Research Interests

Molecular mechanisms of G protein-coupled receptor signalling

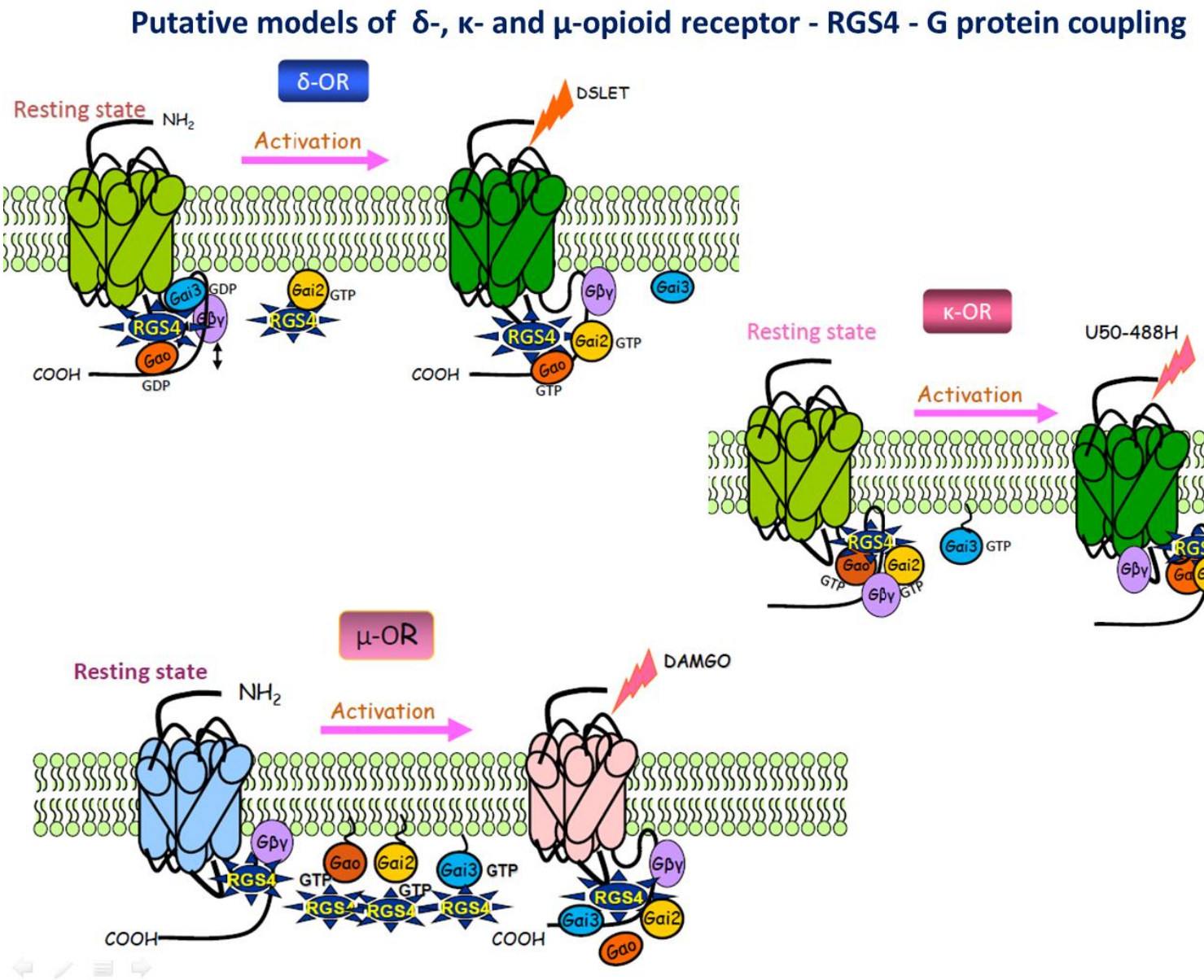
Georgoussi's laboratory studies the molecular mechanisms by which cellular signaling regulates cell function and how signaling mechanisms affect human health. Specifically, we study G-protein signaling cascades and their dynamic regulation by activated receptors, RGS (Regulator of G-protein Signaling) proteins and other factors. We have a long-standing interest in the family opioid receptors (μ, δ and κ), which are implicated in pain mechanisms and addiction processes as well as in neuronal cell survival and differentiation.

