Beyond Platinum: Tricarbonyl Rhenium(I) Complexes with Iminoquinolines as a Novel Strategy in Colorectal Cancer Therapy

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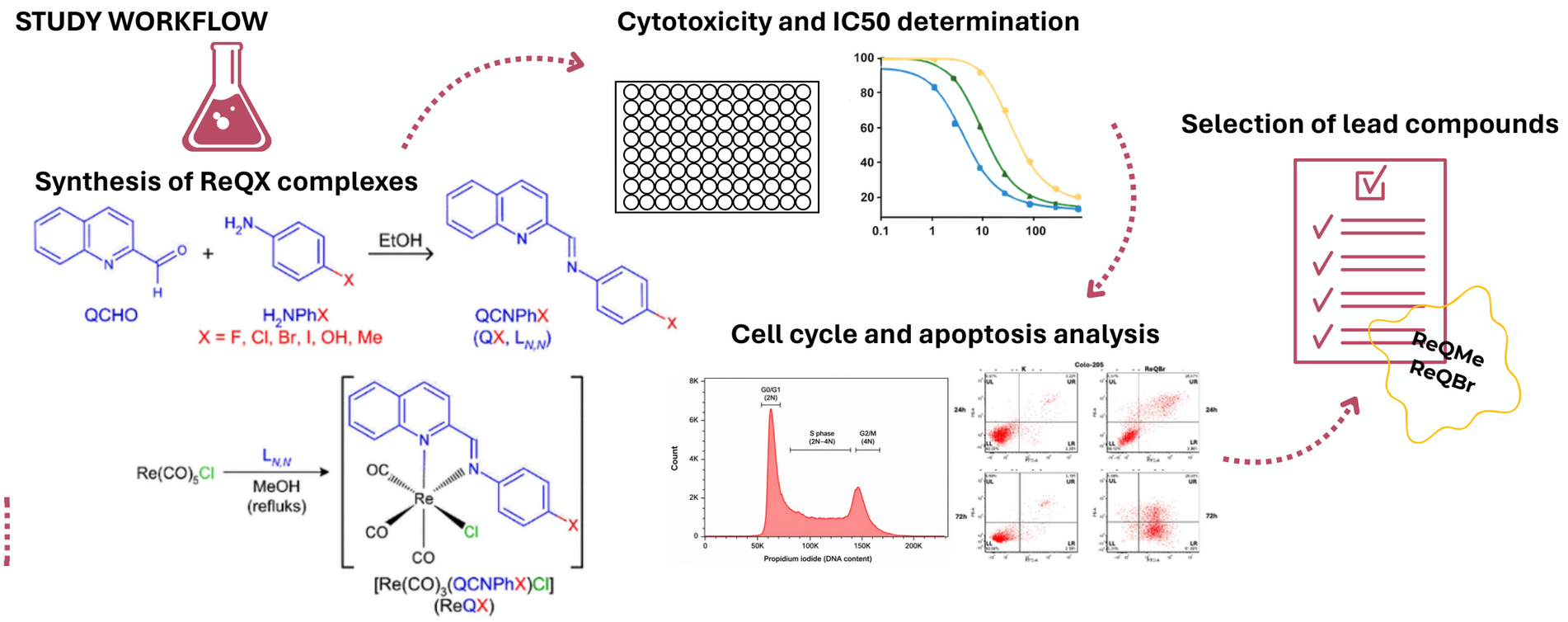
INTRODUCTION

Colorectal cancer (CRC) continues to present significant therapeutic challenges, primarily due to treatment resistance and tumour heterogeneity. Rhenium(I) tricarbonyl complexes have emerged as promising non-platinum metallodrugs with potential anticancer activity.

The present study was conducted with the objective of evaluating the biological effects of a novel series of iminoquinoline-based Re(I) complexes in colorectal cancer cell lines, and of identifying the most promising candidates for further mechanistic investigations.

