carcinoma T47D-INV cells reveal an upregulation of DPP4/CD26 and enhanced glucose uptake and lipid content, whereas Her2-positive Au565-INV cells with down-regulation of DPP4/CD26 and significant decrease of glucose uptake and intracellular lipid content. MDA-MB-231-INV changed neither DPP4/CD26, nor glucose uptake nor total lipid volume.

Secondly, we investigated the effect of the DPPIV inhibitors Teneligliptin and p32/98 on both parental and invasive cells viability, clonogenic survival in combination treatment with radiotherapy or alone and migratory abilities.

For the purposes of those experiments all cell lines were grown in RPMI1640 medium supplemented with 2 mM L-glutamine, 50 U/mL penicillin, 50 µg/mL streptomycin, and 10% fetal calf serum (FCS) (Thermo Fisher Scientific, Vienna, Austria). Cell cultures were maintained in a 5% CO_2 humidified atmosphere.

1) Parental and invasive cells viability

Parental MDA-MB-231 and Au565 cells and their invasive pairs were seeded (1 x 10^5) in 3.0-mL in 6-well plates; parental T47D and T47D-INV cells were seeded 2 x 10^5 and 2,5 x 10^5, respectively. All cells were incubated for 24 hours at 37°C and then treated with different concentrations of Teneligliptin (0.1µM, 1,0µM, 3,0µM and 10µM). Cells were trypsinized and counted using Beckman Coulter Vi-CELL AS cell viability analyzer (Beckman Coulter, Fullerton, CA, USA) 48h and 72 hours after treatment with the inhibitors. Based on these experiments we couldn't find significant effect of Teneligliptin on cells viability.

2) Clonogenic survival after treatment with DPPIV inhibitors alone and after combination treatment with radiotherapy

First, we investigated the effect of Teneligliptin and p32/92 alone on the clonogenic ability of the parental and invasive cells. Parental MDA-MB-231, T47D and Au565 and their invasive pairs were seeded 500 cells in 3.0-mL in 6-well plates. All cells were incubated for 24 hours at 37°C and then treated with different concentrations of Teneligliptin (0.1µM, 1,0µM, 3,0µM and 10µM) and p32/92 (1.0µM, 10µM, 50µM and 100µM) in duplicates. Each pair of parental and invasive cells were incubated for the same time and stained with crystal violet in the same day. The incubation time for the different cell lines was different, since they have different doubling time (as previously reported) and clonogenic abilities.
We observed close, but different response to treatment with Teneligliptin and p32/98 among the MDA-MB-231 and Au 565 parental and invasive cells as Teneligliptin was suppressing or not affecting and p32/98 was stimulating the clonogenic abilities of the cells. In T47D invasive cells we observed 10 times increase in the surviving fraction (SF), when cells were treated with 50µM p32/98, compared to control and more than 6 times increase of SF, when treated with 0.1 µM Teneligliptin. (Figure 3). These results are in line with our previous findings that T47D-INV cells reveal an upregulation of DPPIV, whereas Au565-INV cells reveal down-regulation and MDA-MB-231-INV revealed no change in DPPIV.

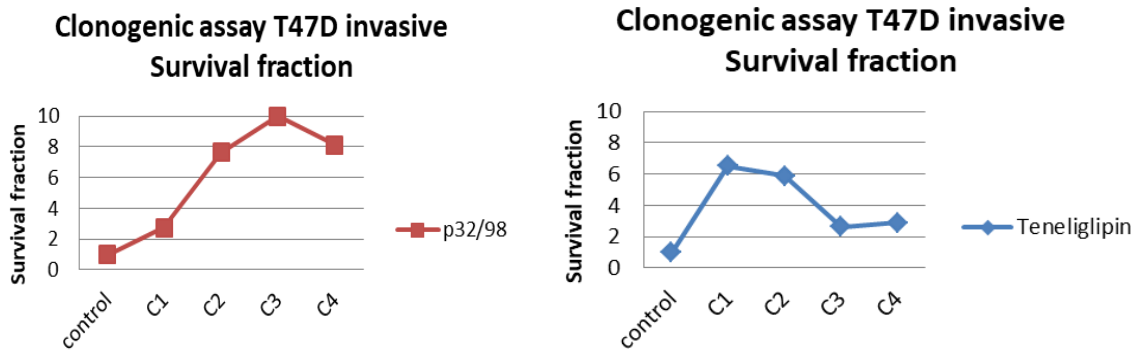

Figure 3. Surviving fraction of T47D invasive cells when treated with different concentrations of p32/98 or Teneligliptin.

