



## RT-qPCR application for analysis of *IL1B* expression in migraine

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### Introduction

Migraine is a disabling chronic neurological disease, and its pathophysiology is still unclear. Interleukin (IL) 1 $\beta$ , a pro-inflammatory cytokine, influences migraine pathophysiology by affecting nociceptors and pain sensitivity. Elevated plasma levels of IL-1 $\beta$  in migraine patients have been reported previously. Thus, studying cytokine gene expression in migraine could promote the identification of diagnostic and therapeutic targets for the disease, particularly in relation to inflammation and vascular response modulation.

### Objectives

This study aimed to design primers and standardize a method for comparative analysis of *IL1B* expression based on reverse transcription coupled to real-time polymerase chain reaction (RT-qPCR), in patients with migraine and control patients.

### Methods

A pair of sense and antisense primers complementary to the human *IL1B* gene were designed based on a consensus sequence generated after analysis of nucleotide sequences deposited in GenBank, and PickPrimer software. Subsequently, the primers were assessed for specificity using BLASTn tool and dimer formation with DinaMelt tool. Standardized cDNA and primer concentrations were used for RT-qPCR validation on migraine patient leukocytes obtained from the peripheral blood. RNA extraction from clinical samples was performed using the Trizol®. A comparative analysis of the relative expression of *IL1B* transcripts was conducted on clinical samples obtained from 50 individuals (32 migraine patients and 18 controls). RT-qPCR was utilized to analyze *IL1B* gene expression.

### Results

Median *IL1B* expression was 10.7 times higher in migraine patients and 3.4 times in controls, though not statistically significant.

### Conclusion

The study identified no significant difference in *IL1B* transcript levels between migraine patients and controls. Nevertheless, this investigation is pioneering in its approach. RT-qPCR proved to be specific for the detection of *IL1B* transcripts and can be standardized for the study of other modulators. Future research involving samples from migraine patients during an acute migraine attack could further elucidate the role of IL-1 $\beta$  in migraine pathophysiology.