

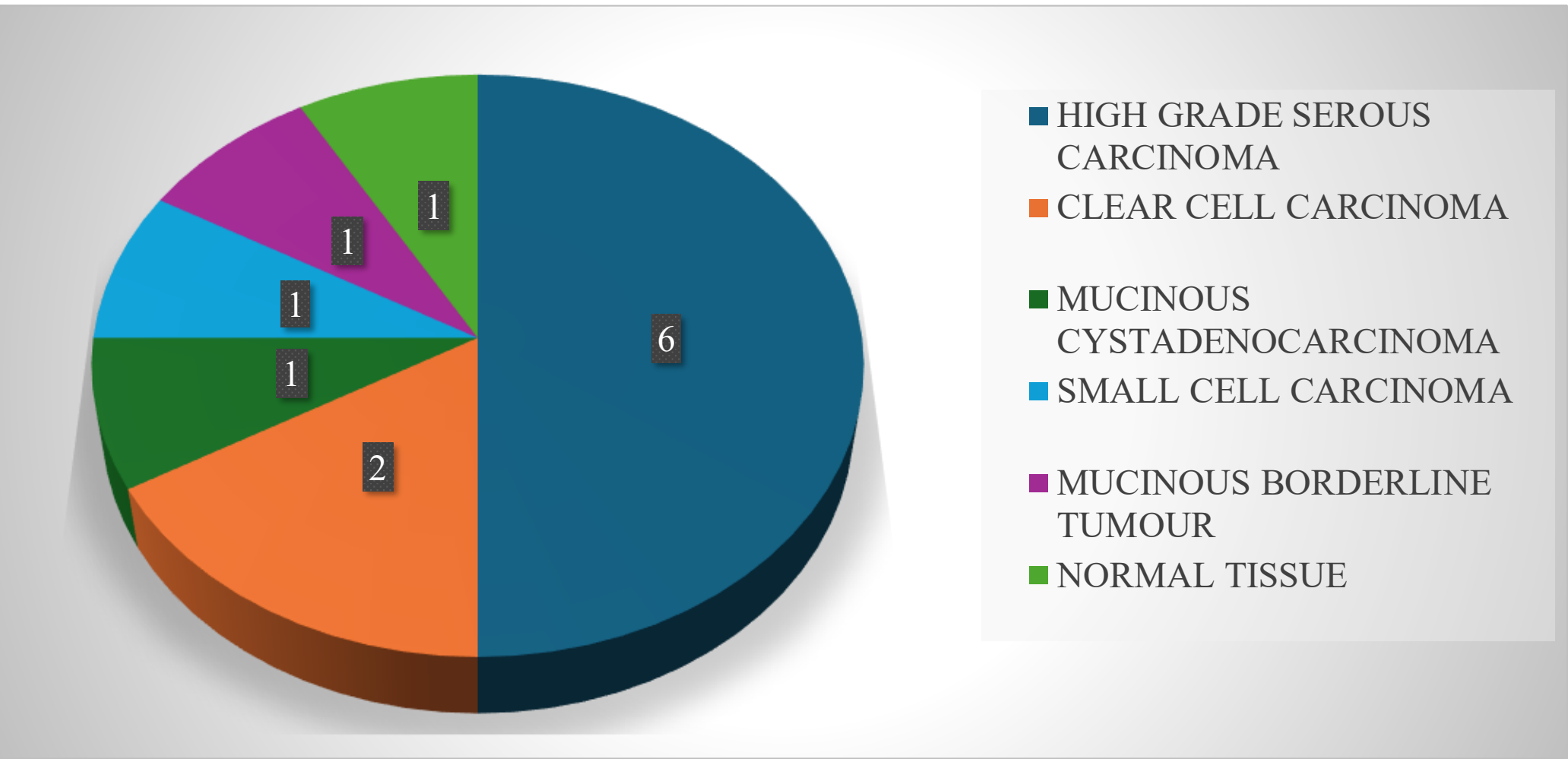
FROM SLIDE TO SPHEROID- STUDYING OVARIAN CANCER CHEMORESISTANCE USING TISSUE AND PATIENT DERIVED ORGANOID HISTOPATHOLOGY STAINING

