DOCKING AND MOLECULAR DYNAMICS ANALYSIS OF A NOVEL RHENIUM(I) COMPLEX BOUND TO JAK1 PROTEIN

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INTRODUCTION

The JAK/STAT signaling pathway, specifically JAK1 protein represents an important therapeutic target in the HuT-78 cell line, because it carries JAK1 mutations within an activated JAK/STAT cascade. Literature shows that inhibitors of this pathway can reduce the proliferation of CTCL cells, including HuT-78 [1] [2]. An MTT assay of the novel rhenium(I) complex **7d'$_{Re}$** shows a low IC_{50} value (2.1 μM) for HuT-78 treatment. In order to asses the metalic ligands potential to inhibit JAK1 protein, docking and molecular dinamics (MD) simulations were performed.

METHODOLOGY

The geometry of the rhenium(I) complex **7d'$_{Re}$** (ligand) was optimized using Gaussian 16, at the ωB97X-D level of theory, with the LANL2DZ basis set and effective core potential for Re and the 6-311++G(d,p) basis set for all other atoms. The crystal structure of the JAK1 kinase domain (PDB code 3EYG) was completed using MODELLER 10.8, protonated at physiological pH, and used for molecular docking with MetalDock 1.0.0. The best-ranked binding pose with preserved rhenium coordination geometry was selected for molecular dynamics simulations. Parameters for the metal complex were generated using easyPARM v4.20, while the protein was described with the ff19SB force field. The complex was prepared in AmberTools25, solvated in a truncated octahedral TIP3P water box, and neutralized with counterions. After four consecutive energy-minimization stages, the system was heated from 0 to 300 K and gradually equilibrated with decreasing positional restraints. Three independent 500 ns production simulations were then performed using AMBER 22 in the NPT ensemble at 300 K and 1 atm, with a 1 fs time step, Langevin temperature control, SHAKE constraints, and an 11 Å nonbonded cutoff. Trajectories were analyzed using CPPTRAJ to evaluate protein and ligand RMSD radius of gyration, ligand–protein hydrogen bonds, and the stability of the Re(I) coordination sphere.

RESULTS

Molecular docking results showed that the ligand binds within the active site of JAK1. The binding pose is primarily stabilized by a hydrogen bond with Leu95, together with additional polar and hydrophobic interactions involving Arg15, Leu17, Ala42, Met92, Glu93, Phe94, Leu146, and Asp157. These interactions indicate that the ligand occupies the catalytic region of the kinase domain and interacts with residues important for ligand binding.

Three independent 500 ns simulations showed that the protein–ligand complex remained structurally stable (Table 1). Backbone RMSD indicated replicate-dependent conformational changes, with replicate 3 showing the lowest overall deviation (Fig. 1). Ligand heavy-atom RMSD revealed higher mobility, but the ligand remained bound and the rhenium coordination sphere was preserved in all simulations (Fig. 2 and Fig 3). The protein radius of gyration remained nearly constant, indicating maintained global compactness. The most persistent protein–ligand hydrogen bond was LEU95 N–H···O5(LIG), with an occupancy of approximately 46–52% across the three replicates, while other hydrogen bonds were less offten (Table 2).

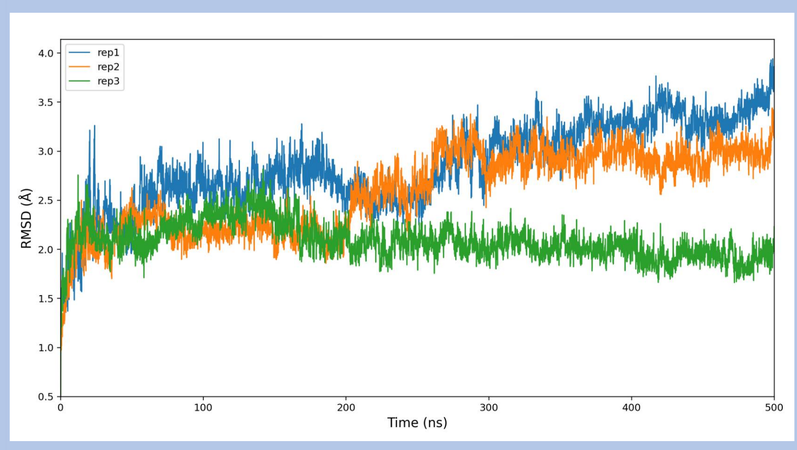

Fig 1. Backbone RMSD during 500 ns.

Table 1. Rhenium coordination distances across 500 ns.

