

ESMO Research Research Fellowship
(November 2021– November 2022)

Gaia Giannone
FINAL REPORT

Host Institute: **Imperial College London**

Mentor: **Prof. I A. McNeish**

Project title: **Predictive biomarkers for response to nintedanib in ovarian clear cell carcinoma**

Home Institute: **Candiolo Cancer Institute**

Introduction

Ovarian clear cell carcinoma (OCCC) is a rare ovarian cancer subtype (approximately 10% of cases in Caucasian women but more frequent in South-East Asia). It is frequently diagnosed at an early stage with a good prognosis in these cases. However, advanced and recurrent disease has a poor outcome, with intrinsic platinum resistance and no effective therapeutic tools (1).

OCCC has few recurrent mutations (the most frequent are *ARID1A*, which is part of the SWI/SNF chromatin remodeling complex, and *PIK3CA*, with activation of PI3K/AKT pathway), while mismatch repair deficiency has been recorded in 2-20% cases (2-5).

Gene expression profiles show some similarities with renal clear cell carcinomas with an upregulation of IL6-STAT3-HIF signalling (6,7).

In a retrospectively collected cohort of 222 OCCC, two gene expression patterns were identified: a good prognosis epithelial group, with high expression of cell-cell adhesion genes and higher rates of SWI/SNF mutations (EpiCC), and an aggressive, poor prognosis mesenchymal subtype (MesCC), with high prevalence of epithelial to mesenchymal transition (EMT) and enrichment in genes involved in extracellular matrix (ECM) organization as well as immune-related genes (8).

Data from a clear cell renal cohort suggest that the MesCC subtype respond better to anti-angiogenic drugs (8).

Rationale and Aim

My hypothesis is that OCCC patients with different gene expression profiles benefit from specific targeted drugs, and our original aim was to study the predictive role of MesCC in patients treated with anti-angiogenic agents. We used samples from the NiCCC trial, a randomised phase II study which evaluated single agent nintedanib in a population of recurrent, platinum-resistant OCCC patients (9). Overall, NiCCC showed no significant benefit from nintedanib treatment compared to standard of care in the overall study population (9). However, I hypothesised that nintedanib might be superior in patients whose tumours demonstrate MesCC gene expression.

Experimental design

Primary hypothesis: nintedanib has significantly greater effect in OCCC that are classified as MesCC and/or lack mutations in *ARID1A*, which are more frequent in the EpiCC subtype.

- Aim 1: To evaluate gene expression profiles in NiCCC samples and correlate gene expression classification (MesCC vs EpiCC) with clinical outcomes.

- Aim 2: To assess somatic single nucleotide variants/short variants in key OCCC genes and correlate with outcomes in the two arms.

Exploratory hypothesis: we will search for and identify markers of poor prognosis at the time of diagnosis and potential therapeutic targets (with a focus on angiogenesis and immune response) which can be exploited in future studies (Aim 3).

Results, Conclusions and Future Perspectives

During the first months of this fellowship, we signed an MTA and DTA with University of Glasgow and we applied for a grant to NIHR Imperial Biomedical Research Centre (BRC) with the aim of covering the costs of RNA sequencing and staining for NiCCC samples (Lead applicant: Prof McNeish, Co-applicant: Dr. Giannone and Dr. McDermott). The application was successful and supported all the consumables from this project.

Once the samples were received, we performed a quality check and RNA was extracted and sequenced. In addition, we stained the slides for ARID1A.

Analyses are ongoing. I am actively analysing RNA results starting from raw data, after being enrolled in a course on bioinformatics held by Wellcome Sanger institute and with the day to day supervision of our senior bioinformatician Mr Hasan Mirza. Moreover, a digital pathology approach has been explored for ARID1A staining using QuPath.

Clinical data have been studied to evaluate the prognostic role of C reactive protein (CRP) and platelet counts (both at baseline and during treatment), in these patients. Our plan is to submit an abstract for the ASCO Meeting 2023 with our findings.

Lastly, we received support from our institution to perform spatial transcriptomic analysis using GeoMx on a pilot set of samples. We will compare some representative FFPE from our cohort with tumours collected using a retrospective set from NHS (patients receiving surgery at Hammersmith Campus outside NiCCC trial). These analyses are planned for beginning of 2023.

