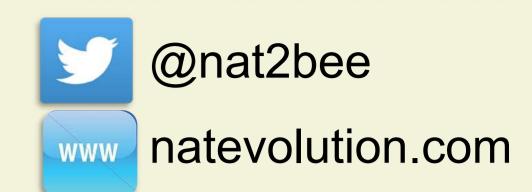
Unveiling the Expression Dynamics of Genes Involved in Bee Sociality

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Introduction

Bees are a great model to study the evolution of social behavior since in this group sociality seems to have evolved in several taxa independently. Thus a great diversity of social life styles was originated. The tribes Apini and Meliponini are comprised only by highly eusocial bee species, whereas various levels of sociality can be detected in other tribes, being the vast majority of bees indeed solitary. Although the molecular evolution of eusociality has been the subject of many studies, specific genetic changes involved in this behavior have not been completely understood. Fundamental questions about shared and derivate gene pathways involved in the different social systems are still open. Recently new sequencing technologies have allowed gene expression studies of non-model and model organisms in a deep and non-directional way, which is promising for evolutionary studies of complex behavioral traits. Herein, some of these new molecular tools were used to investigate the gene expression profile of different bee species performing distinct behaviors.

Material and Methods

High coverage RNASeq data (~50x) were generated for three bee species with distinct behaviors, Tetrapedia diversipes (solitary), Bombus terrestris (primitively eusocial) and Tetragonisca angustula (highly eusocial). Adults samples were from founders and nurses, and larvae from all developmental stages were used. Genes of interest were identified combining results from two designs: 1- Differential expression analyses (DE) in orthologous genes comparing the three species; 2- DE in orthologous genes comparing eusocial and solitary bees. MethylSeq data were also generated for the three species. RNASeq data from additional nine bee lineages were also obtained to validate the extension of the results (Figure 1). Genes of interest were analyzed through Gene Ontology enrichment analyses and Coexpression Network (both in R).

Results and Conclusions

- Comparing the three main species 787 "Life style" genes were selected;
- Life style genes are enriched to six biological processes (Table 1), being all of them already reported in previous behavioral studies;
- Life style genes are co-expressed in 7 network modules (Figure 2);
- According to DNA methylation comparisons between life style genes and other orthologous, methylation context is more relevant for sociality than methylation amount (Figure 3);
- Life style genes are good candidate genes to test in other spp. At least 328 could be found in all spp.

