**BIO_15 - Discovery and characterization of sites through molecular dynamics with probes and virtual screening to propose new immunobiological targeting the PD-1**

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**Introduction:** Cancer is one of the leading public health problems worldwide and is among the top four causes of death in most countries. It is well established that the interaction between PD-1, an immune checkpoint receptor present in the T cell, and its ligand, PD-L1, present in cancer cells, results in immune evasion and maintenance of the tumorigenesis. Current treatments to block the interaction between PD-$1_{PD\text{-}L1}$ involve monoclonal antibodies (mAbs), such as nivolumab and pembrolizumab. Nowadays, there remains a need and interest in searching for new antibodies with better efficacy and activity.

**Objective:** Propose new antibodies that specifically bind and stabilize an unfavorable conformation of PD-1, causing an indirect inhibition in the formation of the complex with PD-L1.

**Methodology:** We use computational biology tools, such as Molecular Dynamics with probes (MixMD), Principal Component Analysis (PCA), and Virtual Screening, to identify and characterize interaction sites, describe novel conformations capable of impairing the interaction with PD-L1 and demonstrate, through stable ligands, how the process of inhibition and locking of the region occurs. PD-$1_{apo}$ (PDB: 2M2D) and PD-$1_{PD\text{-}L1}$ (PDB: 4ZQK) were used as control structures.

**Results:** The MixMD simulations are generally attested to the affinity of PD-1 for aromatic and hydrophobic probes and allowed the identification of an unprecedented site located in the C'D loop, close to the main interface between the two proteins. Furthermore, it is possible to verify that the ligands cause a conformational change in the region, locking the loop through changes in the dihedrals of E84 and S93 and reorganizing the side chain of R86 and E84. This conformation change has a prohibitive effect on the interaction between PD-$1_{PD\text{-}L1}$ given the collision between the F strand, the FG loop, and the N-terminal residues of PD-L1 with the C'D loop of PD-1.

**Conclusion:** Our results reveal a new conformation in PD-1 that prohibits interaction with PD-L1 and can be used as a reference for the formulation of alternative mAbs to block the interaction between the two proteins. From here, we are able, supported by the structural information obtained, to start prospecting for new antibodies targeting the PD-1 and PD-L1 pathway.

**Keywords:** Immunotherapy; Bioinformatics; PD-1