Due to COVID 19 disruption, the number of available samples was lower than expected and it did not allow to plan any inference according to the predictive value of gene expression signature focusing only on this study. For this reason, we decided to review the design of our project, planning on evaluating the role of these two categories of OCCC (MesCC and EpiCC) in an enlarged cohort, treated not only with antiangiogenetic agents but also with immune checkpoint inhibitors (ICI) or standard chemotherapy.

We held virtual and in person meetings with Prof. David Tan (Cancer Science Institute of Singapore) and Dr. Rebecca Kristeleit (Guy's and St Thomas Hospital, London, UK) in which we explored the possibility to merge our cohorts from different trials and we have strengthened our collaboration with their centres. Indeed, they led two phase II studies which enrolled patients with recurrent OCCC treated with ICI and have planned a strong translational project related to their trials (10,11).

We will continue our analyses with the support of Prof Tan's team in Singapore and will probably validate our findings in their cohorts thereafter.

List of Publications and Presentations Resulting from the Translational Research Project "Predictive biomarkers for response to nintedanib in ovarian clear cell carcinoma"

Abstract in preparation for ASCO 2023

List of Publications and Presentations resulting from other projects during the fellowship period (if applicable)

Selected Poster Presentations:

- Ergasti R, Lim MQ, Giannone G, Ennis DP, Dye ICA, Mirza HB, Fagotti A, Scambia G, McNeish IA. Physiologically relevant treatment models to investigate epigenetic mechanisms driving platinum resistance in ovarian High Grade Serous Carcinoma. ESGO 2022

- Targeting PI3K/AKT/mTOR pathway in platinum-resistant ovarian highgrade serous carcinoma: Translational analysis from the randomized phase II OCTOPUS Trial. Giannone G, Ennis D, Mirza HB, Cheng Z, McDermott J, Lewsley L-A, Clamp A, Herbertson R, Glasspool R, Krell J, Hinsley S, Banerji U, Riisnaes R, Banerjee S, McNeish I. ESMO Gynaecological Cancers Congress 2022

Published papers:

- Giannone G, Giuliano AR, Bandini M, Marandino L, Raggi D, Earle W, Ayres B, Pettaway CA, McNeish IA, Spiess PE, Necchi A. HPV vaccination and HPV-related malignancies: impact, strategies and optimizations toward global immunization coverage. *Cancer Treat Rev.* 2022 Dec;111:102467. doi: 10.1016/j.ctrv.2022.102467. Epub 2022 Oct 5. PMID: 36265336.
- Musacchio L, Valsecchi AA, Salutari V, Valabrega G, Camarda F, Tuninetti V, Giannone G, Bartoletti M, Marchetti C, Pignata S, Fagotti A, Scambia G, Di Maio M, Lorusso D. MEK inhibitor as single agent in low grade serous ovarian and peritoneal cancer: a systematic review and meta-analysis. *Cancer Treat Rev.* 2022 Nov;110:102458. doi: 10.1016/j.ctrv.2022.102458. Epub 2022 Aug 24. PMID: 36063572.
- Bartoletti M, Bergamini A, Giannone G, Nero C, Musacchio L, Farolfi A, Passarelli A, Kuhn E, Castaldo D, Lombardo V, Di Palma T, Lorusso D, Puglisi F, De Giorgi U, Valabrega G, Schettino C, Scambia G, Capoluongo E, Pignata S. A fully virtual and nationwide molecular tumor board for gynecologic cancer patients: the virtual experience of the MITO cooperative group. *Int J Gynecol Cancer.* 2022 Mar 21;ijgc-2022-003425. doi: 10.1136/ijgc-2022-003425. Epub ahead of print. PMID: 35314461.
- Giannone G, Castaldo D, Tuninetti V, Scotto G, Turinetto M, Valsecchi AA, Bartoletti M, Mammoliti S, Artioli G, Mangili G, Salutari V, Lorusso D, Cormio G, Zamagni C, Savarese A, Di Maio M, Ronzino G, Pisano C, Pignata S, Valabrega G. Management of Metastatic Endometrial Cancer: Physicians' Choices Beyond the First Line. A MITO Survey. *Front Oncol.* 2022 May 27;12:880008. doi: 10.3389/fonc.2022.880008. PMID: 35692798; PMCID: PMC9185800.
- Cheng Z, Mirza H, Ennis DP, Smith P, Morrill Gavarró L, Sokota C, Giannone G, Goranova T, Bradley T, Piskorz A, Lockley M; BriTROC-1 Investigators, Kaur B, Singh N, Tookman LA, Krell J, McDermott J, Macintyre G, Markowitz F, Brenton JD, McNeish IA. The Genomic Landscape of Early-Stage Ovarian High-Grade Serous Carcinoma. *Clin Cancer Res.* 2022 Jul 1;28(13):2911-2922. doi: 10.1158/1078-0432.CCR-21-1643. PMID: 35398881; PMCID: PMC7612959.

