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**Introduction & Objectives:** PCa is known to be an androgen dependent disease. However, estrogens acting via estrogen receptors (ER) also play important role in the disease development and progression. The two main subtypes of ER are ER alpha and ER $\beta$ , often mediating opposing effects in PCa. Besides, stromal-epithelial interactions are important, with ER $\beta$  expressed in different tissue compartments playing various roles in the overall response to estrogens and other compounds. The aim of the work was to study characteristics of ER $\beta$  expression in clinical PCa samples.

**Materials & Methods:** ER $\beta$  expression was studied in 28 radical prostatectomy samples. Whole prostate was assessed for histopathological findings and 1 representative slice per case was chosen. Heat induced antigen retrieval was used. ER $\beta$  expression was assessed by immunofluorescent technique using primary rabbit polyclonal antibodies (BioGenex, 1:500 dilution) and secondary goat anti-rabbit antibodies labeled with Alexa Fluor 555 dye (ThermoFischer, 1:200). Slides were studied using Olympus BX53 microscope and ER $\beta$  expression was assessed semiquantitatively in 3-15 random non-crossing high power fields (HPF, x400) per case (total n=251), separately in cancer and stromal cells. For each HPF the proportion of stained nuclei (0 – absent, 1 – up to 1/3, 2 – 1/3 to 2/3, 3 - >2/3) and staining intensity (1 – weak, 2 – moderate, 3 – strong) were assessed with multiplying them to get the total staining score (TSS) per HPF. Mean TSS per case was also calculated.

**Results:** ER $\beta$  expression was present in all studied cases both in epithelium and stroma, with slightly higher rates in epithelium. The numbers of HPFs with various staining characteristics are shown in the table.

Score	Epithelium		Stroma	
	Proportion	Intensity	Proportion	Intensity
0	13 (5.2%)	13 (5.2%)	8 (3.19%)	8 (3.19%)
1	62 (24.7%)	133 (53.0%)	61 (24.3%)	110 (43.8%)
2	44 (17.5%)	41 (16.3%)	69 (27.5%)	73 (29.1%)
3	132 (52.6%)	64 (25.5%)	112 (44.6%)	59 (23.5%)

Mean TSS per HPF was 4.14 for both epithelium and stroma. Mean TSS per case was 4.18 (range 0.14-9) for epithelial ER $\beta$  and 4.15 (0.57-8.75) for stromal. Significant positive moderate correlation was observed for epithelial and stromal ER $\beta$  levels ( $r=0.57$  at HPF level and  $r=0.62$  at case level,  $p_{\text{Spearman}} < 0.05$ ). No statistically significant differences were observed between ER $\beta$  expression in PCa with Grading group 1-2 or 3-5 and in pT2 vs. pT3 tumors ( $p_{\text{Mann-Whitney}} > 0.05$ ) but this may be due to a relatively small sample size.

**Conclusions:** ER $\beta$  expression in PCa is highly prevalent and present in all cases, with most cells stained but with weak intensity. Levels of epithelial and stromal ER $\beta$  are positively correlated, however, no significant associations with major clinicopathological variables were observed. Further analysis of ER $\beta$  associations with other receptors and epithelial-mesenchymal transition and cancer stem cell markers in PCa is ongoing.