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INTRODUCTION

- Ovarian cancer is a heterogenous disease , with differences in molecular expressions and genetic mutations¹.
- Primary chemotherapy resistance is a key challenge in management of ovarian cancer.
- Organoids are in –vitro 3D structures grown from stem cells , that self organize through lineage commitment; they recapitulate histological architecture and genetic alterations of original tumour².
- We aimed to generate patient derived ovarian cancer organoids to evaluate their potential as functional precision oncology models for high -throughput screening and treatment stratification.

RESULTS- Organoids retain histopathological fidelity to parent tumour



GRAPH 1 – Histopathological subtype distribution in our study.

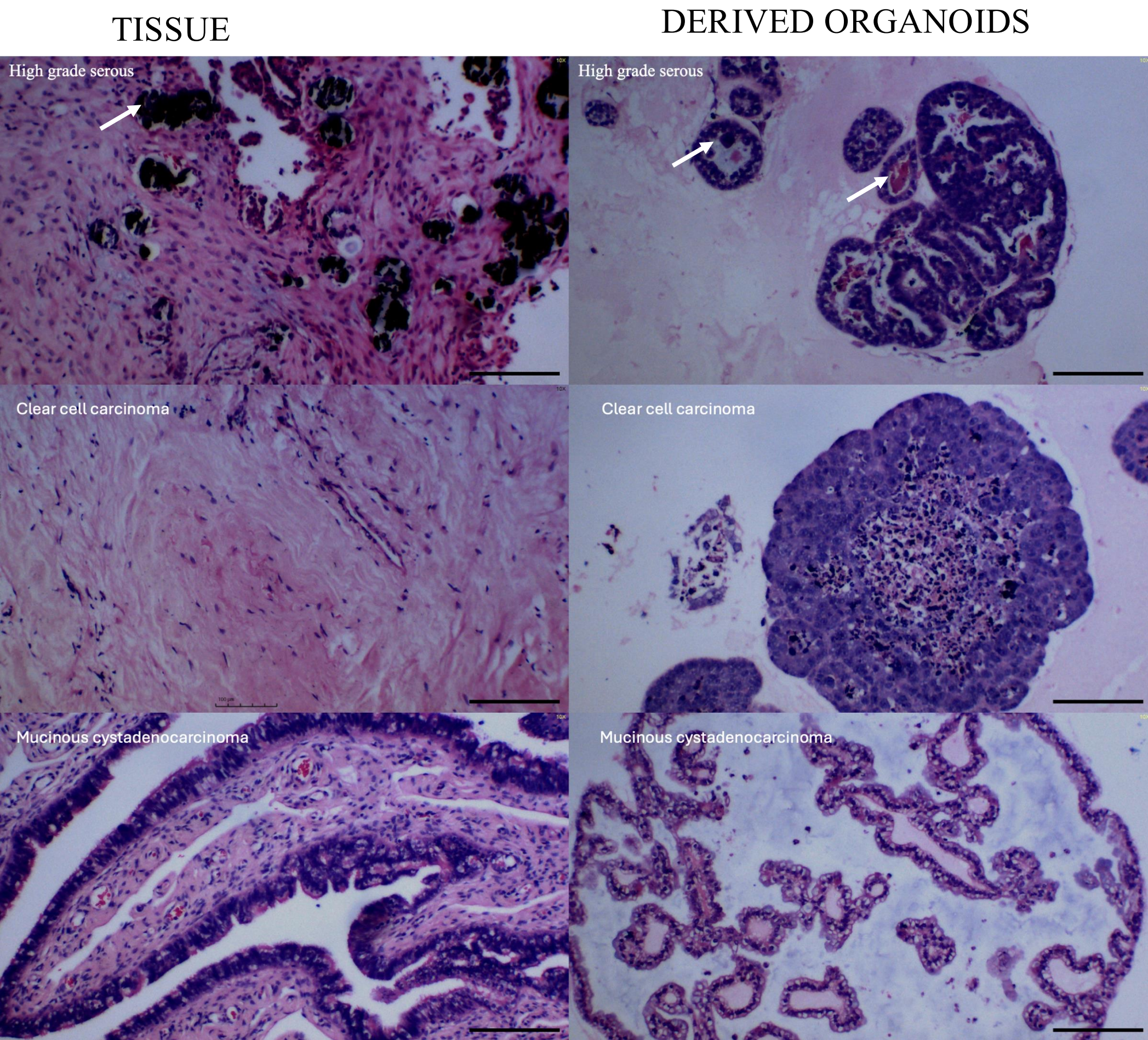


Figure 2 shows the brightfield microscopic images of H&E staining of the donor tumour tissue and the derived organoids from the respective tumour tissue. Three types of morphology noted in derived organoids- solid, cystic and glandular. The images were captured under 10X objective , with scale bar of 150 µm. White arrows represent Psammoma bodies.

