FLUORESCENIN FOR EVALUATION OF THE UROTHELIAL BARRIER IN BLADDER PAIN SYNDROME

Hypothesis / aims of study
Bladder pain syndrome is a serious chronic condition with an unknown aetiology. There is consensus in literature that a deficient barrier is a key element in this condition. This can enable irritant solutes from the urine to invade the bladder wall and cause inflammation and activation of nerves. In order to corroborate this hypothesis objective clinical measurement of urothelial permeability is pivotal. In this study, fluorescein is validated as a marker for permeability in-vitro.

Study design, materials and methods

Porcine urothelial cells were isolated and cultured on membranes over which the transepithelial electrical resistance (TEER) could be measured using electrodes in the upper (A) and lower (B) compartment (see figure). This is a measure for the urothelial barrier function. Three groups were tested: healthy urothelium n=5, damaged urothelium n=5 and empty membranes n=3. To damage the urothelium, protamine was used what breaks down essential barrier components. TEER was measured after which fluorescein was incubated at the apical side of the cell layer. The fluorescein concentration in both compartments was measured using spectrofluorometry after 15, 30, 60 and 90 minutes. The ratio of this was measured between the groups using an unpaired T-test.

Results
Enzymatic treatment of the urothelial cells showed a significant decrease of resistance measured by TEER. In the damaged urothelium there was a significant increase of fluorescein concentration in the lower compartment as was with the empty membranes (both p<0.001) as opposed to the healthy urothelium. In the damaged group there was fluorescein leakage of 2.1% versus 0.7% in healthy urothelium which was significantly different (p=0.04).

Interpretation of results
Damaging of the urothelium leads to a decrease in TEER. With this, there is an increased permeability for fluorescein.

Concluding message
Fluorescein permeability correlates with electrical resistance measurements of urothelium in-vitro. Hence, fluorescein may serve as a marker for urothelial permeability to give an objective evaluation of damaged urothelium.

Disclosures
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