METHODS



RESULTS

CYTOTOXICITY SCREENING

The antiproliferative activity of the investigated compounds was evaluated in three human colorectal cancer cell lines **(HT-29, SW48, and Colo-205)** after 24, 48, and 72 h of incubation at concentrations ranging from 2 to 50 μM. In contrast to the free ligands, which did not exhibit detectable cytotoxic activity under the applied experimental conditions, the corresponding rhenium(I) complexes demonstrated pronounced antiproliferative effects, indicating that coordination of the used ligands to the Re(I) center is essential for biological activity. **Moreover, the observed response was strongly concentration-, time-, and cell line-dependent, suggesting a selective biological mechanism rather than nonspecific cytotoxicity.**

IC_{50} values of the investigated Re(I) complexes against colorectal cancer cell lines after 48 h of incubation.

	HT-29	SW48	Colo-205
ReQBr	3.7μM	4.3μM	2.0μM
ReQI	6.7μM	5.1μM	6.5μM
ReQF	18.6μM	19μM	20.4μM
ReQMe	4.3μM	5.1μM	11.1μM

Dose–response analysis using a four-parameter nonlinear regression model showed that three of four complexes had IC50 values below 10 μM after 48 hours, indicating strong potency against CRC cells. **ReQBr and ReQMe were most effective across cell lines. The results suggest small changes in ligand structure significantly impact activity and highlight these rhenium(I) compounds as promising candidates for further study.**

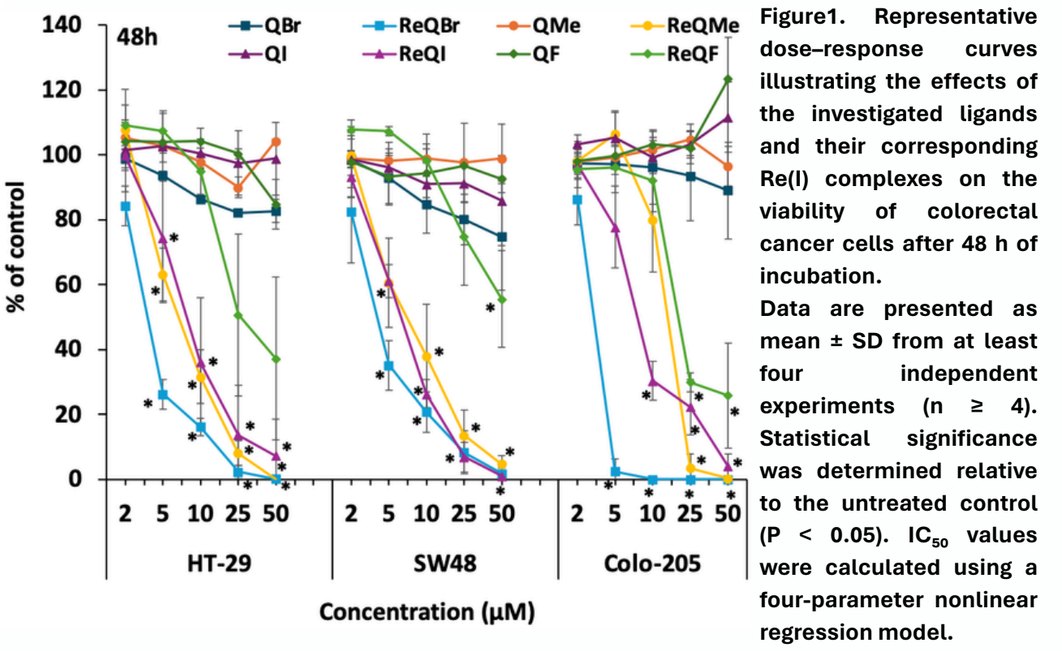


Figure1. Representative dose–response curves illustrating the effects of the investigated ligands and their corresponding Re(I) complexes on the viability of colorectal cancer cells after 48 h of incubation.
Data are presented as mean ± SD from at least four independent experiments (n ≥ 4). Statistical significance was determined relative to the untreated control ($P < 0.05$). IC_{50} values were calculated using a four-parameter nonlinear regression model.