Papers under review:

- Banerjee S, Giannone G, Clamp A, Ennis D, Glasspool R, Herbertson R, Krell J, Riisnaes R, Mirza HB, Cheng Z, McDermott J, Green C, Kristeleit RS, George A, Gourley C, Lewsley L-A, Rai D, Banerji U, Hinsley S, McNeish IA. A Randomised, Multi-centre Phase II Trial of Weekly Paclitaxel and Vistusertib in Platinum-Resistant Ovarian High-Grade Serous Carcinoma: OCTOPUS Arm 1. *Medrxiv* doi: <https://doi.org/10.1101/2022.05.26.22275618>
- Smith P-S, Bradley T, Morrill Gavarró L, Goranova T, Ennis D, Mirza H, De Silva D, Piskorz A, Al-Khalidi S, Sauer C, Funingana I-G, Reinius M, Giannone G, Lewsley L-A, Stobo J, McQueen J, Bryson G, Eldridge M, The BriTROC Investigators, Macintyre G, Markowitz F, Brenton JD, McNeish IA. The copy-number landscape of recurrent ovarian high grade serous carcinoma: the BriTROC-1 study. *Medrxiv* doi: <https://doi.org/10.1101/2022.10.21.22280992>

Selection of Courses and Workshops Attended During the Fellowship

Wellcome Connecting Science-Next Generation Sequencing Bioinformatics, Wellcome Trust Genome Campus.

Data Science: R Programming course, Imperial College London.

Data Science: Data Processing with R, Imperial College London.

Acknowledgements

Prof I McNeish, for his continuous and strenuous support, for dedicating so much time to mentor me and for providing me with a stimulating environment and endless chances to learn, grow and understand how to move on in my career path.

Dr Darren Ennis for being an amazing supervisor and for teaching me so much every day; the whole McNeish lab, which puts together incredible researchers and wonderful people, because it is always a great pleasure to work within such a smart and nice team.

Imperial College and Ovarian Cancer Action Research Centre, for hosting me and making me feel welcome every day.

Prof Aglietta and Valabrega for encouraging my ambitions and my interest in translational research and for believing in the role of networking in Oncology. Prof Pignata, for setting an example of how to do research in a collaborative group and for nurturing the next generation of oncologists.

ESMO for being an inspiring network and an incessant source of opportunities for young clinicians; for providing outstanding mentors and shaping the future of oncology, with the aim of providing better care for our patients.

All the patients and their families, for supporting research and actively helping us understand the real unmet needs and how to improve diagnosis and treatment.

Personal Statement

I am extremely grateful to ESMO for this lifechanging fellowship. It marked a crucial moment of my career path as a young oncologist and helped me understand what translational research is and the fundamental role it plays in oncology. Being actively involved in a translational project, with both successes and setbacks, gave me a taste of what being a clinician scientist means and how complex and at the same time rewarding this job can be.

This experience will profoundly influence my career choices and change my perspective on how to be an oncologist nowadays. Indeed, I am currently enrolled in a 3-years translational project at Imperial, hoping to gain more and more experience in this field.

In summary, to all my young colleagues I would say: take a leap and apply for the ESMO translational fellowship, it will enrich your personal and professional life.

