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The complexity of *Gelsemium sempervirens* effects on human neuronal cells: a qualitative and functional evaluation

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Background: *Gelsemium s.* preparations contain a complex mixture of active compounds and is traditionally used in homeopathy and phytomedicine as remedy against anxiety, neuralgias and inflammation. As previously reported (Florence 2012, Barcelona 2013), we are currently assessing the genetic responsiveness of neuronal cells to low and high dilutions of *Gelsemium s.*

Methodology: We re-analyzed the complex "omics" dataset of cells, treated for 24 h with different dilutions of *Gelsemium s.,* using bioinformatic tools that help to discover functional roles of differentially expressed genes (DEG; drug vs. placebo), e.g. the Ingenuity Pathways Analysis software (Ingenuity Systems). In a second series of microarray experiments the neurons in activated or resting conditions were challenged with low dilutions of *Gelsemium s.* (2c and 3c) for short time (1-2 h) to investigate the early response of the cells. In parallel, we are also using RT-PCR array to explore further approaches to these drug effects.

Results: It could be observed a fine tuning of a core of genes that changed their expression level after 24h of exposure. The greatest changes in gene expression were found in 2c and 3c dilutions, but significant genes were changed also in higher dilutions. The DEG could be grouped according to their biological functions: the most significant group of genes shared the general function of cell-to-cell signalling and interaction; in this group, 29 genes are involved in activation of cells (Fisher p val= 2.3×10^{-8}). The genes in this group code for cytokines, growth factors and transmembrane receptors. Almost all those genes were downregulated by *Gelsemium s.*, suggesting that the drug may have an effect in the modulation of inflammatory reactions (Activation z-score= -2.3).

A group of 12 genes (Fisher p val=3.5×10^{-4}) are suggested as important in neurotransmission and include G-protein coupled receptors and ion channels. Another group of genes may have a role in nervous system development and function (22 genes) and contain transcription regulators, growth factors and neuropeptides that modulate axon guidance, neuronal differentiation and plasticity. Interesting observations could be done comparing the type of genes engaged by *Gelsemium s.* and the direction of the expression changes in the different conditions and times of exposure. RT-PCR array results focused on two neuron-specific genes which were down regulated in treated cells.

Conclusions: At a cellular level, the mechanism of action of *Gelsemium s.*, which is composed by many principles, appears to involve the modulation of several groups of genes. The use of a sensitive analytical approach, based on the detection of whole genome expression changes, allow us to gain new info about the complexity of its action.

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