CELL CYCLE ANALYSIS AND APOPTOSIS INDUCTION

To determine whether the observed reduction in cell viability resulted from cell cycle perturbation and activation of programmed cell death pathways, cell cycle distribution and apoptosis were analyzed by flow cytometry using propidium iodide (PI) staining and Annexin V-FITC/PI double staining, respectively.

The obtained results demonstrated that exposure of CRC cells to the investigated Re(I) complexes led to marked disturbances in cell cycle progression, indicating a pronounced antiproliferative effect. Concomitantly, distinct morphological alterations were observed, particularly following treatment with ReQBr and ReQMe. Treated cells with time lost their characteristic epithelial morphology, detached from the culture surface, and acquired a rounded and shrunken phenotype, consistent with profound cellular stress and loss of proliferative capacity (Fig. 2).

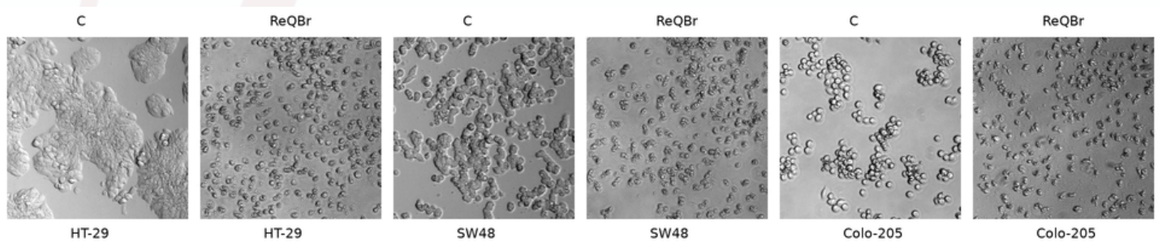


Figure 2. Representative phase-contrast micrographs showing morphological alterations induced by the investigated ReQBr complex in CRC cell lines after 48 h of treatment. Images were acquired using a 10× objective.

Cell cycle analysis further revealed a significant accumulation of cells within the sub-G1 fraction, a hallmark of apoptotic DNA fragmentation. This observation was subsequently confirmed by Annexin V-FITC/PI staining, demonstrating that treatment with the investigated Re(I) complexes (10 μM) effectively induced apoptotic cell death.

Among the evaluated compounds, ReQBr and ReQMe exhibited the strongest and most reproducible proapoptotic activity, with a clear time-dependent increase in the apoptotic cell population. After 72 h of incubation, the percentage of apoptotic cells reached approximately 80–95% for ReQBr and 42–80% for ReQMe, depending on the CRC cell line analyzed.

