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## Expression of TLR-2, 4 and 5 and proinflammatory (IL-6, CXCL-11 and CXCR-3) and anti-inflammatory cytokines (IL-10) among patients with irritable bowel syndrome

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Evidence of low grade inflammation and altered host-microbial interactions suggests that innate immune response may play an imperative role in the pathogenesis of irritable bowel syndrome (IBS). We aimed to study of TLRs (2, 4 and 5), cytokines (IL-6 and IL-10) and chemokines (CXCL-11 and CXCR-3) expression in colonic biopsies among patients with IBS. Quantitative real-time PCR was used to determine mRNA level of TLRs, cytokines and chemokines in 47 patients with IBS (Rome III criteria) and 25 controls. Expression of TLR-4 and TLR-5 was confirmed at protein level using immunohistochemistry. Of 47 patients with IBS, 20 had constipation (IBS-C), 20 had diarrhea (IBS-D) and 7 unclassified (IBS-U). Expression of TLR-4 and TLR-5 was up-regulated in IBS patients than controls (P=0.013 and P<0.001, respectively). Protein level of TLR-4 and TLR-5 was 4.2 and 6.6-fold higher in IBS-D patients than controls. The mRNA level of IL-6 (P=0.003), CXCL-11 (P<0.001) and CXCR-3 (P<0.001) was higher in IBS patients than controls. Expression of IL-6 (P=0.002), CXCL-11 (P<0.001) and CXCR-3 (P<0.001) was higher, while IL-10 (P=0.012) was lower in IBS-D patients than controls. A positive correlation was found between TLR-4 and IL-6 (P=0.043), CXCR-3 and CXCL-11 (P=0.047), and CXCR-3 and IL-6 (P=0.003). Stool frequency per week showed positive correlation with mRNA level of TLR-4 (P=0.016) and CXCR-3, inversely correlated with IL-10 (P=0.002). Up-regulations of TLRs (4 and 5), proinflammatory cytokine (IL-6) and chemokines (CXCL-11 and CXCR-3) and down-regulation of anti-inflammatory cytokine (IL-10) may influence intestinal inflammation in a subgroup of IBS patients.

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## Immunological assay and arbitration inspection from single to multiple mycotoxins in agro-food

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Multiplexed mycotoxins with strong carcinogenesis and toxicity are fatal threats in food and feed safety, and require highly sensitive and high-throughput detections greatly. Rapid immunoassay and arbitration detection methods play a critical role on two sides of one coin. In the rapid immunoassay, a series of high specific and high affinity monoclonal antibody, recombinant antibody and nano-body against aflatoxin B1 (AFB1), ochratoxin A (OTA), and zearalenone (ZEA), etc., were developed as the key recognition reagents. Based on these specific antibodies, simultaneous detection for multiplexed mycotoxins was studied by using the Au (or Europium)-based lateral flow strip and non-fouling antigen microarray. The limit of detection was lowered down to pg mL-1 level (0.3 pg $mL^{-1}$), depending on mycotoxins in food and feed samples. On the other hand, simultaneous arbitration detection method based on LC-MS/MS was investigated. Either multiplexed immuno affinity column or solid phase extraction column was used in the sample extraction. The internal standard allowed precise determination of mycotoxins regardless of matrices. Multiplexed mycotoxins (AFB1, B2, G1, G2, OTA, ZEA and T-2 toxin) were successfully identified by using a new multi-immuno affinity column in a single run. Furthermore, a promising proposal was suggested to achieve the rapid, sensitive, ultra high-throughput detection of 96-384 contaminants in food and feed, including bio toxins, pesticides, veterinary drugs, etc., based on immuno chemiluminescence biosensors using Hadamard transformation imaging (iHT).

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