ABSTRACT

p53 is a transcription factor with tumour suppressor properties, which is able to induce mitochondrial apoptosis independently of its transcriptional activity. Analogues of the spiro[imidazo[1,5-c] thiazole-3,3′-indoline] -2′,5,7(6H,7aH) -trione, previously synthesized from my research group, as p53 modulators were synthesized during my PhD, aiming to explore new structural requirements at the thiazolidine domain to increase the antiproliferative activity and improve p53 modulation. Derivative 5-bromo-3′- (cyclohexane carbonyl) -1-methyl-2oxospiro[indoline-3,2′-thiazolidine] (**SM13**) emerged as the most potent compound of all series, inhibiting, in vitro, 30% of p53−MDM2 interaction at 5 μM and the cell growth of different human tumor cells at nanomolar concentrations. Docking studies confirmed the interactions of SM13 with the well-known Trp23 and Phe19 clefts, explaining the reasons for its binding affinity to MDM2. SM13 at 50 nM is capable of inducing the accumulation of p53 protein, inducing significant apoptotic cell death without affecting the cell cycle progression. Comparative studies using nutlin in the same cellular system confirmed the potential of **SM13** as a tool for increasing understanding of the process involved in the nontranscriptional proapoptotic activities of p53. Thus, the effectiveness of this compound in tumors carrying a mutated form of the p53 gene without transcriptional activity was verified.The effectiveness of **SM13** in cancer cell lines carrying WT, mutated and null p53 gene were evaluatedi vitro. At the same time, in vivo studies were performed in BALB/c nude mice and the signal-dependent mitochondrial apoptosis was evaluated by western stain. **SM13** reduced cell proliferation and induced apoptosis in the in vitro studies, suggesting that its effect is independent of p53 transcriptional activity. On the contrary, **SM13** had no effect in a null p53 cell line. In vivo, **SM13** induced tumor cell death in a dose dependent manner through the activation of death mitochondrial-dependent signaling in cells mutated p53. Overall these studies highlights the efficacy of **SM13** as anticancer cancer to be used for the treatment of p53-dependent tumors, even in the absence of transcriptional activity of p53.

A second part of my PhD was, instead, dedicated, to the synthesis of a series of highly functionalized DNTQ-based derivatives. Most of the synthesized compounds exerted, in vitro, a cytotoxic effect against several tumour cell lines greater than doxorubicin. In particular N-(4-chlorobenzyl)-4,9-dioxo-3-(2-(piperidin-1-yl) acetamido)-2,3,4,9-tetrahydronaphtho[2,3-b]thiophene-3- carboxamide (compound **14**), showed a reduced cardiotoxicity, inducing, at the ame time, cell differentiation and was distributed mainly in the cytoplasm in the human glioblastoma LN229 cell line. Moreover, compound **14** reduced both cellular glucose uptake and serine/threonine kinase AKT expression, and triggered cell apoptosis. These findings suggest that highly functionalized DTNQ-based derivatives are promising pharmacological tools for the study of human solid tumours.