Dissection of opioid receptor signaling

We are exploring the ability of RGS proteins to attenuate GPCR signaling. In this respect, we have demonstrated that RGS4 and RGS2 proteins interact with δ- μ- and κ-opioid receptors to confer selectivity for Gi/o proteins (Figure 1) and thus modulate negatively their signaling by affecting adenylyl cyclase activity, MAP kinase phosphorylation and agonist induced internalization in a differential manner for each receptor subtype (Georgoussi et al., 2006 ; Le ontiadis et al., 2009
; Papakonstantinou et al., 2015;
http://en.wikipedia.org/wiki/CCG-4986
.

Parallel studies have shown that spinophilin, a ubiquitous multidomain scaffold protein, which occurs abundantly in dendritic spines, interacts also with the μ- and δ-opioid receptors and forms a signaling complex with Gi/o and RGS4 proteins to positively regulate μ- and δ-opioid receptor signaling (Fourla et.,2012 ; Georgoussi et al., 2012). Current studies focusing on neuronal cells attempt to establish whether spinophilin i) is regulated by opioid administration and ii) is implicated in neuronal development and nervous system disorders in RGS4
-/-
knockout animals.

Figure 1

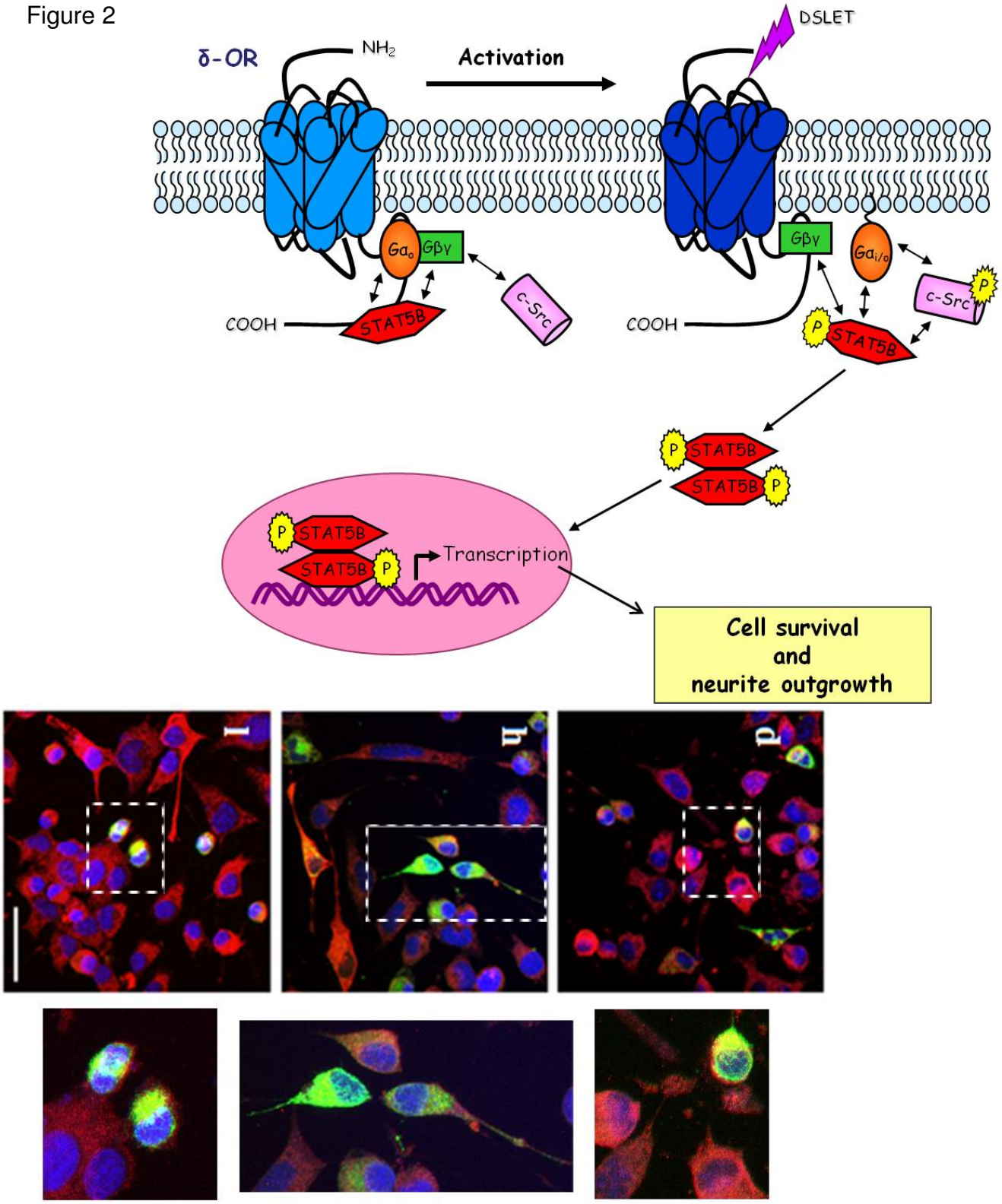


Fuctional analysis of genes whose expression is altered by opioid administration.

A landmark of our research has been the finding that members of the Signal Transducers and Activators of Transcription (STATs) such as STAT5A/B directly interact with the δ- and μ-opioid receptors and are phosphorylated in response to opioids in neuronal cells (Mazar akou and Georgoussi, 2005 ; Georgoussi et al., 2012 Georganta et al., 2010 ; Georgoussi et al., 2012). This interaction is mediated by a multi-component signalling complex – a signalosome - a platform composed of STAT5B, c-Src kinase and selective Gαi/o, Gβγ subunits (Figure 2),

which assemble on using the C-terminus of the δ-opioid receptor .

Figure 2



A putative signalling pathway of the δ-opioid receptor-phosphorylated-STAT5B-G protein complex, implicated in neuronal differentiation. Confocal images of cells expressing the δ-OR

