

Poster Communication 14

## Targeting the Warburg effect in breast cancer with chiral derivatives of flavonoids

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### Abstract

**Background:** Triple-negative breast cancer (TNBC) is the breast cancer subtype with the worst prognosis due to a lack of expression of human epidermal growth factor receptor 2 or hormone receptors [1]. Furthermore, TNBC cells exhibit metabolic reprogramming, prioritizing glycolysis even when oxygen is available. This metabolic shift, known as the Warburg effect, not only enhances tumor resistance to cytotoxic agents but also fuels its invasive behavior [2]. Flavonoids are examples of bioactive compounds with demonstrated antitumor activity, and their conjugation with amino acids has been reported to increase their cytotoxic effects [3]. Our studies further demonstrate that chiral derivatives of flavonoids (CDFs) showed promising anticancer potential. **Objective:** This study aims to investigate the cytotoxicity and mechanistic profile of a specific enantiomeric pair of CDFs (TriCe-LTrp and TriCe-DTrp) on two TNBC cell lines (MDA-MB-231 and Hs578T) and a non-tumor control cell line (MCF-10A). **Methods:** Cell viability was assessed to determine the selectivity index of the selected CDFs. The metabolic profile, evaluated by quantifying glucose and lactate, and the induction of apoptosis, measured by Annexin V flow cytometry, were analyzed following treatment. Additionally, cell cycle distribution was characterized using propidium iodide flow cytometry to further evaluate the effects of the compounds on the tumor cell lines. **Results:** Both TriCe-LTrp and TriCe-DTrp exhibited low GI<sub>50</sub> values in MDA-MB-231 and Hs578T cell lines. TriCe-LTrp demonstrated high selectivity on both tumor lines, with high selectivity indices, whereas TriCe-DTrp showed no selectivity, highlighting the critical role of chirality. Regarding metabolism, TriCe-DTrp significantly decreased lactate production in both lines and glucose consumption in Hs578T cells. Conversely, TriCe-LTrp reduced glucose consumption in both cell lines, but only significantly in Hs578T cells, though its impact on lactate was not significant. Flow cytometry analysis using Annexin V and propidium iodide revealed that TriCe-LTrp significantly induced apoptosis in MDA-MB-231 cells, while similar trends for TriCe-LTrp in Hs578T and for TriCe-DTrp in both lines did not reach significance. **Conclusions:** These findings identify TriCe-LTrp as a highly selective and potent lead compound for triple-negative breast cancer therapy due to its dual ability to modulate tumor metabolism and induce apoptosis.

**Keywords:** breast cancer; chiral flavones; antitumor activity

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## References

1. Li, Y. et al. Recent advances in therapeutic strategies for triple-negative breast cancer. *J Hematol Oncol* **2022**, *15*, 121, doi:10.1186/s13045-022-01341-0.
2. Liu, S. et al. Correlation between the Warburg effect and progression of triple-negative breast cancer. *Front Oncol* **2023**, *12*, 1060495, doi:10.3389/fonc.2022.1060495.
3. Pinto, C. et al. Chiral flavonoids as antitumor agents. *Pharmaceuticals* **2021**, *14*, 1267, doi:10.3390/ph14121267.



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