Secondly, we investigated the effect of combined treatment of Teneligliptin and radiotherapy (RT) on MDA-MB-231 parental and invasive cells in two treatment schedules– if the treatment is applied consequently or simultaneously.
MDA-MB-231 parental and invasive breast cancer cells were seeded (1 x 10^5) in 3.0-mL medium in 6-well plates. After 24 hours incubation cells were treated with different concentrations of Teneligliptin (0.1µM, 1,0µM, 3,0µM and 10µM) and incubate for another 24 hours. After that 500 cells from each treatment concentration were seeded in 6 well plates in duplicates and incubated for 24 hours, when radiotherapy in dose of 2Gy and 6Gy was applied.
In the second approach 500 cells from the MDA-MB-231 parental and invasive cells were seeded in 6 well plates. In 24 hours the cells were treated with different concentration of Teneligliptin and 4 hours later were irradiated with dose of 2Gy and 6Gy.
Cells, treated without irradiation and with 2Gy, from both experiments were incubated for 9 days. At this point there were

no clones developed by cells treated with 6Gy. MDA-MB-231 invasive cells were able to form more clones compared to the parental. Cells, treated with 6Gy, were incubated for 16 days in total and then stained. Only big single clones were observed. We observed dose dependent response to treatment in both parental and invasive cells, most significant, when cells are treated with 3,0μM Teneligliptin. Further experiments are needed to confirm these results in this and the others cell lines.

3) Migratory abilities – scratch assay

Migratory capacities of the investigated BC cells were assessed with automated live cell imager Lionheart FX (BioTek, Vermont, USA). When a full cell monolayer was obtained a scratch was made in a straight line with a p200 pipet tip. The medium was then changed with medium containing different concentrations of Teneligliptin and cells were incubated in the live cell imager for 72 hours. Images were obtained every 2 hours. We observed the fastest closure in both control and treated cells in Au parental cells – for 22 hours and 18 hours, respectively. T47D invasive cells were not affected by Teneligliptin and the "scratch" in those cells monolayer was never closed by the end of the incubation period. In T47D parental cells the "scratch" was not closed at the 72hour in the control and was almost closed in the cells treated with 10μM Teneligliptin. Therefore we could conclude that Teneligliptin is affecting the migratory abilities of MDA-MB-231 INV cells, Au 565 parental and invasive cells and most significantly – the T47D parental cells.

Further experiments are needed in order to confirm the reported results.

List of Publications and Presentations Resulting from the Translational Research Project "* Breast cancer metastasis and lipid metabolism implication*"

G. Negro, B. Aschenbrenner, S. K. Brezar, M. Cemazar, A. Coer, G. Gasljevic, D.Savic, M. Sorokin, A. Buzdin, M. Callari. I. Kvitsaridze, A. Jewett, M. Vasileva-Slaveva, U. Ganswindt1, I. Skvortsova S. Skvortsov. Molecular heterogeneity in breast carcinoma cells with increased invasive capacities Radiol Oncol 2020; 54(1): 103-118

List of Publications and Presentations resulting from other projects during the fellowship period

Poster presentations:

1. M. Vasileva-Slaveva, M. Dimitrova-Mladenova, J. Simeonova, A. Konsoulova, E. Valerieva, Y. Marincheva, E. Chupryna, H. Ivanovska-Peneva, S. Maslyankov, A. Yordanov. Surgery of the primary tumor in de novo metastatic breast cancer patients. European Journal of Surgical Oncology supplement. (*in press*).
2. M. Vasileva-Slaveva, Y. Marincheva, A. Valerieva, K. Angelov, D. Kostova-Lefterova, Kr. Nikolov, et al. Outcomes of patients with brain metastases in Bulgaria. Cancer Metastases conference 2019, Seefeld-in-Tirol, Austria, Abstract book - ISBN: 978-2-88963-094-3 DOI: 10.3389/978-2-88963-094-3. p 139-141
3. A. Konsoulova, Y. Marincheva, Kr. Nikolov, P. Bochev, Zh. Dancheva, ….. M. Vasileva-Slaveva. Population-based study on epidemiology and survival of patients with brain metastases in Bulgaria. Cancer Metastases conference 2019, Seefeld-in-Tirol, Austria, Abstract book - ISBN: 978-2-88963-094-3 DOI: 10.3389/978-2-88963-094-3. p 191-193
4. M. Vasileva, Z. Inic, M. Jevric, at al. Men with breast cancer - surgical management in the metastatic setting, Abstract book of the Advanced Breast Cancer 5th ESO-ESMO International Consensus Conference 2019 Lisbon, Portugal The Breast Journal, vol 48, Supp 2, November 2019, Abstract PO105, S62;
5. A. Konsoulova, Z. Inic, M. Jevric, A. Jordanov, G. Zieafetova, ……, M. Vasileva. Men with breast cancer –role of endocrine treatment for disease progression, Abstract book of the Advanced Breast Cancer 5th ESO-ESMO International Consensus Conference 2019 Lisbon, Portugal The Breast Journal, vol 48, Supp 2, November 2019, Abstract PO87, S55;
6. A. Konsoulova, A. Jordanov, G. Zieafetova, Ts. Kondzhova, K. Nikolov, ….. M. Vasileva. Men with breast cancer- survival and prognostic factors in the metastatic setting in Bulgaria, Abstract book of the Advanced Breast Cancer 5th ESO-ESMO International Consensus Conference 2019 Lisbon, Portugal The Breast Journal, vol 48, Supp 2, November 2019, Abstract PO81, S52;

