The impact of mycophenolate mofetil on androgen receptor activity in prostate cancer cells 22Rv1

Ondřej Ženáta, Zdeněk Dvořák and Radim Vrál
Department of Cell Biology and Genetics, Faculty of Science, Palacky University, Stejskalova 27, 783 71 Olomouc, Czech Republic

1. Introduction:
Mycophenolate mofetil (MYC) (Figure 1) is widely used immunosuppressive drug in the prevention of organ rejection after heart or kidney transplantation. It’s primary target is the inhibition of inosine monophosphate dehydrogenase (IMPDH), enzyme which is essential in the synthesis of the guanosine nucleotides. After oral administration, MYC is rapidly hydrolyzed to mycophenolic acid (MPA) with high affinity to IMPDH II isoform, which is present in primary lymphocytes.

Recently, our research group find that MYC is strong activator of aryl hydrocarbon receptor (AhR), which is involved cell differentiation, apoptosis and immunity. On the other hand, it acted like an antagonist in glucocorticoid receptor (GR) signaling since it abolished the effects of GR agonist dexamethasone (Vrál et al., 2015).

Recent research show that some kinds of cancer are highly sensitive to MYC treatment (Dun et al., 2013). Treatment by AVN944, IMPDH II inhibitor, had antiproliferative effects and induced cell death in 4 cancer cell lines (Flynkt et Thompson, 2008). Thus, similar effects may be expected for MYC. To that purpose, we decide to investigate the effect of MYC on the activity of androgen receptor (AR) in androgen-independent cell line (22Rv1) compared to androgen-dependent line (LNCaP).

2. Androgen receptor is activated by mycophenolate mofetil:
As a first step, we had to determine non-toxic concentrations of MYC. We used the range correspondent with plasma concentrations (Pescovitz et al., 2000) with maximum 20 μg/ml (approx. 62μM). In all used concentrations, there was no decrease by more than 10% in AIZ-AR cell line (Figure 2).

As a second step, we used recently developed AR-responsive cell line AIZ-AR, which has been derived from 22Rv1 cells transfected with a construct containing three copies of androgen response regions (ARs) and one copy of androgen response element, respectively (Bartonková et al., 2015) to determine if MYC can activate AR.

In the agonist setting, there was significant induction of luciferase activity (1.4-1.7 fold) for three highest concentrations (1, 10 and 20 μg/ml), see in Figure 3A. Dihydropyrosterone (DHT) was used as a positive control of the functionality of the system (18-fold). In the Antagonist setting, we observed significant effect only for two lowest concentrations of MYC, which increased luciferase activity about 13% above DHT alone (Figure 3B).

3. Mycophenolate mofetil modulates KLK3 expression and proliferation of prostate cells:
One of the most known target genes of AR is KLK3 (kallikrein related peptidase 3), also known as PSA (prostate specific antigen). We observed effect of MYC on KLK3 mRNA in 22Rv1 (which the AIZ-AR cell line was derived from) and then on LNCap cell line. In 22Rv1, positive control significantly induced KLK3 mRNA about 2-fold, while MYC induced significantly about 60% growth of DHT for three highest concentrations (Figure 4A). Higher induction was observed in LNCap (approx. 15-fold) for positive control DHT, but surprisingly the highest MYC induction was only 1.5-fold for one concentration (about 3.5% of DHT effect, Figure 4B).

4. Conclusion:
We investigated the effect of MYC on the activity of AR in two prostate cancer cell lines. We found that MYC activated androgen receptor in androgen-independent cells 22Rv1 and induced AR-target gene expression (KLK3 mRNA) in both androgen-dependent as well as independent cells. However, there was a big difference in strength of activation AR target gene KLK3 (60% of DHT induction in 22Rv1 compared to 3.5% in LNCap). Moreover, the combination of MYC and DHT synergistically stimulated KLK3 mRNA induction in 22Rv1 cells whereas the opposite, i.e. the suppression was observed in LNCaP cells. Proliferation profile was similar for both cell lines, which can be the effect of MYC (IMPDH inhibition). This research provides novel information about the implications for widespread used transplant drug.

References:

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