Identification of reliable miRNA targets based on combinatorial miRNA action and integration of different prediction tools

ID - 208

Re Angela[1], Caselle Michele[1,2]

[1]Department of Theoretical Physics, University of Turin, Turin

[2]Interdepartmental Centre on Complex Systems in Biology and Molecular Medicine, Turin

Motivation

MicroRNAs (miRNAs) are a family of ~22 nucleotide-long non-coding RNAs which negatively regulate gene expression at the post-transcriptional level in a wide range of organisms.

Even though the precise mechanism of action of miRNAs is not very well understood, the current paradigm is that miRNAs are able to negatively affect the expression of a target gene via mRNA cleavage or translational repression, after antisense complementary base-pair matching to specific target sequences in the 3' UTR of the regulated genes. In animals the complementarity is restricted to the 5' regions of the miRNA, in particular requiring a binding sequence of 7 nucleotides, usually (but not always) from nucleotides 2 to 8. While several databases of experimentally validated miRNAs already exist much less is known from an experimental point of view on their targets. Thanks to the above mentioned complementarity between the 5' end of the miRNAs and their targets, starting from the miRNA sequence one could in principle identify miRNA targets by pure bioinformatics methods. Several prediction tools which perform this type of analysis exist, the most popular being PicTar and MiRBase.

However all these tools are affected by a large number of false positive predictions.

The aim of our project is to select among all the putative candidates a list of highly reliable targets by looking for instances of cooperative interactions among different miRNAs

Methods

Our strategy relies on two major features: the quantitative estimation of the combinatorial action of miRNA-mediated regulation and the comparative study of putative targets predicted by algorithms that exploit complementary features of miRNA targeting.

It is widely assumed that miRNA regulation has a combinatorial nature. Indeed the majority of experimentally validated examples of miRNA regulation support this assumption.

To implement this observation we looked for the presence of binding sites of different miRNAs in the same 3 UTR and considered this event as supporting for the mRNA being a target of this miRNA.

To this end we constructed a miRNA-based regulation network whose nodes are miRNAs and a link between two nodes is drawn if these miRNAs share a statistically significant subset of targets.

After performing network topology analysis, we are able to provide a set of highly selected miRNA targets based on the fact that they appear to be cooperatively regulated by two or more miRNAs.

We applied this analysis using as input the miRNA-target interactions obtained by the two freely available computational tools mentioned above: miRBase and PicTar (we include both versions of PicTar which differ in their cross-species conservation requirements). Since these programs use very different binding sites properties to define their search space for prediction we consider the possible existence of common subnetworks in the intersection of the two networks as highly significant and use it to further select optimized miRNA target sets.

Results

We describe a method to identify reliable miRNA targets looking for instances of cooperative regulation. We applied the method to the predictions of the miRBase and PicTar tools.

Besides targets resulting from the two tools separately, we also found twelve miRNA pairs in the intersection of the two networks. The components based on such pairs target a highly significant number of mRNAs ranging from 7 to 33. We consider these targets as the most reliable ones. Work is in progress to characterize these targets and for a possible experimental validation.

Email: angelare@to.infn.it