Presentations:

M. Vasileva, *A. Konsoulova.* Building a career of young oncologist. How it works? Second winter school of Medical oncology of the Young oncologist club – Bulgaria. 1-3 Nov 2019

Publications:

1. M. Vasileva-Slaveva, J. Simeonova, A. Konsoulova , A. Yordanov, E. Valerieva, Y. Marincheva, H. Ivanovska-Peneva, D. Kostova-Lefterova, S. Maslyankov, M. Dimitrova-Mladenova. Patterns of metastatic spread and survival outcomes in Bulgarian breast cancer patients – submitted
2. A. Yordanov, I. Ivanov, T Dineva, S. Popovska, M. Karcheva, S. Kostov, S. Slavchev, S. Strashilov, A. Konsoulova, M. Vasileva-Slaveva. The role of EBV and HPV infection in lymphoepithelioma-like cervical cancer. Systematic review and single center experience. European Journal of Gynaecological Oncology (in print)
3. S. Strashilov, A. Yordanov, M. Vasileva-Slaveva, A.Konsoulova. Re-excision within a radius of 2 cm in patients with melanoma of the skin – sufficient for local oncological radicalness. 2020 Jun. Arch Med Sci
4. A. Yordanov, L.Tantchev, P. Vasileva, S. Strashilov, M. Vasileva-Slaveva and A. Konsoulova Uterine smooth muscle tumors of uncertain malignant potential: single-center experience and review of the literature. Prz Menopauzalny. 2020 Mar; 19(1): 30–34.
5. A. Yordanov, M. Karamanliev, L.Tantchev, A. Konsoulova, S.I Strashilov, M.Vasileva-Slaveva. Mucoepidermoid Carcinoma of the Uterine Cervix—Single-Center Study Over a 10-Year Period. Medicina, 2020,Vol. 56, Issue 1, p37.
6. A. Yordanov, M. Karamanliev, M.Karcheva, A. Konsoulova, M. Vasileva-Slaveva, S. Strashilov. Single-Center Study of Lymphoepithelioma-Like Carcinoma of Uterine Cervix over a 10-Year Period. Medicina, 2019, Vol.55, Issue 12, p780.

Selection of Courses and Workshops Attended During the Fellowship

October 2020 – ESSO 2020 Virtual Congress
September/October 2020 – ESMO 2020 Virtual Congress
October 2020 - Breast Cancer in Young Women: Fifth ESO-ESMO International Symposium
August 2020 - ESMO Academy
March 2020 – Think Pink Europe Innovation award – second prize
December 2019 - International Conference „Cancer Metastasis“, Seefeld-in-Tirol, Austria
December 2019 - ESMO Advanced Course on Biomarkers for Precision Medicine, Zurich, Switzerland
November 2019 - Advanced Breast Cancer 5th ESO-ESMO International Consensus Conference 2019 Lisbon, Portugal

Personal Statement

The ESMO translational research fellowship gave me once in a lifetime opportunity to learn and gain new experience in a field that I wouldn't otherwise have the chance to explore in such details. In the year that has past I have learnt so much, every day. I have enriched my view and changed my perspective for research, not only translational, but in the whole field of oncology. Beside scientific work, the last year have taught me to think more for others and strengthen my belief that we, working together, can actually have an impact on our life and life of the others.

Before coming to the ESMO TR fellowship I wanted to see what would be to be involved in a cutting –edge research that can really make the change for people. Working in a lab, learning so much and seeing the gaps in research and treatment led to the development of the project ROSE: Rising the Oncology patients' Survival among the European countries- a project for breast cancer digital diary, which is connecting translational and clinical research.

The year that has past was also very hard for me as I believe for many others. It was a year in which we all had to re-think our priorities, sacrifice our comfort and realize that we could be running out of time. This year was my 33th year and above all it was truly special one. I want to thank to ESMO, for giving me this chance, for changing my life and for supporting me all the way.

