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Alternative & Integrative Medicine

Review in 3 weeks

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GENOMIC PROGRAMS FOR NEURONAL APOPTOSIS

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NEURONAL APOPTOSIS

The *programmed cell death* or *apoptosis*, characterized by specific biochemical and morphological events, is a crucial phenomenon to determine the fate of neurons, both in physiological and in pathological conditions.



Elucidating the molecular mechanisms underlying neuronal apoptosis hence may contribute to our understanding of basic developmental biology and human neuropathology.

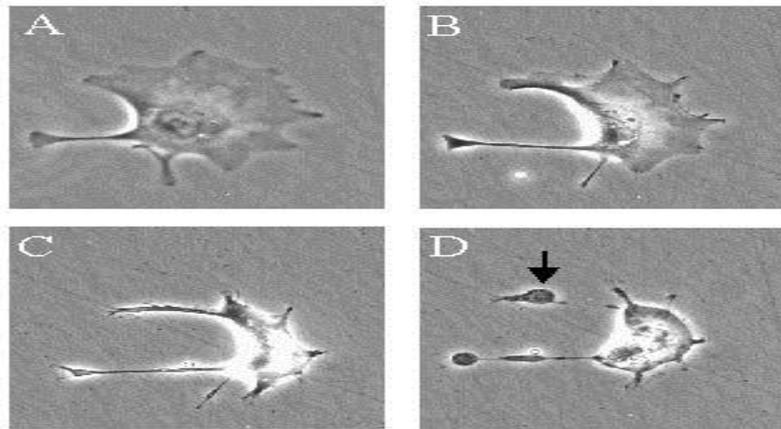


Fig.1 Apoptosis by microscopy

- ✓ Cytoplasm shrinks due to cleavage of nuclear lamins and actin (A)
- ✓ Chromatin is broken down as nucleus condenses (often "horse-shoe" shaped) (B)
- ✓ Cells shrink making an easy meal for phagocytes that are responsible for clearing the apoptotic cells (C)
- ✓ Cells undergo plasma membrane changes – move phosphatidylserine from inner to outer leaflet of the membrane; this attracts macrophages. At the end, membrane blebs appear (D). Small vesicles called apoptotic bodies are also sometimes observed (D, arrow).

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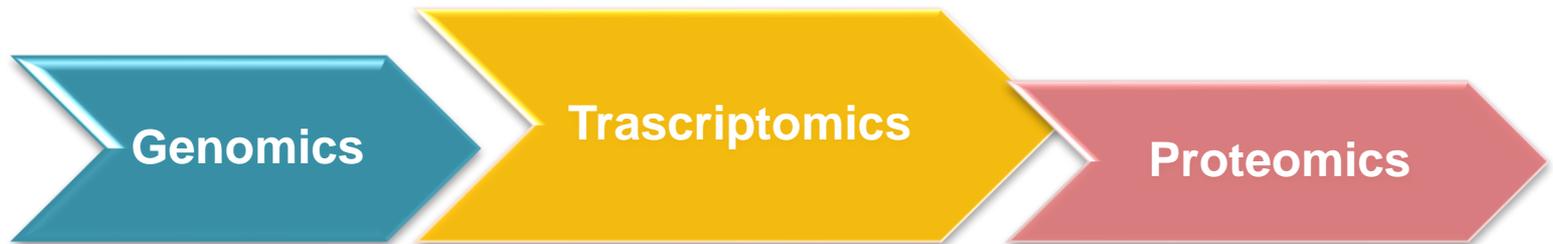


EXPRESSING THE STORY OF LIFE FROM THE GENETIC CODE

The analysis of the **TRASCRIPTOME**, defined as the complete set of transcripts and their relative levels of expression, is going to be a very important issue for the clinical implications



MICROARRAY TECHNOLOGY propelled functional genomics into the spotlight, because it allowed functional analysis of genome-wide differential RNA expression between different samples, states and cell types



