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Sequencing the ovine pancreatic RNase A gene, a potent immunotoxin, in Iranian wild and domestic sheep

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Abstract: Ribonucleases (RNase) are promising agents for use in cancer therapy. Among the different ribonucleases described to be cytotoxic, pancreatic ribonuclease drew attentions due to similarity with Onconase[®]. Ruminants produce high levels of pancreatic ribonuclease in the digestion tract to deal with high content of nucleic acids. The activity of this enzyme in ruminants is also the highest among other mammalian species. The ovine pancreatic RNase A gene characteristics is not well investigated in compare to bovine and human orthologs. In this study, we sequenced complete pancreatic RNase A gene from10 samples of Baluchi and wild sheep from Khorasan-e-Razavi. Seven pairs of overlapping primers have been designed to cover 4347 bp of the RNase gene. Genomic DNA was extracted from fresh blood samples and standard Sanger sequencing was conducted after PCR amplification of fragments from ovine RNase gene. Alignments of 4347 bp from studied animals showed a 99.78% homology between Baluchi and wild sheep and a 99.61% homology with the reference sequence registered in the NCBI GenBank. However, coding region alignment showed complete homology between Baluchi and wild sheep and the reference sequence, which demonstrated a high degree of conservation. Phylogenetic comparison of RNase A sequences from different mammalian species revealed that the closest species to sheep is Goat's RNase A and then cattle. Sequences obtained from this study will be deposited in the GenBank in the near future after in silico protein structure predictions.

Keywords: sheep; RNase A; pancreas, cytotoxicity