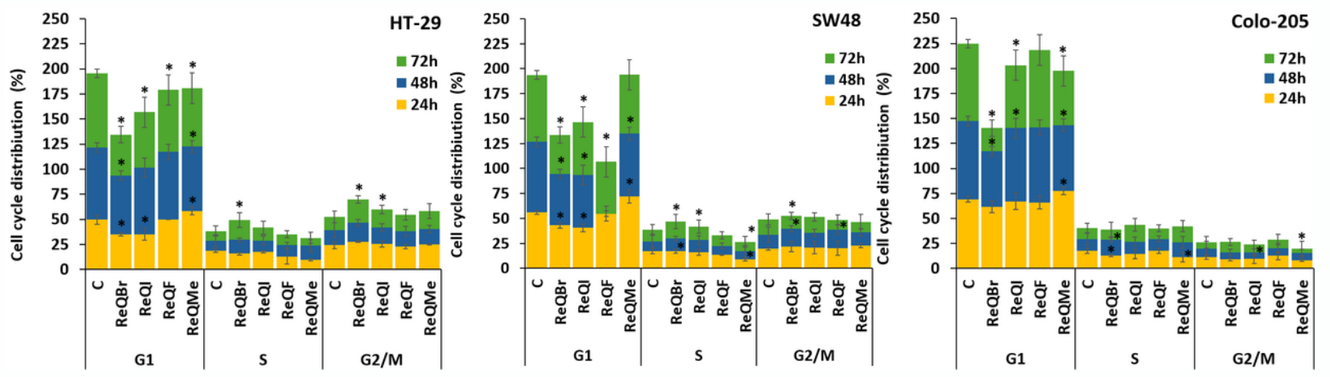


Figure 3. Changes in cell cycle distribution of CRC cells following treatment with Re(I) complexes (10 μM), assessed by flow cytometry. Data are presented as mean ± SD (n ≥ 5 independent experiments). *P < 0.05 vs. control.

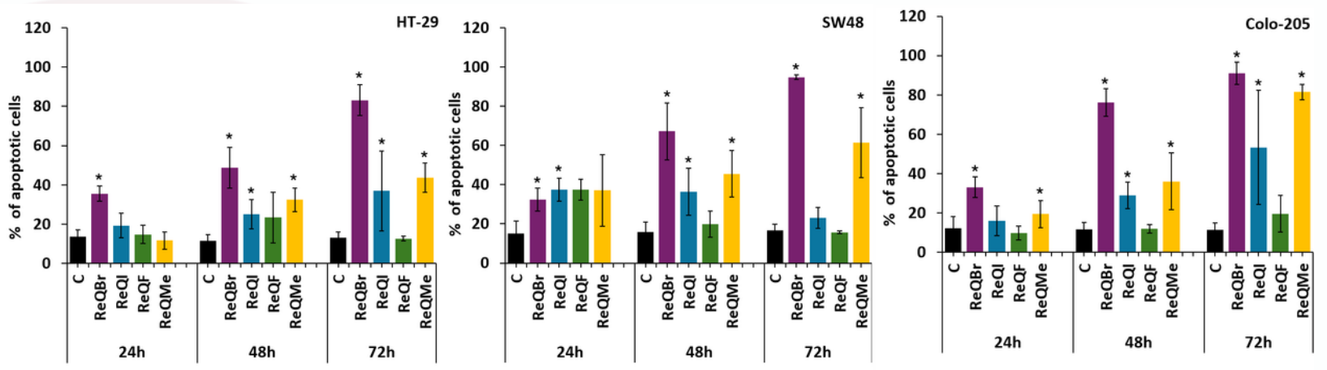


Figure 4. Apoptosis induction in CRC cells following treatment with Re(I) complexes (10 μM), assessed by flow cytometry. Data are presented as mean ± SD (n ≥ 5 independent experiments). *P < 0.05 vs. control.

Collectively, these findings indicate that the biological activity of the investigated Re(I) complexes is mediated through regulated antiproliferative and proapoptotic mechanisms rather than nonspecific cytotoxicity. The coordinated modulation of cell cycle progression and apoptosis strongly suggests the involvement of specific intracellular signaling pathways, the identification of which constitutes one of the major objectives of the proposed project.

DISCUSION

The obtained results identify ReQBr and ReQMe as promising leading compounds for further investigation. Future studies will focus on transcriptomic profiling and mechanistic characterization to elucidate the molecular pathways responsible for their anticancer activity and evaluate their potential in advanced 3D colorectal cancer models.

CONLUSION

- **Re(I) coordination transformed inactive ligands into potent anticancer agents.**
- **ReQBr and ReQMe showed the highest activity across CRC cell lines.**
- **Active complexes induced cell cycle arrest and apoptosis.**
- **Biological effects were time- and concentration-dependent.**
- **Re(I) complexes are promising candidates for further mechanistic studies.**