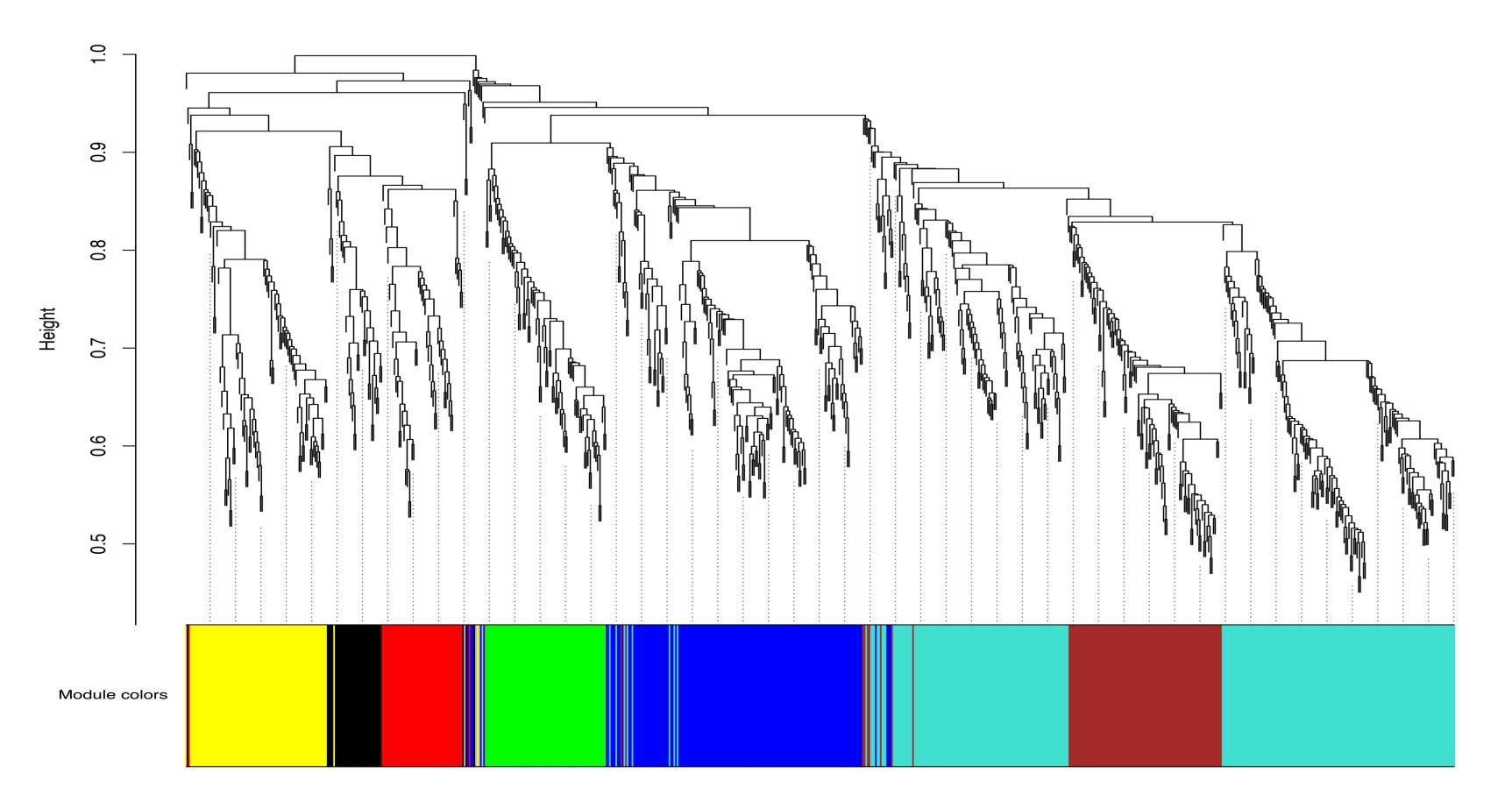


Figure 2 Modules of co-expression identified among the life style genes. Core genes in these networks are supposed to have great influence in the behavior and are great candidates for further validation studies.

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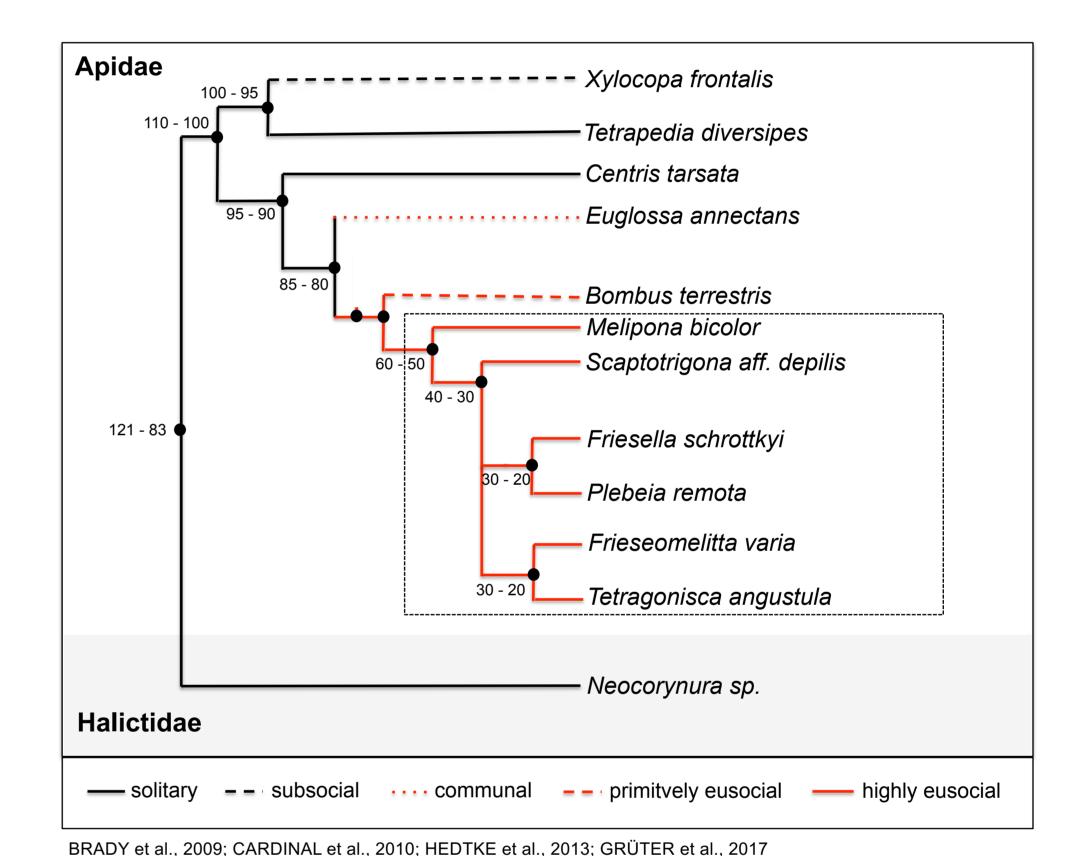


