The marine-derived antibiotic chromomycin A5 targets the oncogenic TBX2: Anew strategy to treat breast cancer

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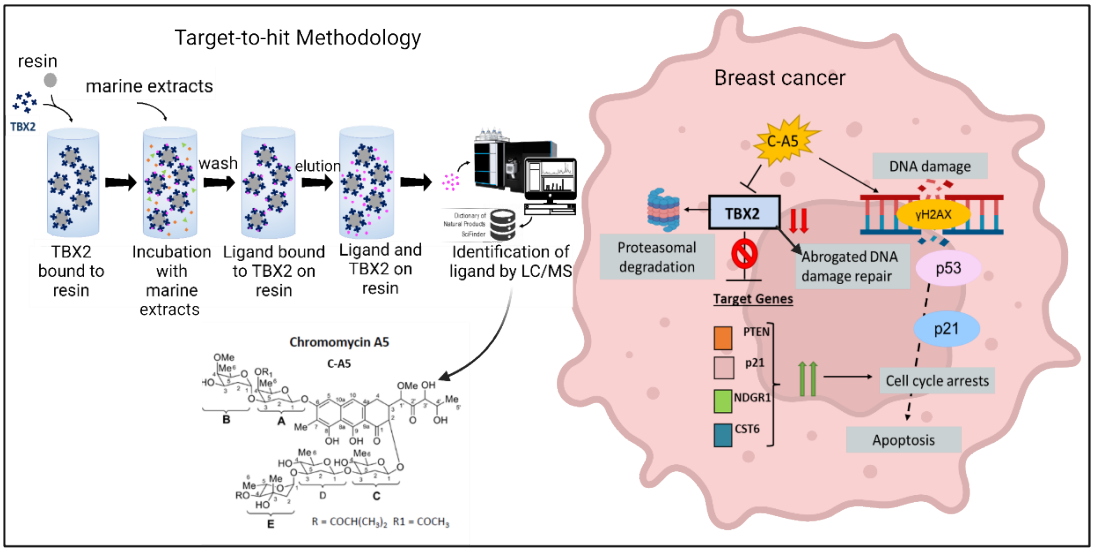

Graphical Abstract: *Identification of Chromomycin A5 as a TBX2-targeting agent with anti-breast cancer activity.*

Introduction: Breast cancer (BC) is the most common cancer and the leading cause of cancer-related death in women globally[1]. The transcription factor, TBX2, is commonly over-expressed in BC where it drives proliferation and promotes DNA damage repair to suppress cell death and confer drug resistance[2]. TBX2, although important during mammary gland development, plays no known function in adult breast tissue and is therefore an attractive anti-BC target. We therefore screened marine natural compounds for TBX2-binding affinity and identified Chromomycin A5 (C-A5), an aureolic acid producedby actinobacteria *Streptomyces*, as having a strong affinity for TBX2[3]. This project investigates C-A5 as a TBX2-targeting anti-cancer compound in BC.

Methodology: The short- and long-term cytotoxicity of C-A5 were assessed in MCF-7 and T47D ER+BC, and the normal breast epithelium MCF12a cell lines by MTT, clonogenic assays and spheroid analyses. The ability of C-A5 to target TBX2 for degradation via the proteasome-26s pathway was assessed using the MG132 inhibitor and western blotting, and TBX2 target genes assessed by qRT- PCR. We next looked at the ability of C-A5 to induce DNA damage, cell cycle arrests and apoptosis bywestern blotting, immunofluorescence, and flow cytometry.

Results: C-A5 was shown to exhibit potent and cytotoxicity in BC cells and to inhibit BC spheroid growth. Mechanistically, C-A5 targeted TBX2 for degradation through the proteasome 26s pathway, resulting in the de-repression of key BC tumour suppressor genes ordinarily repressed by TBX2. TBX2 knockdown and over-expression studies revealed C-A5 cytotoxicity was dependent on the availability of TBX2. Importantly, the inhibition of TBX2 by C-A5 corresponded with DNA damage, cell cycle arrests, and apoptosis in BC cells.

Discussion and conclusion: Here we demonstrate that C-A5 exhibits anti-breast cancer activity whichoccurs, in part, through its ability to degrade TBX2

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