ISGE
GYNECOLOGICAL ENDOCRINOLOGY
THE 18TH WORLD CONGRESS
7-10 MARCH 2018 FLORENCE, ITALY

Physiological and pathophysiological relevance of ZIP9, a zinc transporter, androgen receptor, and G-protein-coupled signal transducer

L Konrad (DE) [1], G Scheiner-Bobis (DE)

Context
In the so-called “classical” signaling pathway, testosterone binds to the nuclear androgen receptor (AR) that, in turn, regulates gene expression. In the “non-classical” pathway, testosterone activates cytosolic signaling cascades that are normally triggered by growth factors. The nature of the receptor mediating these signaling events has been a source of controversy.
Objective
To assess the nature of the AR mediating the non-classical signaling of testosterone.
Methods
We addressed testosterone effects in various cell lines by employing SDS-PAGE, western blots and immunofluorescence to detect and localize phosphorylated proteins, RT-PCR to address changes in expression, siRNA to abrogate the expression of proteins, in situ proximity assays to identify interacting proteins, and molecular modelling and docking experiments to identify and characterize the androgen binding site involved in the non-classical signaling of testosterone.
Patients
n.a.
Interventions
n.a.
Main Outcome Measures
n.a.
Results
In the spermatogenic cell line GC-2, non-classical signaling of testosterone, characterized by the activation of Erk1/2 and transcription factors like CREB or ATF-1 is mediated through ZIP9, a Zn2+ transporter that interacts with the G-protein Gn?11. siRNA-induced abrogation of the expression of either ZIP9 or Gn?11 completely prevented all testosterone effects addressed. Silencing of AR expression did not affect the signaling events investigated.
In the Sertoli cell line 93RS2, that lacks the classical AR, all testosterone effects were prevented by silencing ZIP9 expression or by bicalutamide, an anti-androgen of clinical use. Computer-based modelling and docking experiments revealed an extracellular androgen binding site of ZIP9 which could be labelled by the membrane-impermeable testosterone analogue T-BSA-FITC.
In the prostate cancer cell line LNCaP testosterone stimulated the phosphorylation of FAK and paxillin and stimulated the migratory behaviour of the cells. Abrogation of ZIP9 expression prevented all of these testosterone actions.

Conclusions

ZIP9 is a membrane-bound androgen receptor mediating non-classical signaling of testosterone. The accessibility of its androgen binding site from the extracellular milieu positions ZIP9 as an attractive pharmacological target for pro- or anti-androgens.

[1] Institute of Gynecology and Obstetrics, Giessen