References

1. Organization WH. Obesity and overweight - key facts 2020 [Available from: https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight.
2. Whitlock G, Lewington S, Sherliker P, Clarke R, Emberson J, Halsey J, et al. Body-mass index and cause-specific mortality in 900 000 adults: collaborative analyses of 57 prospective studies. Lancet. 2009;373(9669):1083-96.
3. Chan DS, Vieira AR, Aune D, Bandera EV, Greenwood DC, McTiernan A, et al. Body mass index and survival in women with breast cancer-systematic literature review and meta-analysis of 82 follow-up studies. Ann Oncol. 2014;25(10):1901-14.
4. Pan Hongchao GRG, and on behalf of the Early Breast Cancer Trialists' Collaborative Group. Effect of obesity in premenopausal ER+ early breast cancer: EBCTCG data on 80,000 patients in 70 trials. Journal of Clinical Oncology 2014 32 503-.
5. Blair CK, Wiggins CL, Nibbe AM, Storlie CB, Prossnitz ER, Royce M, et al. Obesity and survival among a cohort of breast cancer patients is partially mediated by tumor characteristics. NPJ Breast Cancer. 2019;5:33.
6. Lee K, Kruper L, Dieli-Conwright CM, Mortimer JE. The Impact of Obesity on Breast Cancer Diagnosis and Treatment. Curr Oncol Rep. 2019;21(5).
7. Silberg Jordan NK, Larose Meredith, Wright Christopher, Simone Nicole Lynn. Effect of elevated BMI on radiation toxicity in early stage breast cancer patients. Journal of Clinical Oncology. 2016;32.
8. Dillekås H, Rogers MS, Straume O. Are 90% of deaths from cancer caused by metastases? Cancer Med. 2019;8(12):5574-6.
9. Afifi AM, Saad AM, Al-Husseini MJ, Elmehrath AO, Northfelt DW, Sonbol MB. Causes of death after breast cancer diagnosis: A US population-based analysis. Cancer. 2019.
10. Blücher C, Stadler SC. Obesity and Breast Cancer: Current Insights on the Role of Fatty Acids and Lipid Metabolism in Promoting Breast Cancer Growth and Progression. Front Endocrinol (Lausanne). 2017;8:293.
11. Munir R, Lisec J, Swinnen JV, Zaidi N. Lipid metabolism in cancer cells under metabolic stress. Br J Cancer. 2019;120(12):1090-8.
12. Yan F, Zhao H, Zeng Y. Lipidomics: a promising cancer biomarker. Clin Transl Med. 2018;7(1):21.
13. Wu Q, Li B, Li Z, Li J, Sun S. Cancer-associated adipocytes: key players in breast cancer progression. J Hematol Oncol. 2019;12(1):95.
14. Chu DT, Phuong TNT, Tien NLB, Tran DK, Nguyen TT, Thanh VV, et al. The Effects of Adipocytes on the Regulation of Breast Cancer in the Tumor Microenvironment: An Update. Cells. 2019;8(8).
15. Hoy AJ, Balaban S, Saunders DN. Adipocyte-Tumor Cell Metabolic Crosstalk in Breast Cancer. Trends Mol Med. 2017;23(5):381-92.
16. Lee Isla Crake R, Phillips E, Kleffmann T, Currie MJ. Co-culture With Human Breast Adipocytes Differentially Regulates Protein Abundance in Breast Cancer Cells. Cancer Genomics Proteomics. 2019;16(5):319-32.
17. Röhrborn D, Wronkowitz N, Eckel J. DPP4 in Diabetes. Front Immunol. 2015;6:386.
18. Sell H, Blüher M, Klöting N, Schlich R, Willems M, Ruppe F, et al. Adipose dipeptidyl peptidase-4 and obesity: correlation with insulin resistance and depot-specific release from adipose tissue in vivo and in vitro. Diabetes Care. 2013;36(12):4083-90.
19. Yang F, Takagaki Y, Yoshitomi Y, Ikeda T, Li J, Kitada M, et al. Inhibition of Dipeptidyl Peptidase-4 Accelerates Epithelial-Mesenchymal Transition and Breast Cancer Metastasis via the CXCL12/CXCR4/mTOR Axis. Cancer Res. 2019;79(4):735-46.
20. Pujadas G, De Nigris V, Prattichizzo F, La Sala L, Testa R, Ceriello A. The dipeptidyl peptidase-4 (DPP-4) inhibitor teneligliptin functions as antioxidant on human endothelial cells exposed to chronic hyperglycemia and metabolic high-glucose memory. Endocrine. 2017;56(3):509-20.
21. Moro K, Kawaguchi T, Tsuchida J, Gabriel E, Qi Q, Yan L, et al. Ceramide species are elevated in human breast cancer and are associated with less aggressiveness. Oncotarget. 2018;9(28):19874-90.

SIGNATURES

Award Recipient full name	
Dr Mariela Vasileva-Slaveva	23.10.2020

Research Mentor full name	Signature and Date
Prof. Ira-Ida Skvortsova	

This ESMO Translational Fellowship Research Project was supported by an educational grant from ESMO