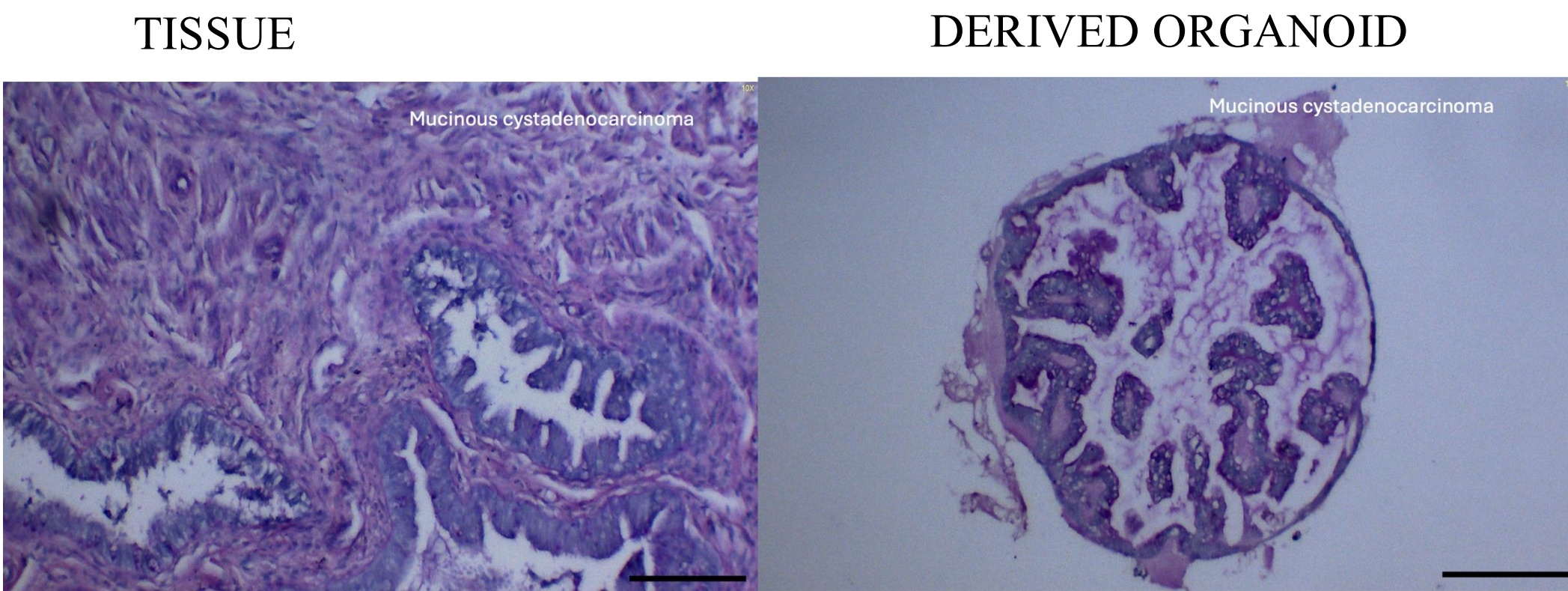


Figure 3 shows the PAS stain on tissue and the corresponding derived organoid in a donor with mucinous cystadenocarcinoma. (Tissue and organoid image = 10X objective, 150 µm scalebar). Organoid image shows scattered lakes of glycoprotein lined by columnar cells.

