

## Apoptosis-Assay after Staurosporin treatment in Organotypic Hippocampal Slice Cultures (OHCs)

Compounds of interest can be assessed *in vitro* for their

- Neuroprotective properties after Staurosporine treatment (Propidium iodide uptake)
- Antiapoptotic properties after Staurosporine treatment (Detection of active Caspase 3)

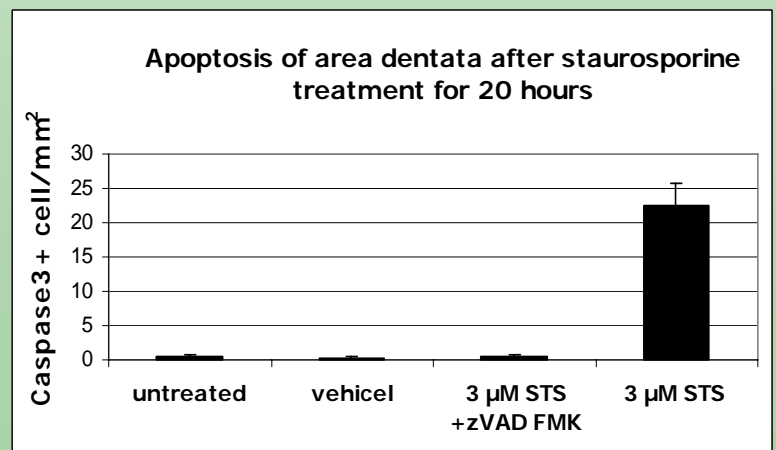
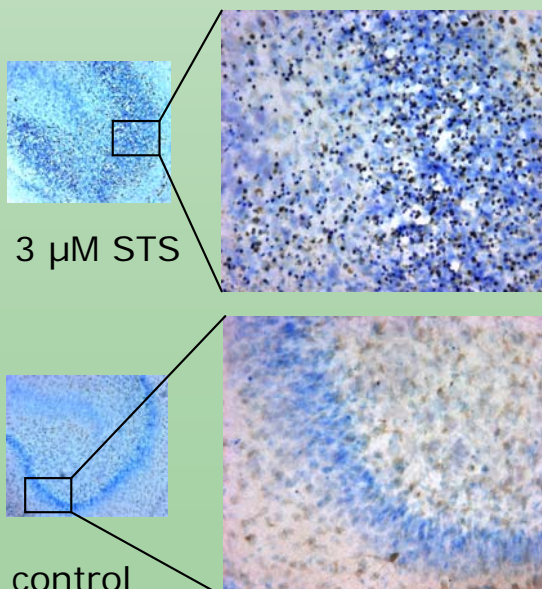
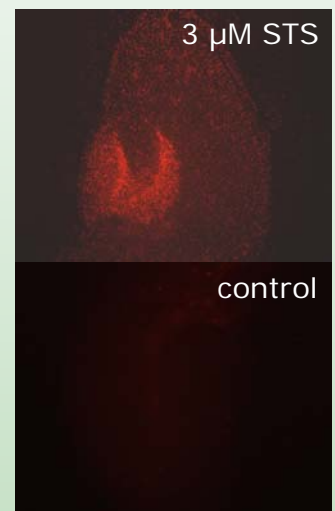
### Models

#### Cell degeneration assay

Neuronal cell death after Staurosporine treatment in serum free OHCs is assessed by densitometric measurement of propidium iodide (PI) uptake. Fluorescent images will be acquired semi-automatised (Nikon motorised stage; LUCIA software) and analysed by densitometry to quantify necrotic cell death (LUCIA Image analysis software). Cell death is expressed as % PI-uptake of the respective regions of interest, here the area dentata (AD).

#### Apoptosis/Necrosis assay

A combination of Caspase 3- Active immunohistochemistry and Nissel staining is used to identify apoptotic and necrotic neurons. The number of caspase-3 active positive cells is determined by transmission light microscopy.



**Fig.1** Quantification of apoptotic cell death under serum-free conditions in area dentata after staurosporine treatment. (n=6/bar). zVAD FMK = pancaspase inhibitor