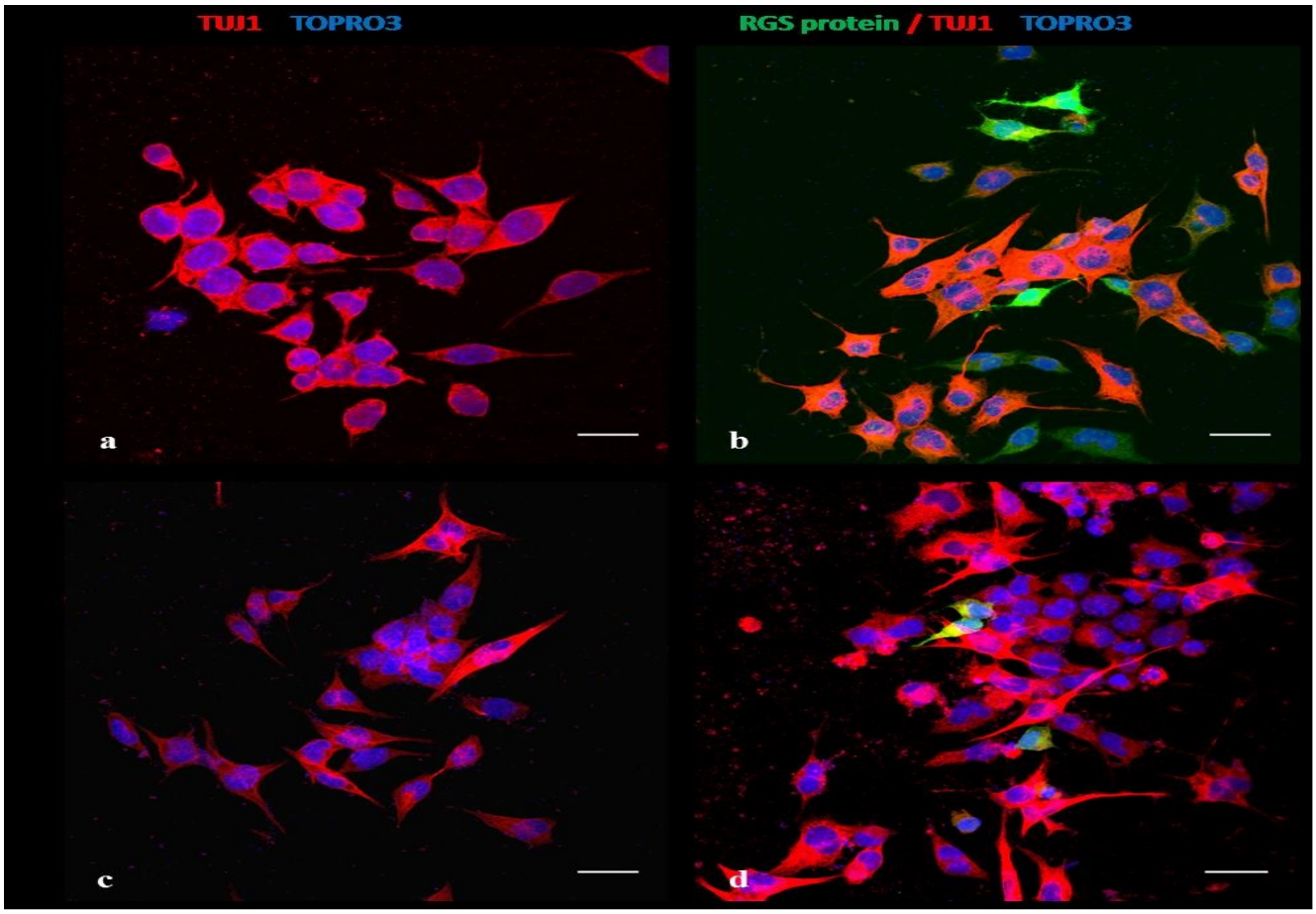
(green) indicate an increased neurite length after DSLET administration (Tuj-1-labelling yellow) (
Mazarakou and Georgoussi 2005
;
Georganta et al., 2010
),
Georganta et al., 2013
)

This dynamic protein complex is implicated in cell survival, neurite outgrowth and differentiation following opioid administration to Neuro-2A cells and primary neuronal cortical cultures Georganta et al., 2010
;
Georganta et al., 2013
; Pallaki et al., manuscript submitted).

Based on such findings we are currently dissecting in more detail the role of STAT5B and RGS4 in neurogenesis in the context of opioid receptor-mediated function using RGS4$^{-/-}$ mice. Our studies on neuronal stem cells from such mice, suggest that the interplay between G and RGS4 proteins with STAT5 is important for neuronal differentiation and outgrowth (Figure 3). Our current goal is to define whether RGS4-STAT5B coupling is modulated by opioids and constitutes an underlying factor for neuronal and neuroinflammatory diseases.

Figure 3.

RGS proteins alter neurite outgrowth mediated by the activated δ-OR



New therapetical agents: Identification and pharmacological characterization

As a member of the EU- network " NORMOLIFE " we have characterized the pharmacological properties of new opioid compounds with potent
in vivo
analgesic effects using cell based assays. (
EKT
,
Pasquinucci et al., 2010
;
Vandormael et al., 2011
;
Pasquinucci et al., 2012
). Finally, in collaboration with the laboratory of
Insect Molecular Genetics & Biotechnology
, we develop reporter gene and/or second messenger cell-based high throughput screening platforms to monitor cellular responses for identification of lead compounds with relevant pharmaceutical potential in plant extracts.(Swewers et al. 2005; Douris et al. 2006; Tsitoura et al. 2010; Greek Patent office 20040100397)