METHODS

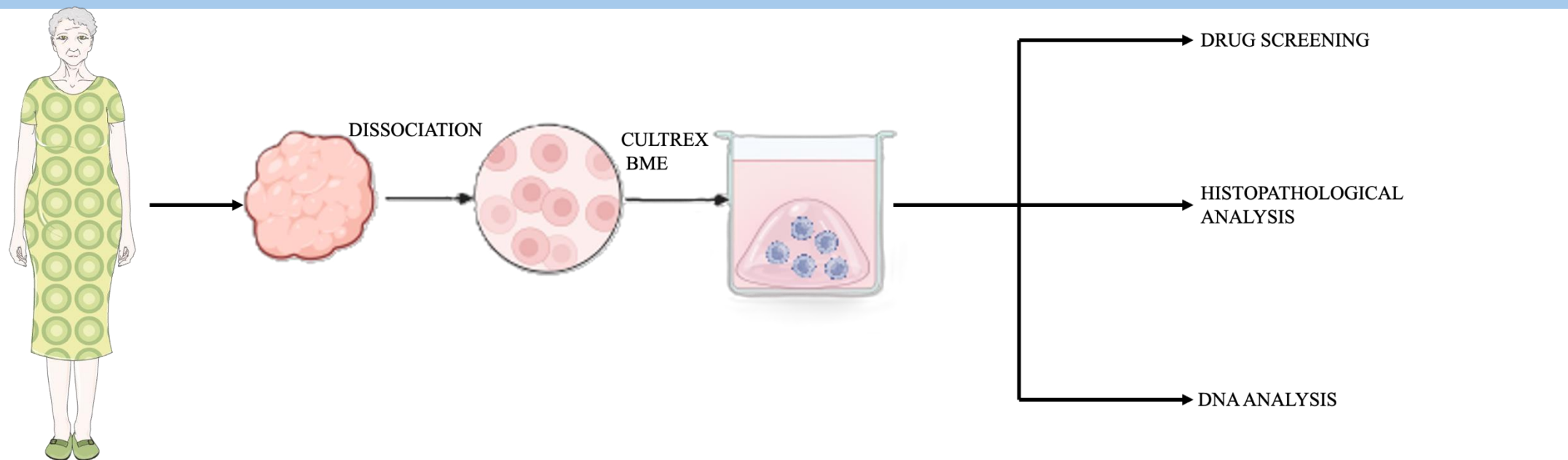


Figure 1 is a graphical representation of the methods involving organoids derivation and the workflow used for histopathological validation. Image generated using Biorender.com

