Introduction

Human 3α -hydroxysteroid dehydrogenase type 5 (HSD17B5 or AKR1C3), belonging to aldo–keto reductase superfamily, is a NADPH-dependent enzyme. AKR1C3 catalases different enzymatic reactions among biosynthesis of bile acids and metabolism of steroid hormones and prostaglandins (PG) (*Penning et al. Mol Cell Endocrinol. 2004, 215:63*). AKR1C3 is highly expressed in testes, basal cells of the prostate, mammary glands and liver; it is overexpressed in prostate cancer (PCa) (*Byrns et al. J Sterois Biochem Mol Biol. 2011, 125:95*) and a strong correlation between AKR1C3 and colorectal cancer was also demonstrated (*Nakarai et al. Clin Exp Med. 2015, 15(3):333*).

The inhibition of AKR1C3 could be a new therapeutic strategy for cancers. Some compounds, as indomethacin, phenylanthranilic acids, flavonoids and cinnamic acids and cyclopentane derivatives, have shown the ability to inhibit AKR1C3, but no selective inhibitors of AKR1C3 are in clinical use at the moment (*Byrns et al. J Sterois Biochem Mol Biol. 2011, 125:95*).

The Medsynth group of the Department of Drug Science and Technology of Turin synthesized a series of derivatives of indomethacin and flufenamic acid, with the aim to obtain compounds with a selective inhibitory activity on AKR1C3. As COX activity of these drugs depends on a carboxylic acid moiety, which forms a salt bridge with Arginine 120, in these new compounds hydroxyazoles replace the carboxilic acid moiety, in order to increase the selectivity towards AKR1C3 over AKR1C2, losing COX activity. Structure–activity relationship studies show that the activity of those compounds derives from the trifluoromethylphenyl group and the 3-hydroxy-1,2-benzoxazole (*Pippione et al. Eur J Med Chem. 2018, 150:930*).

Materials and methods

To evaluate the selectivity of the new synthetized compounds, inhibitory activity assay was performed using recombinant purified enzymes with the oxidation of S-tetralol in the presence of NADP+, compared to flufenamic acid and indomethacin. Once demonstrated their selectivity, new derivatives were tested on three PCa cell lines: 22Rv1, LNCaP, VCaP and two colorectal cancer cell lines, HT-29 and HCT 116. The expression of AKR1C3, AR and PSA in the reported above cell lines was analysed by western blot experiments, while their antiproliferative activity was investigated by assessing cell viability and dosing ATP content with a luminescent cell viability assay (CellTiter-Glo®, PerkinElmer).

Results

AR and PSA are expressed in PCa cells lines and their expression is modulated by new compounds in a cell dependent way. Probably this can be linked also to the characteristics of the three cell lines utilized, as they show different levels of expression of these proteins, and AR can be present in normal or mutated form. We have also evidenced that AKR1C3 is present in both colorectal cancer cell lines. Treatment with new compounds, decrease cellular vitability in PCa and colocancer cell lines, showing a correlation between cell proliferation and AKR1C3 inhibition.

Discussion and conclusion

The new synthetized compounds represent a new generation of AKR1C3 inhibitors that have been designed using a bioisosteric approach and by replacing the carboxylic acid function of flufenamic acid and indomethacin with hydroxylated azoles. The activities of these compounds are found only at relatively high concentration, but the approach utilized to synthetized them could be optimized for the design of more potent and selective AKR1C3 inhibitors, allowing to improve our knowledge about AKR1C3's role in cancer development and other disorders.