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Poster presentation

GLUCOSYLCERAMIDE SYNTHASE INHIBITION DELAYS THE ONSET OF ACQUIRED RESISTANCE IN NSCLC MODELS IN THE ERA OF OSIMERTINIB

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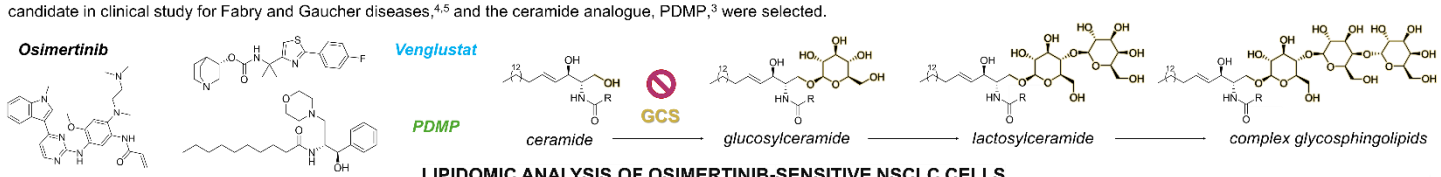
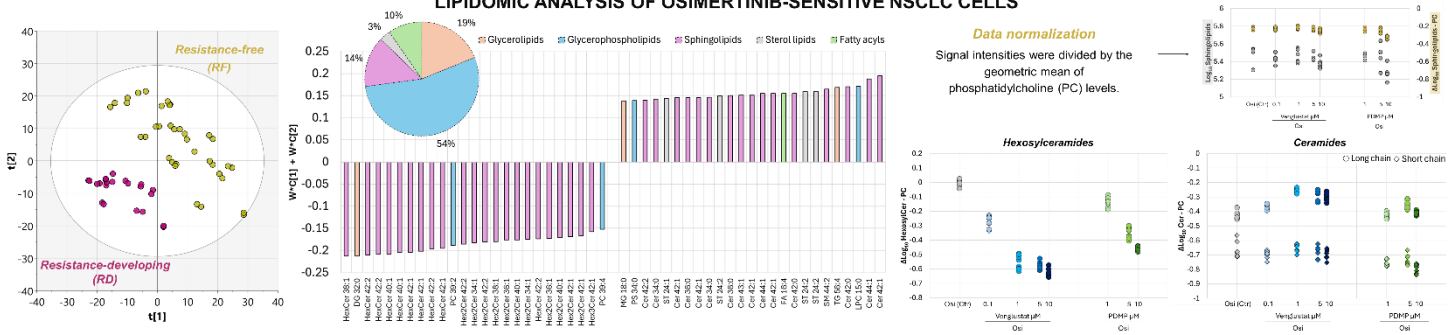
INTRODUCTION & AIM OF THE WORK

- Osimertinib (osi) represents the front-line treatment for non-small cell lung cancer (NSCLC) patients with activating mutations of EGFR. Despite its clinical efficacy, the occurrence of resistance phenomena urges the identification of novel strategies to fight osi-resistance in NSCLC.[1] While second-line treatments for osi-resistant NSCLC patients are still lacking, the progression-free survival (PFS) could be improved delaying the insurgence of resistance to osi treatment.
- Besides genetic mutations, the involvement of metabolism remodeling in the development and maintenance of drug resistance has been demonstrated.[2] Upregulation of ceramide glycosylation with overexpression of glucosylceramide synthase (GCS) has been recently proposed as a hallmark of metabolic reprogramming and cancer progression in osi-resistant clones.[3]
- Guided by lipidomic analysis, we evaluated the use of GCS inhibitors to delay the onset of acquired resistance to osi in NSCLC cells and in xenograft models. Two GCS inhibitors, venglustat, a drug candidate in clinical study for Fabry and Gaucher diseases,[4,5] and the ceramide analogue, PDMP,[3] were selected.

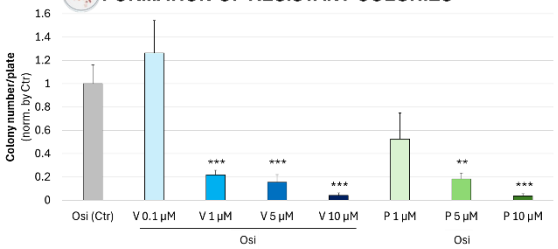


LIPIDOMIC ANALYSIS OF OSIMERTINIB-SENSITIVE NSCLC CELLS



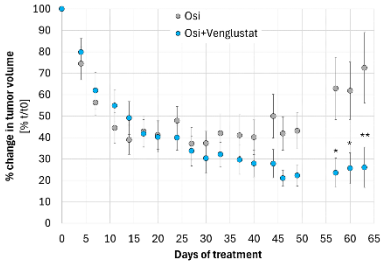
- Lipidomic analysis of PC9 cells treated with osi or with the association of osi (30 nM) and venglustat/PDMP at increasing concentrations was performed employing a UHPLC-HRMS system.
- PLS-DA on 800 lipid features reveals clear separation of ***RD*** and ***RF*** groups. Hexosylceramides and ceramides represent the top-discriminant features with opposite directions.
- Collective concentration-response curves of hexosylceramides and long-chain ceramides (≥ 40 carbon atoms) confirm the role of GCS inhibitors in preventing the insurgence of resistance.

FORMATION OF RESISTANT COLONIES



- Osi-sensitive NSCLC cells (PC9) develop long-term resistance to increasing concentrations of osi (30 nM – 500 nM). GCS inhibitors block the formation of resistant colonies.
- Venglustat is more potent that PDMP in inhibiting the development of resistance.

IN VIVO STUDY ON XENOGRAFT MODELS



- The combination of osi (3 mg/kg/day, p.o.) and venglustat (10 mg/kg/day, i.p.) to BALB/c-Nude female mice subcutaneously injected with PC9 cells avoids the long-term progression of tumor growth.

Time	Venglustat plasmatic conc.
3 h	2.35 µM ± 0.48
6 h	2.19 µM ± 0.14

- Plasmatic concentrations of venglustat at 3 and 6 hours are similar to those inhibiting hexosylceramide synthesis and formation of resistant colonies *in vitro*.

CONCLUSIONS

- Lipidomic analysis confirms the role of hexosylceramides as mediators of NSCLC cell progression to osi-resistance.
- *In vitro* and *in vivo* studies support the association of osi and venglustat as a promising strategy to prolong the PFS of EGFR-mutant NSCLC patients who have progressed to osi-resistance.

REFERENCES

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