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To cite this version:
Marc Legeay, Béatrice Duval, Jean-Pierre Renou. Context-specific co-expression networks to explore the impact of anti-sense transcription. 13th conference on Computational Methods for Systems Biology (CMSB’15), Sep 2015, Nantes, France. 2015. hal-02516452

HAL Id: hal-02516452
https://univ-angers.hal.science/hal-02516452
Submitted on 23 Mar 2020

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Context-specific co-expression networks to explore the impact of anti-sense transcription

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Motivations
Understanding the regulation mechanisms in a cell is a key issue in bioinformatics. Anti-sense RNAs are endogenous RNA molecules whose partial or entire sequences exhibit complementarity to other transcripts. Their different functional roles are not completely known but some studies suggest that they play an important role in stress response mechanisms. A recent study with a full genome microarray for the apple has detected anti-sense transcription for 65% of expressed genes, which suggests a high potential of anti-sense regulation. Our work integrates anti-sense transcripts into context-specific gene networks and proposes a differential analysis of those networks.

Biological materials
We use microarray data of RNA extracted from the apple fruit (Malus × domestica) thanks to the chip AryANE v1.0 containing 63011 predicted sense genes and 63011 complementary anti-sense sequences. We study the fruit ripening process described by two conditions: harvest (H) and 60 days after harvest (60DAH), and for each condition, 22 samples of apple fruit have been analyzed.

Methods
We start with expression data of sense and anti-sense genes in two stress-response conditions. From there, we propose five steps to analyse the role of anti-sense transcripts:

- Identification of genes of interest: genes (sense and anti-sense) differentially expressed.
- Functional analysis of genes of interest to compute the revealed-by-AS terms.
- Contextual construction of gene network for the genes of interest. We construct a co-expression gene network for each condition from the expression of the genes of interest.
- Module detection: In each network, we look for highly correlated genes by grouping them in modules thanks to a hierarchical clustering.
- Network analysis, this work is in progress.

Functional analysis
Revealed-by-AS terms allow us to identify biological functions which could not have been noticed by enrichment analysis without anti-sense data.

For the dataset described before, one of the most significant revealed-by-AS terms is the term “response to cold” (red arrow in Figure 5). Ripening clearly involves cold reaction since between the conditions H and 60DAH, the apples are stored cold; yet this term is not observed if we only consider the differentially expressed sense genes.

We define the evolution of a probe as the expression difference between the two conditions with respect to a threshold of 1 log change. We observe that almost half of the probes associated with the term “response to cold” have the same evolution pattern: the sense expression level is nearly constant between the two conditions whereas the anti-sense expression level decreases (highlighted in Figure 6). This confirms that anti-sense data provide valuable information to study this process.

Figure 3: Genes of interest distribution.

Figure 4: Ontology of all the genes of interest.

Figure 5: Revealed-by-AS terms.

Figure 6: Evolution of probes associated with the term “response to cold".

Legends:
S: sense data
AS: anti-sense data