

# FUNCTIONAL CHARACTERIZATION OF RECOMBINANT INTERLEUKIN (IL)-17A/F1 IN THE JAPANESE PUFFERFISH (*TAKIFUGU RUBRIPES*)

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## ABSTRACT

In mammals, interleukin (IL)-17A and IL-17F are hallmark inflammatory cytokines, which are expressed by Th17 cells. In contrast, teleost IL-17A and IL-17F homologs named as IL-17A/F has been identified, and its functional aspect is poorly understood. Here, the recombinant protein of the Japanese pufferfish, *Fugu (Takifugu rubripes)* IL-17A/F1 was produced and purified using mammalian cell line expression system where the protein folding was achieved correctly. The recombinant (*r*) IL-17A/F1 was added into head kidney (HK) and intestinal endothelial (IE) cells prepared from *Fugu* to stimulate the immune regulations, and then phagocytic activity, superoxide anion production by NBT assay, and cytokine gene expressions in the treated HK and IE cells at 0, 3, 6, 12, 24 and 48 hours post-stimulation (hps) were conducted. The *r*IL-17A/F1-treated both HK and IE cells showed an increased phagocytic activity at 24 hps. Superoxide anion production increased significantly at 48 hps in the HK cells and also increased moderately at 48 hps in the IE cells. Moreover, the *r*IL-17A/F1 displayed an ability to enhance expression of pro-inflammatory cytokine mRNAs, such as IL-1 $\beta$ , TNF- $\alpha$  and IL-6 in *Fugu*. The expression of IL-1 $\beta$ , TNF- $\alpha$  and IL-6 genes was significantly up-regulated during 3 to 12 hps. These bioactivity and gene expression results suggest that *Fugu* IL-17A/F1 probably plays an important role in promoting inflammatory and host innate immune responses as a pro-inflammatory cytokine in fish.

## KEYWORDS

IL-17A/F, pro-inflammatory cytokine, phagocytic activity, gene expression, Japanese pufferfish

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