Pre-miRNA and mature miRNA in human mitochondria

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Introduction

Because of the central functions of the mitochondria in providing metabolic energy and initiating apoptosis, we hypothesized that some miRNA could be present in the mitochondria for post-transcriptional regulation by RNA interference. We intend to identify miRNA localized in the mitochondria isolated from human skeletal primary muscular cells.

Materials and methods

Bio-informatic analysis

To investigate the potential origin of mitochondrial miRNA, we in silico searched for putative
• pre-miRNA and mature miRNA sequences
• miRNA target sites
in the miRNA reference sequence. All the analysis were performed using miRBase and bio-informatic tools.

Results

Bio-informatic analysis

Mitochondrial miRNA candidates

Twenty-five human pre-miRNA and 33 miRNA alignments (E-value<0.01) were found in the reference mitochondrial sequence. The most significant alignments with human miRNA were obtained with four pre-miRNA (pre-mir-302a, pre-let-7b, pre-mir-1267 and pre-mir-1296; E-value<0.01) and with the two miRNA (mir-365 and mir-31; E-value<0.1). The presence of the best candidates in myoblastic mitochondria was further evaluated by in situ hybridization.

Colocalization of pre-miRNA and mRNA in mitochondria by in situ hybridization

In situ hybridization of pre-mir-302a, pre-let-7b, let-7b and mir-365, using specific labelled locked nucleic acids (LNA) probes, demonstrated that these miRNA were colocalized in mitochondria of human myoblasts.

MicroRNA detection by RT-qPCR

The detection of 742 human miRNA (miRBase) were monitored by RT-qPCR at three increasing miRNA inputs. Forty-six miRNA were significantly expressed for the smallest RNA input concentration and 204 miRNA for the maximum RNA input concentration.

Conclusion

The present study experimentally demonstrated for the first time the presence of pre-miRNA and miRNA in the human mitochondria isolated from skeletal muscular cells. A set of miRNA were significantly detected in mitochondria fraction. The origin of these pre-miRNA and miRNA should be further investigated to determine if they are imported from the cytosol and/or if they are partially processed in the mitochondria.

Reference:

Pre-microRNA and mature microRNA in human mitochondria.