

Patterning of Protocol to Evaluate of a microRNA (has-miR-1) circulating as a non-invasive biomarker to the Chronic Chagas Cardiomyopathy

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INTRODUCTION: The Chagas Disease (CD) is a pathology that about one third of people infected evolved to the Chronic Chagas' Cardiomyopathy (CCC), clinic form responsible for the majority number of deaths. Many studies has gave special attention to seek of the factors involved with the progression to this disease form. Of which has shown that the pattern deregulate of the target gene's expression contributes to a resulting pathological phenotype. **OBJECTIVE:** This study goals the validation of protocol to extraction of circulating miRNA, reverse transcriptase and confirmed on agarose gel, in order to patterning of extraction conditions, quantity of genetic material (cDNA) and primers, seeking to optimized before start the real-time PCR (qPCR). **MATERIAL AND METHODS:** Recruitment of individuals that presents TCLE and agree to participate the study project (groups: non-Chagas disease and without heart disease, cardiac patients without Chagas disease, Chagas disease in FI and contractility segmental changes on echocardiogram, Chagas disease in FI and without alteration in the echocardiogram and patients with CCC); collection of peripheral blood with serum separation and storage; purification of circulating miRNAs from the serum of recruited individuals; reverse transcription to cDNA synthesis and conventional PCR with GAPDH primer. **DISCUSSION AND RESULTS:** We confirmed the success of the process of extracting the total RNA and reverse transcription from conventional PCR with GAPDH primer, and the ideal concentration f the primer that will be used as "housekeeping" (GAPDH) in the real-time PCR. In addition to establishing the optimal amount of cDNA for amplification and visualization in agarose gel. **CONCLUSION:** Considering the difficulty to work with genetic material which has only 21 to 22 nucleotides, like the circulating miRNAs, the patterning of maximum possible factors is necessary for optimization purposes. Once the next step will be the real-time PCR for quantitative and qualitative analysis of has-miR-1, possible nom-invasive marker to be evaluate.

Key words: Chagas Disease; Biomarker; has-miR-1.

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