I would be more than happy to give feedback to anyone who needs more information (twitter account @gaia_giannone)

References

1. Chan JK, Teoh D, Hu JM, Shin JY, Osann K, Kapp DS. Do clear cell ovarian carcinomas have poorer prognosis compared to other epithelial cell types? A study of 1411 clear cell ovarian cancers. *Gynecologic Oncology*. 2008;109(3):370-6.
2. Kuo K-T, Mao T-L, Jones S, Veras E, Ayhan A, Wang T-L, et al. Frequent Activating Mutations of PIK3CA in Ovarian Clear Cell Carcinoma. *The American Journal of Pathology*. 2009;174(5):1597-601.
3. Jones S, Wang T-L, Shih I-M, Mao T-L, Nakayama K, Roden R, et al. Frequent Mutations of Chromatin Remodeling Gene ARID1A in Ovarian Clear Cell Carcinoma. *Science*. 2010;330(6001):228-31.
4. Uehara Y, Oda K, Ikeda Y, Koso T, Tsuji S, Yamamoto S, et al. Integrated Copy Number and Expression Analysis Identifies Profiles of Whole-Arm Chromosomal Alterations and Subgroups with Favorable Outcome in Ovarian Clear Cell Carcinomas. *PLOS ONE*. 2015;10(6):e0128066.

5. Morikawa A, Hayashi T, Kobayashi M, Kato Y, Shirahige K, Itoh T, et al. Somatic copy number alterations have prognostic impact in patients with ovarian clear cell carcinoma. *Oncology Reports*. 2018.
6. Yamaguchi K, Mandai M, Oura T, Matsumura N, Hamanishi J, Baba T, et al. Identification of an ovarian clear cell carcinoma gene signature that reflects inherent disease biology and the carcinogenic processes. *Oncogene*. 2010;29(12):1741-52.
7. Coward J, Kulbe H, Chakravarty P, Leader D, Vassileva V, Leinster DA, et al. Interleukin-6 as a Therapeutic Target in Human Ovarian Cancer. *Clinical Cancer Research*. 2011;17(18):6083-96.
8. Tan TZ, Ye J, Yee CV, Lim D, Ngoi NYL, Tan DSP, Huang RY. Analysis of gene expression signatures identifies prognostic and functionally distinct ovarian clear cell carcinoma subtypes. *EBioMedicine*. 2019 Dec;50:203-210.
9. Glasspool R, Mcneish I, Westermann A, Hinsley S, Ledermann J, Ray-Coquard I, et al., editors. 596 A randomised phase II study of nintedanib (BIBF1120) compared to chemotherapy in patients with recurrent clear cell carcinoma of the ovary or endometrium. (NICCC/ENGOT-OV36). *Ovarian Cancer*; 2020 2020-12-01: BMJ Publishing Group Ltd.
10. Tan, D.S.P., Choi, C.H., Ngoi, N., Sun, H., Heong, V., Ow, S.G.W., Chay, W.Y., Kim, H.S., Lim, Y.W., Goss, G., Goh, J.C., Luo, V., Tai, B.C., Lim, D., Kaliaperumal, N., Au, V.B., Connolly, J., Kim, J.-W., Friedlander, M., Kim, K., 2022. A multicenter phase II randomized trial of durvalumab (D) versus physician's choice chemotherapy (PCC) in patients (pts) with recurrent ovarian clear cell adenocarcinoma (MOCCA/APGOT-OV2/GCGS-OV3).. *Journal of Clinical Oncology* 40, 5565–5565.
11. R. Kristeleit, A.R. Clamp, C. Gourley, R. Roux, M. Hall, M. Devlin, R. Nirsimloo, V. Kounnis, L. Hughes, N. Counsell, L. Farrelly, R. Miller. Efficacy of pembrolizumab monotherapy (PM) for advanced clear cell gynaecological cancer (CCGC): Phase II PEACOC trial. *Annals of Oncology* (2022) 33 (suppl_7): S235-S282. 10.1016/annonc/annonc1054

SIGNATURES

Award Recipient full name	Signature and Date
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Research Mentor full name	Signature and Date
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