RESULTS- Organoids represent reliable platform for IHC and IF analyses

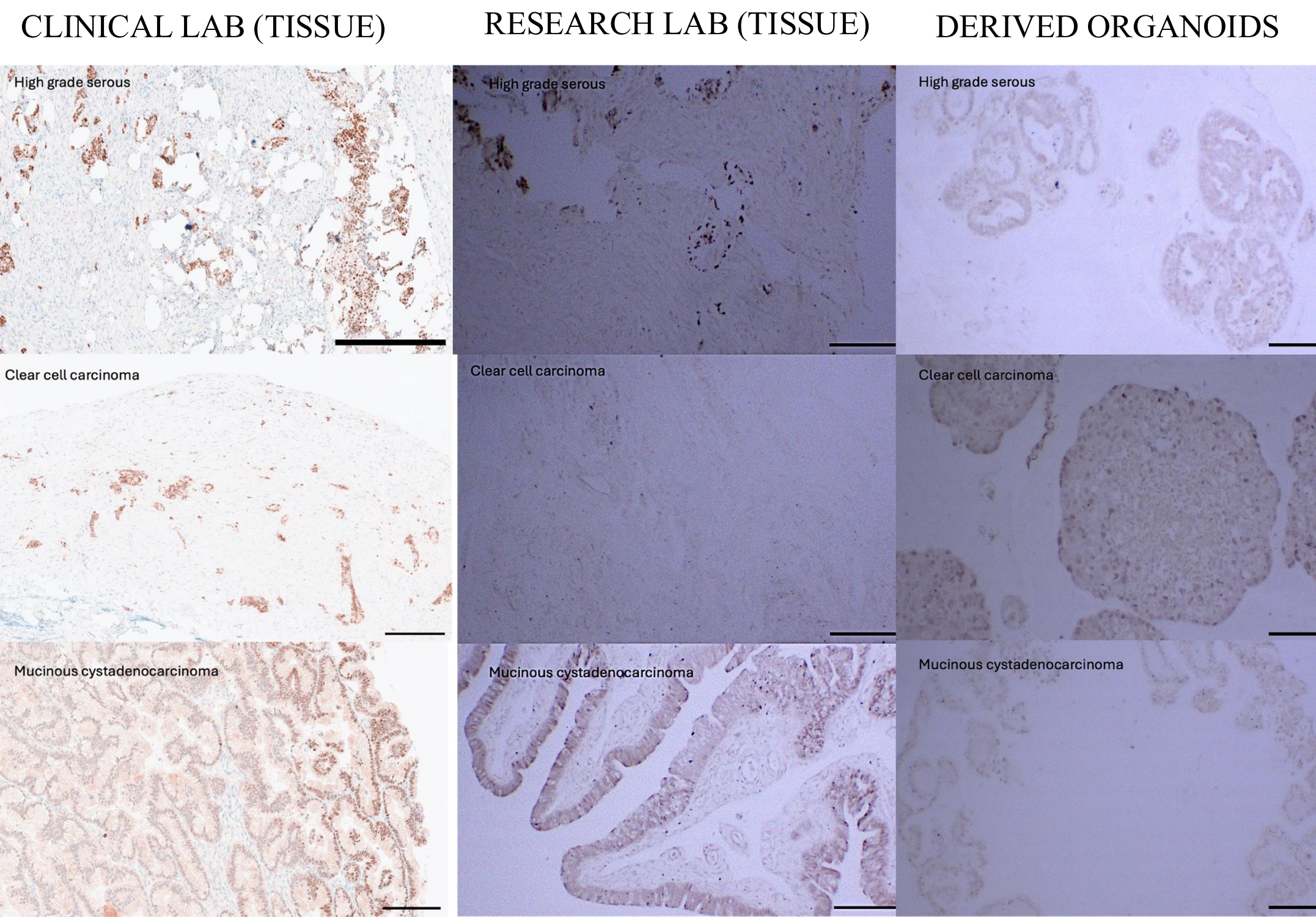


Figure 4 shows the brightfield microscopic images of PAX8 Immunohistochemistry stain of the samples. The images are from donor samples stained in clinical diagnostic lab, the tissue and organoids stained in research lab. All images- 10X objective, 150 µm scalebar.