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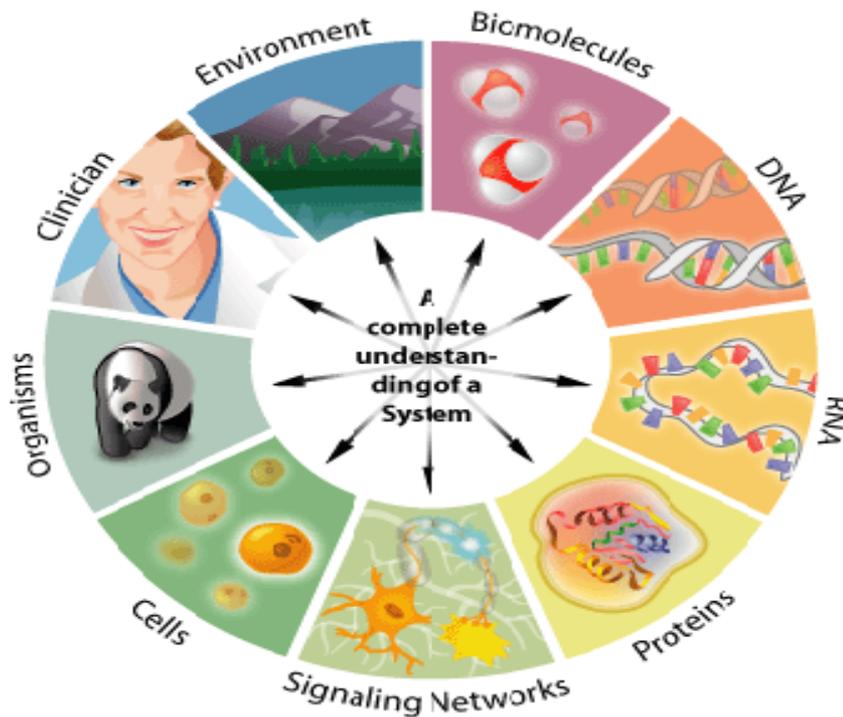
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A NEW APPROACH TO DECODING LIFE SYSTEM BIOLOGY



The **GOAL** is...

- ✓ *To decode biological meaning of gene expression changes*
- ✓ *To organize gene expression changes into molecular processes of a cell*
- ✓ *To promote the knowledge about the molecular mechanisms underlying neuronal apoptosis*

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...NEW WAYS OF THINKING

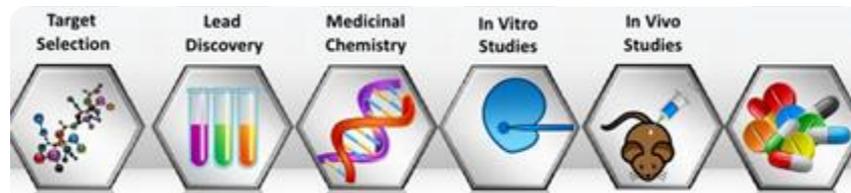
SYSTEM BIOLOGY FOR DRUG DISCOVERY



Leads to identify...

POTENTIAL DRUG TARGET

where intervention could make a difference in progression of neurodegenerative disease





CORE OF RESEARCH INTEREST

What makes CGNs decide to commit suicide?

✓ Withdrawal of serum and lowering of extracellular potassium from 25 to 5mM

On the basis of a previous study

What rescues apoptotic CGNs ?

✓ CGNs treatment by a maximal effective dose (100 nM) of Gastric inhibitory polypeptide (Gip)

WHAT ARE THE MOLECULAR PATHWAYS UNDERLYING CGN_s ESCUE AFTER GIP TREATMENT ?

Cent Nerv Syst Agents Med Chem, 2011 Sep 1;11(3):210-22.

Gastric inhibitory polypeptide and its receptor are expressed in the central nervous system and support neuronal survival.

Paratore S¹, Ciotti MT, Basille M, Vaudry D, Gentile A, Parenti R, Calissano P, Cavallaro S.



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“Transcriptional analysis of apoptotic cerebellar granule neurons following rescue by gastric inhibitory polypeptide.”

Maino B, Ciotti MT, Calissano P, Cavallaro S

In our study

✓ we conducted whole-genome expression profiling to obtain a comprehensive view of the **previously unknown transcriptional program** underlying the rescue effect of Gip in CGN.

✓ By using DNA microarray technology, we identified 65 genes, we named **survival related genes**, whose expression is significantly de-regulated following Gip treatment (Fig.2).