Figure 1 Species used for RNASeq analyses.

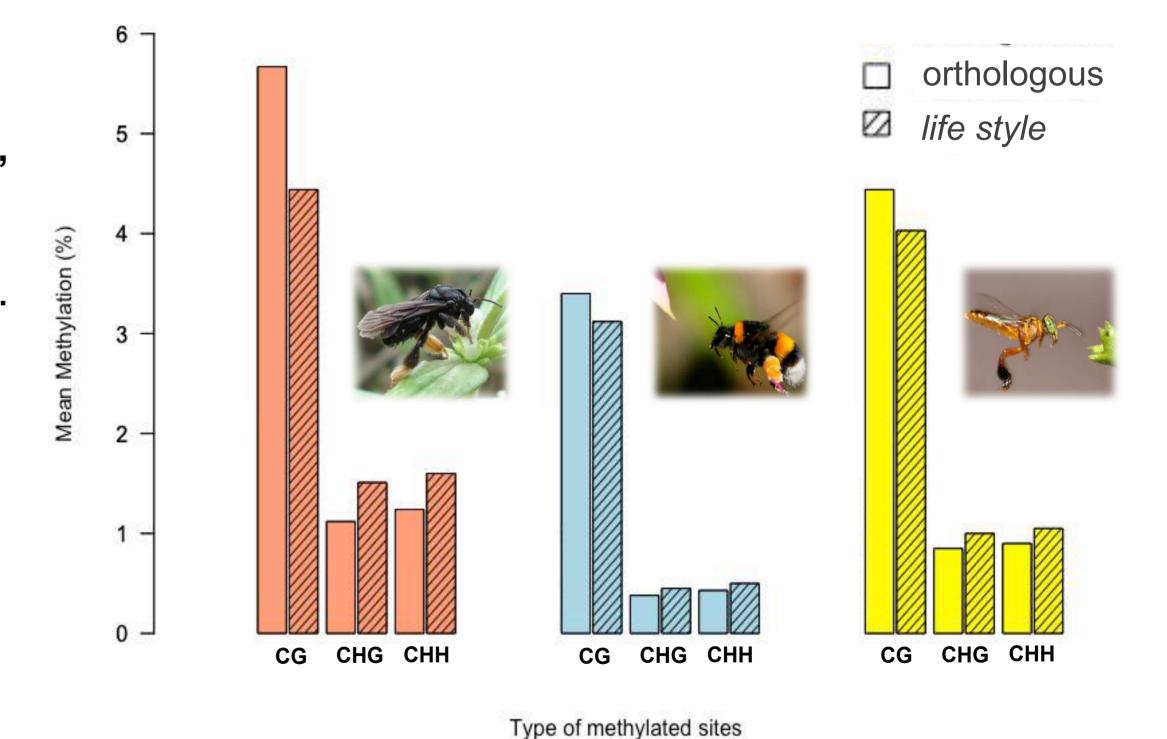


Figure 3 DNA methylation levels and context comparison between all orthologous and *life style* genes.

Table I. Biological processes enriched among the life styles genes when compared to all orthologous.

GO.ID	Term	classicFisher
GO:0060560	developmental growth involved in morphogenesis	0.0023
GO:1901136	carbohydrate derivative catabolic process	0.005
GO:0006518	peptide metabolic process	0.0068
GO:0043043	peptide biosynthetic process	0.0068
GO:0045292	mRNA cis splicing, via spliceosome	0.0078
GO:0043604	amide biosynthetic process	0.0098





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