PROTEIN-LIGAND COMPLEX FOR STRUCTURE-BASED DESIGN: IMPACT ON THE AFFINITY AND ANTITUMOR ACTIVITY OF NEW TUBULIN LIGANDS

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Microtubules, made of αβ-tubulin heterodimers, are the key components of the cytoskeleton and play a crucial role in many cellular processes, such as cell motility, morphogenesis and mitosis.[1] Interference with microtubule dynamics induces cell cycle arrest during mitosis and triggers cell death. Compounds that interact with tubulin, especially those binding at the colchicine domain, have been deeply investigated as anticancer drugs due to their dual mechanism of action as antimitotics and as vascular disrupting agents.[2,3] Our research group has recently described a new family of colchicine-domain binders, based on a cyclohexanedione skeleton, with potent antiproliferative activity against tumor and endothelial cells.[4] Moreover, to gain insight into the binding mode of these cyclohexanediones, we have determined the crystal structure of αβ-tubulin in complex with our hit compound (TUB075). Based on this detailed information and by applying the affinity maps program cGRILL, a structure-based synthesis of new cyclohexanedione derivatives has been accomplished with the objective of improving their affinity for tubulin and their antitumor activity. Following this approach, we have obtained new compounds with potent antiproliferative activity against tumor and endothelial cells (IC_{50}=8-31 nM) and with the highest K_b value reported for compounds binding at the colchicine site in tubulin. Additional studies have shown that they arrest cell cycle at G2/M and disrupt a network of endothelial cells. Moreover they keep antiproliferative activity against cell lines overexpressing P-gp, further supporting the potential of these compounds. **Acknowledgements:** The financial support of the Spanish MINECO (SAF2012-39760-C02-01 and SAF 2015-64629-C2-1-R), Comunidad de Madrid (BIPEDD2; ref P2010/BMD-2457) and the COST action CM1407 (to M J. P.P., S.L., M.O.S. and J.F.D.) is sincerely acknowledged.

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