We rendered a portrait of the molecular framework involved in CGN survival program (Fig. 3)

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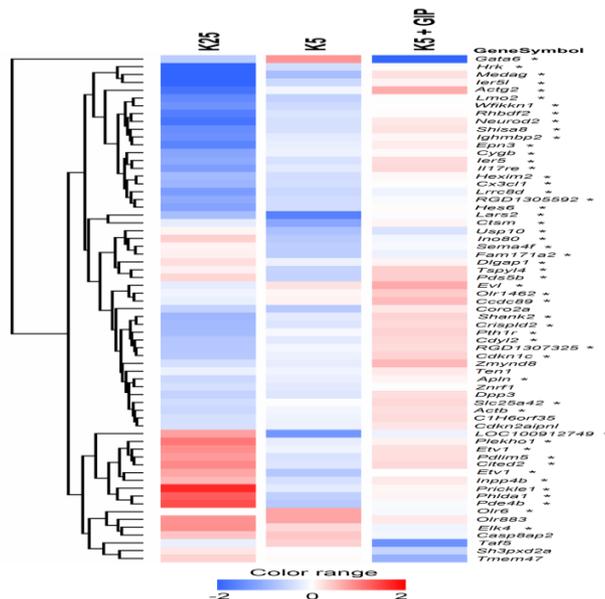
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HIERARCHICAL CLUSTERING of genes differently expressed in CGNs treated with Gip during the pre- commitment of apoptosis



✓ Data are presented in a **matrix format**: each row is equivalent to a single gene and each column is equivalent to one of the three different experimental conditions (K25, K5, K5 + Gip).

✓ The **color of the corresponding cell** in the matrix indicates the averaged normalized intensity from replicates. Red, blue and white respectively represent transcript levels below, equal or above the median abundance across all conditions.

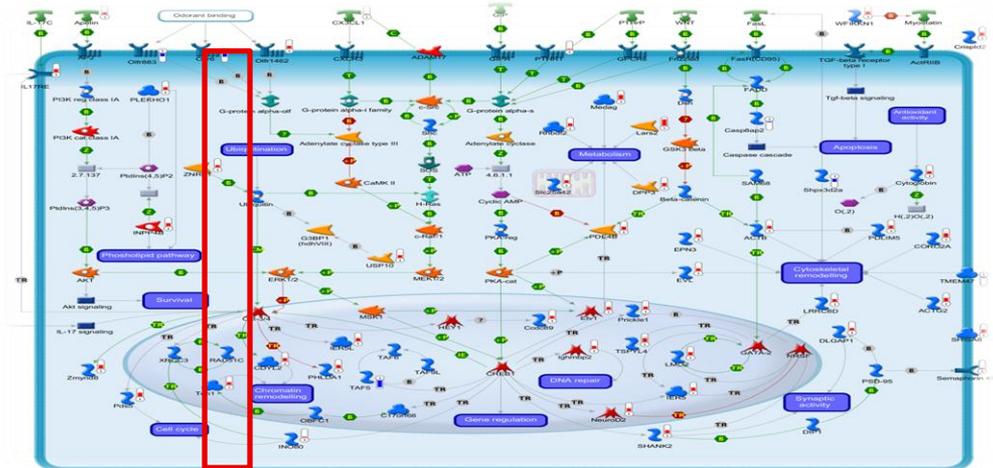
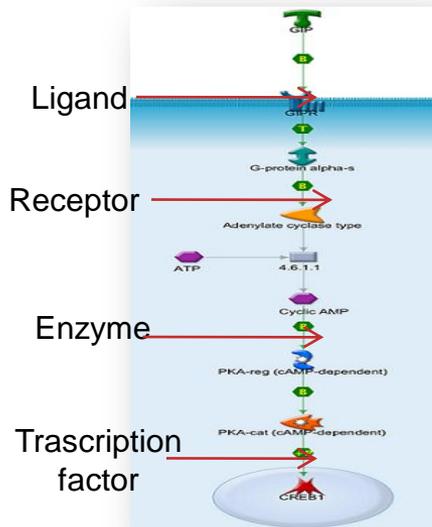
✓ **Color intensity** reflects the magnitude of the deviation from the median (see scale below).





GIP-RESCUE PATHWAY IN CGNs

The survival effects of Gip are initiated by a G-protein-coupled receptor and activate a variety of intracellular second messengers. These signaling pathways converge into the nucleus and regulate a transcriptional program governing neuronal cell life.





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CONCLUSION



Our results reveal:

- ✓The existence of a previously unknown transcriptional program associated to neuronal survival.

Our results form:

- ✓The basis for further functional analyses and pharmacological exploitation to identify neuroprotective drugs.

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THE FUTURE ?

Pharmacological exploitation of potential targets will help to determine their cause-relationship and identify new clues for neuroprotective drugs.



Further studies will be interesting for understanding complex biological systems as well as DRUG DISCOVERY and NEURODISEASE DIAGNOSIS.

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