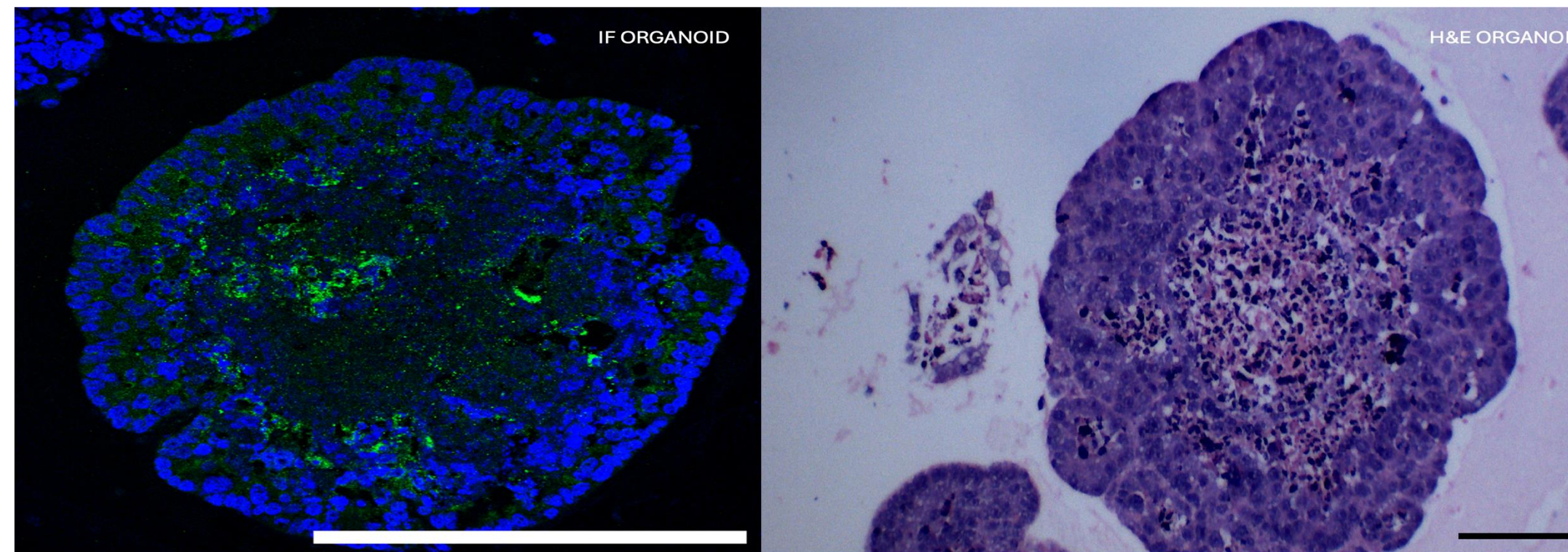


Figure 5 shows the immunofluorescence stain of CD44 and CD133 stem cell markers on organoid (clear cell carcinoma). IF image (20X objective) is an overlay of 3 channels (red, green and blue). (Red= CD44, Green=CD133, Blue= DAPI). The image shows predominantly CD133 signal with blue DAPI nuclear stain

RESULTS

- 12 donors were recruited to our study (GRAPH 1)
- Figure 2 shows the H&E brightfield images comparing tissue and organoids
- Figure 3 shows the PAS stain of fallopian tube and the derived organoid from a donor with mucinous cystadenocarcinoma. The organoid image shows preservation of secretory cells of the fallopian tube.
- Figure 4 shows the corresponding PAX8 IHC comparing images from tissue stained during clinical diagnostic analysis, tissue stained in the research lab and organoid stained in the research lab. Pre-diluted IHC PAX8 kit was used in research lab.
- Figure 5 shows the immunofluorescence image of organoid stained for CD44 and CD133 stemcell antibodies. (used to study chemoresistance)

CONCLUSIONS

- Patient derived ovarian cancer organoids faithfully recapitulate the histopathological architecture of the parent tumour.
- Patient derived ovarian cancer organoids are reliable platform to study key diagnostic cancer markers using IHC and IF.
- Patient derived ovarian cancer organoids can be used in research towards next generation precision (or personalized) medicine.