Effects of 5HT on Cellular and Cytokine Activation Profiles during Ex Vivo Stimulation of Human Leukocytes

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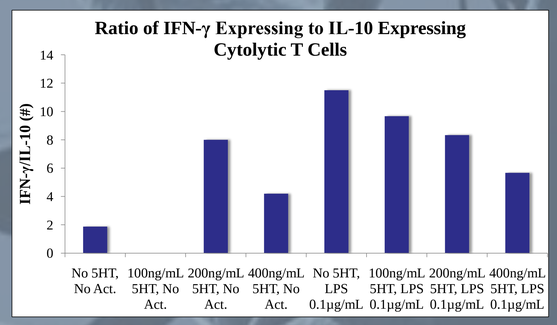
Introduction

Serotonin (5HT) is primarily known as a neurotransmitter but has recently been thought to be a modulator of the immune system. Outside the brain, 5HT is normally produced from enterochromaffin cells and taken up by circulating platelets. When an inflammatory process is triggered, platelets aggregate at the site of inflammation and release 5HT into the microenvironment, activating the immuno-regulatory effects of 5HT. During the early stage of inflammation, 5HT serves as an immunostimulant, yet reverses to become an immunosuppressant during the late stage.

Purpose

Our series of experiments sought to quantify and examine the anti-inflammatory effects of 5HT at physiological conditions, mimicking an *in vivo* environment. By confirming the anti-inflammatory property of 5HT, we aim to provide potential foundations for medicinal treatments using 5HT-based drugs, especially for immuno-regulatory malfunctions illnesses such as those pertaining to chronic inflammation.

Results



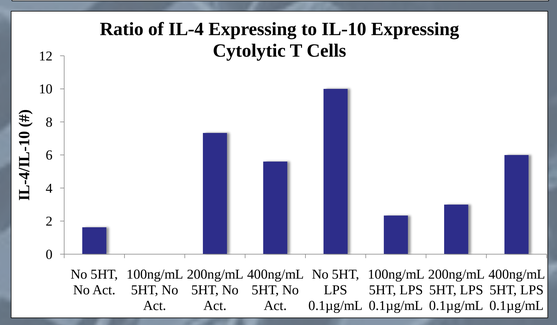
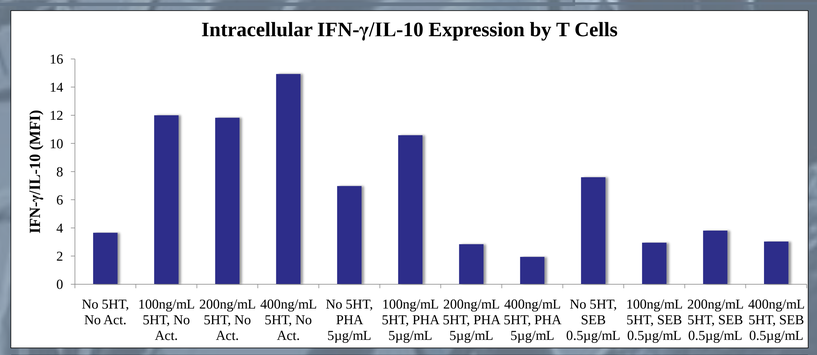


Figure 1. Effect of 5HT on the ratios of IFN-γ expressing and IL-4 expressing to IL-10 expressing cytolytic T cells at 21 hours post-activation. During activation by LPS, both IFN-γ/IL-10 and IL-4/IL-10 expressing cytolytic T cells ratios generally decreased with addition of 5HT. This result was produced from 3 independent trials.



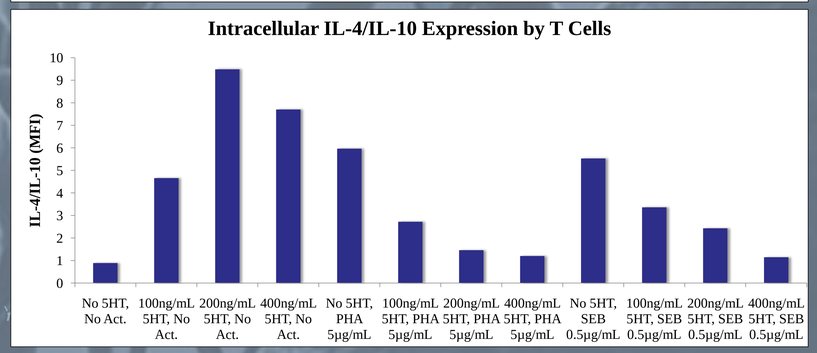


Figure 2. Effect of 5HT on the IFN-γ/IL-10 and IL-4/IL-10 intracellular expression ratios in T cells at 21 hours post-activation. During activation by PHA or SEB, 5HT dose-dependently decreased IFN-γ/IL-10 and IL-4/IL-10 intracellular expression ratios. This result was produced from 3 independent trials.

Methods

In our experiments, we used concentrations of 5HT that would represent both baseline level (100-200ng/mL) and pathologically activated level (400-800ng/mL). We also used the activating agents Lipopolysaccharide (LPS), Phytohaemagglutinin (PHA), and Staphylococcal Enterotoxin B (SEB) to trigger the inflammatory process. Thereafter, we quantified cytokine expression and release by flow cytometry and multiplexed quantitative assays. We hypothesized that during long-term incubation, the presence of 5HT would dose-dependently augment the resolution of inflammation, illustrated by either absolute increases in IL-10 expression or lower pro- to anti-inflammatory ratios.

Discussion

During activation by LPS, PHA, or SEB, 5HT decreased IFN-γ/IL-10 and IL-4/IL-10 ratios in T cells. Our results show that the presence of 5HT was able to inhibit pro-inflammatory cytokines expression but only after taking into account the relative cytokine concentrations in the microenvironment. However, while these results indicate that addition of 5HT down-regulated both Th1 and Th2 immune responses during general activation, it displayed more potency in down-regulating the Th1 immune response during PHA or SEB stimulation.

Conclusion

These experiments showed that physiological concentrations of 5HT effectively induced anti-inflammatory effects on T-cells and phagocytes. Using these results, future studies should focus on both reproducing these experiments on a larger number of trials and expand in time and concentrations used. Because our series of experiments contain only a small number of independent trials, it is limited in predictive power. However, the fact that our results supplement and stand concordantly with literature suggest credibility and potential for further exploring the effects of 5HT on a molecular level using agonists and antagonists, leading to the development of 5HT-based drugs to dually treat chronic inflammation and depression.