Coordination bond	rep1 (Å)	rep2 (Å)	rep3 (Å)
Re–N1	2.077 ± 0.058	2.076 ± 0.057	2.076 ± 0.057
Re–N2	2.093 ± 0.052	2.094 ± 0.052	2.095 ± 0.052
Re–C9	2.054 ± 0.080	2.056 ± 0.080	2.057 ± 0.080
Re–C10	2.060 ± 0.072	2.057 ± 0.072	2.057 ± 0.072
Re–C11	2.054 ± 0.090	2.054 ± 0.089	2.053 ± 0.090
Re–Cl1	2.365 ± 0.048	2.365 ± 0.048	2.365 ± 0.048

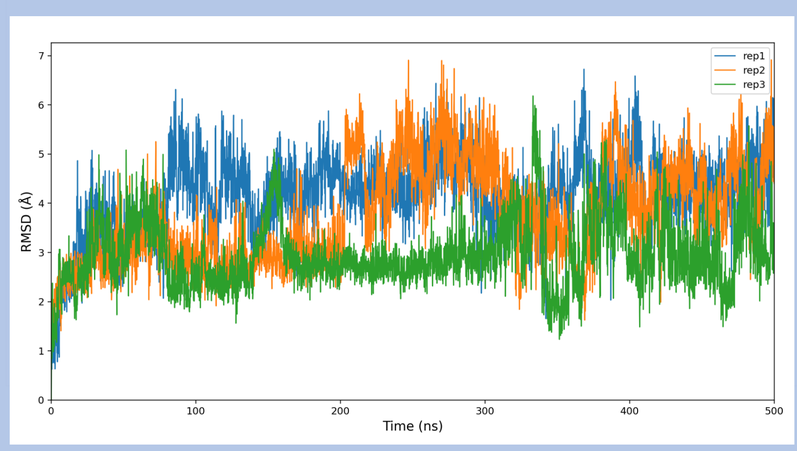

Fig 2. Ligand heavy atom RMSD during 500 ns.

Table 2. Representative conventional protein–ligand hydrogen bonds observed during 500 ns.

Hydrogen bond	rep1 (%)	rep2 (%)	rep3 (%)	Mean (%)	Distance (Å)	Angle (°)
LEU95 N–H···O5(LIG)	51.58	45.67	45.63	47.63	2.886	159.7
GLN40 NE2–HE22···O3(LIG)	1.47	0.42	0.13	0.67	2.908	152.1
N4(LIG)–H8···LEU95 O	0.03	0.64	0.00	0.22	2.842	158.4
N4(LIG)–H8···GLU93 O	0.00	0.37	0.05	0.14	2.904	163.9
SER99 N–H···O5(LIG)	0.00	0.16	0.22	0.13	2.876	150.4

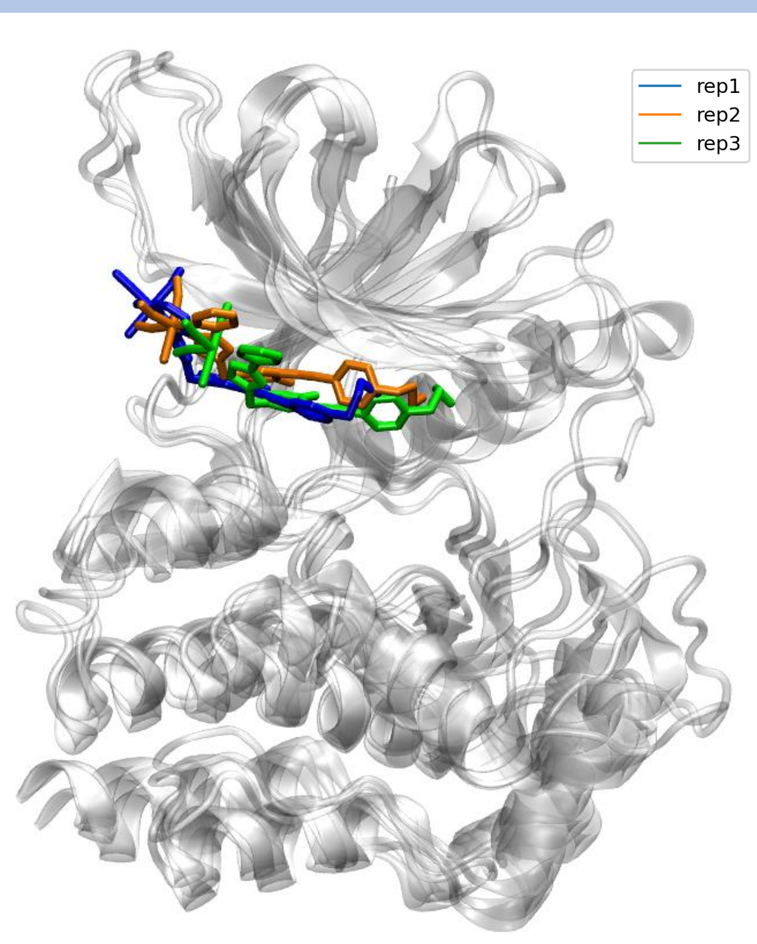

Fig 3. Ligan-protein complexes overlays at final frames.

CONCLUSION

The three 500 ns simulations confirmed that the protein–ligand complex remained globally stable. Protein compactness and the rhenium coordination sphere were preserved, while the ligand showed conformational flexibility. A persistent LEU95–ligand hydrogen bond contributed to stabilization of the complex. The same bond is present beween original ligand and JAK1 protein in PDB database.

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[2] C. Pérez, J. González-Rincón, A. Onaindia et al., Haematologica 100 (2015) e450–e453.

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