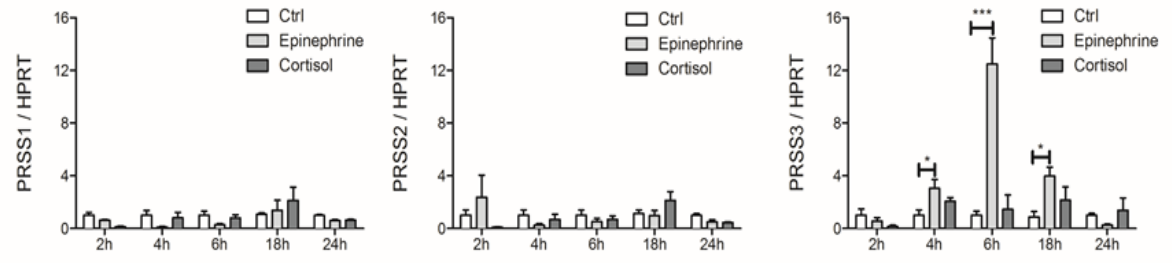
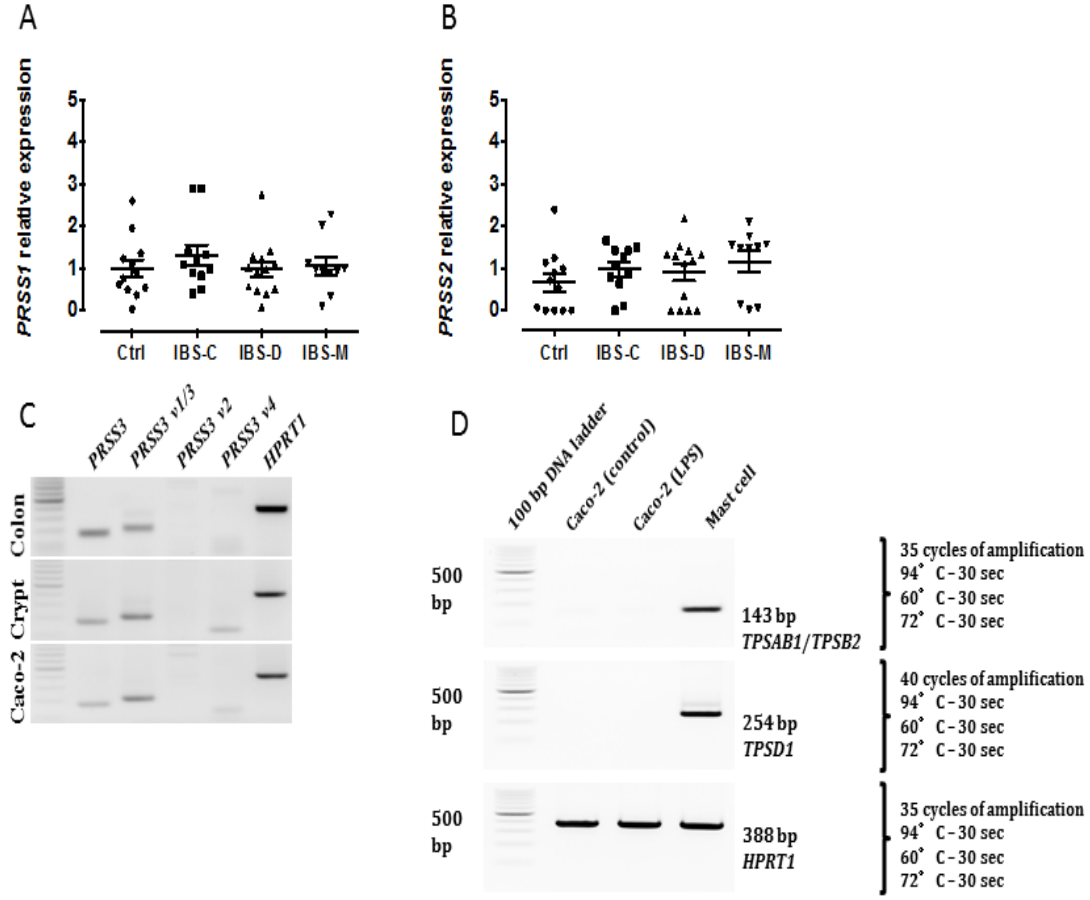
Supplementary figures:

**Supplementary Figure 1:** Kinetic expression of the Trypsin genes PRSS1, PRSS2 and PRSS3 alternative splice variants in intestinal epithelial cells (Caco-2) at different times after their exposure to Epinephrine or to Cortisol. Data are expressed as mean ± SEM.



**Supplementary Figure 2:** Expression of the Trypsin genes PRSS1, PRSS2 and PRSS3 alternative splice variants, and the Tryptase genes TPSAB1, TPSB2, TPSD1. Relative gene expression of **A.** *PRSS1* (cationic trypsin: Trypsin-1 precursor) and **B.** *PRSS2* (anionic trypsin: Trypsin-2 precursor) in colonic biopsies from control and IBS patients. Data are expressed as mean ± SEM and were compared using Student's *t*-test. **C.** Analytical agarose-gel electrophoresis of RT-PCR products performed with RNA extracted from human colonic biopsies (control patient), isolated colonic crypts or Caco2 cells. Amplicons were amplified with oligonucleotides for total *PRSS3* (170 bp), or the alternative splice variants 1 (coding for Trypsinogen IV; 202 bp), 2 (coding for Mesotrypsinogen; 667 bp), 3 (330 bp) and 4 (122 bp) and *HPRT1* (388 bp). **D.** Representative agarose-gel electrophoresis of RT-PCR products performed on RNA extracted from Caco-2 cells stimulated or not with LPS, or primary mast cells derived from human CD34⁺ precursor cells. The amplicon in the top panel was amplified with oligonucleotides common for the genes coding for tryptase-β1 and tryptase-β2 (*TPSAB1* and *TPSB2*, respectively). The amplicon in the middle panel was amplified with oligonucleotides specific for tryptase-δ1 (*TPSD1*). Amplicons in the bottom panel were amplified with oligonucleotides specific for the house-keeping gene *HPRT1*.



**<u>Supplementary Figure 3.</u> Trypsin-3-evoked calcium signals in mouse nociceptive DRG neurons and human submucosal neurons are PAR1 and PAR4 independent. A.** Average amplitude of $[Ca^{2+}]_i$ rises and percentage of responding neurons to Trypsin-3 (10 nM) exposure in mouse nociceptive DRG neurons in the presence or absence of the PAR1 (SCH79797) or the PAR4 (ML-354) specific antagonists. Data are expressed as mean ± SEM and were analyzed by One-Way ANOVA followed by Kruskal Wallis's post-test, (n=6 mice per group), *** and * different from basal for p<0.001, P<0.05 respectively. **B.** Average amplitude of $[Ca^{2+}]_i$ rises and percentage of responding neurons to Trypsin-3 (10 nM) exposure in human submucosal neurons in the presence or absence of the PAR1 (SCH79797) or the PAR4 (ML-354) specific antagonists. Data are expressed as mean ± SEM and were analyzed by One-Way ANOVA followed by Kruskal Wallis's post-test, (n=3 subjects per group), ***p<0.001.