

## Meeting Report

**18th Euroconference on Apoptosis in Ghent, Belgium,  
1–4 September 2010**W Declercq<sup>\*1,2</sup>, P Agostinis<sup>3</sup> and P Vandenabeele<sup>1,2</sup>*Cell Death and Differentiation* (2011) 18, 1540–1542; doi:10.1038/cdd.2011.76; published online 3 June 2011

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The 18th Euroconference on Apoptosis was held at Ghent University, Ghent, Belgium. With its first-rate scientific program, the meeting attracted 283 participants from 36 countries. The participants represented an excellent mixture of PhD students, postdocs and leading scientists. This was evidenced by the many lively discussions during the lectures and poster sessions. We look forward to welcome you at the 19th Euroconference on Apoptosis 'Metabolism, Epigenetics and Cell Death' and 8th Training course on 'Concepts and Methods in Programmed Cell Death' on September 14–17 2011, Stockholm, Sweden. For updated information have a look at <http://www.ecdo.eu>.

During the last years, the Euroconferences were preceded by a training course on 'Concepts and Methods in Programmed Cell Death'. This training course intends to review several evolving concepts or state-of-the-art methodologies in cell death research. Markus Rehm (Dublin, Ireland), Francis Blankenberg (Stanford, CA, USA), Laurence Calzone (Paris, France), Lorenzo Galluzzi (Villejuif, France) and Klaus-Michael Debatin (Ulm, Germany) gave excellent talks on imaging of subcellular events during cell death, *in vivo* imaging of cell death, systems biology approaches, *in vitro* cell death analysis and perspectives for therapeutic targeting of apoptosis, respectively. All training course lectures are publically available at the ECDO website (<http://www.ecdo.eu/gent2010/index.html>).

The ECDO keynote lecture by Guy Salvesen (La Jolla, CA, USA) was an excellent up-to-date overview on caspase activation, specificity and substrates. Importantly, he discussed a novel caspase-8 activation mechanism involving cFLIP. This agrees with the similarity in the phenotypes of caspase-8 and cFLIP KO mice.

**Molecular Mechanisms of Cell Death**

Andreas Villunger (Innsbruck, Austria) reported on the *in vivo* relevance of the PIDDosome, a multiprotein complex composed of PIDD, RAIDD and pro-caspase-2. He reported

that caspase-2 has a PIDDosome-independent tumor suppressor function. Rosario Rizzuto (Padova, Italy) discussed how signaling pathways involved in autophagy and apoptosis affect the different VDAC isoforms, thereby modulating mitochondrial  $Ca^{2+}$  homeostasis. Delphine Merino (Melbourne, Australia) found that the level of pro-apoptotic BH3-only protein Bim is the major determinant of the sensitivity of Bcl-2-overexpressing lymphoid cells to the BH3 mimetic ABT-737. Donat Kögel (Frankfurt, Germany) reported that the pan-Bcl-2 inhibitor (gossypol) efficiently induced cell death in glioma cells, but not in primary astrocytes.

**Non-apoptotic Roles of Death Mediators**

David Wallach (Rehovot, Israel) presented new insights into the skin inflammation in caspase-8<sup>EKO</sup> (epidermis-specific knock-out) mice. He also showed that caspase-8 associates with the RIG-I helicase complex in Sendai virus-stimulated cells and cleaves RIP1. A role for ubiquitination in the proteolytic sensitivity of RIP1 was suggested. Manolis Pasparakis (Cologne, Germany) reported that FADD<sup>EKO</sup> mice also develop skin inflammation. FADD<sup>EKO</sup> mice could not be rescued by crossing them to p38<sup>EKO</sup>, MyD88<sup>-/-</sup>, IL1R<sup>-/-</sup> or TNFR1<sup>-/-</sup> mice, however, the appearance of the skin phenotype was delayed. Thirumala Kanneganti (Memphis, TN, USA) showed that NLPR3<sup>-/-</sup>, caspase-1<sup>-/-</sup> and ASC<sup>-/-</sup> mice are more prone to develop colitis and colon tumors upon challenge with DSS. Apparently, the phenotype was dependent on IL-18 and IFN $\gamma$  levels. Geert van Loo (Ghent, Belgium) generated liver- and intestine-specific A20 (an ubiquitinating editing enzyme) KO mice and found that the sensitivity to TNF-induced cell death was increased in these organs. Sandrine Jouan-Lanhouet (Rennes, France) showed that a reduction in the extracellular pH can modify the sensitivity of human cancer cells to TRAIL-induced RIP1- and RIP3-dependent necrosis.

<sup>1</sup>Molecular Signaling and Cell Death Unit, Department for Molecular Biomedical Research, VIB, Ghent, Belgium; <sup>2</sup>Department of Biomedical Molecular Biology, Ghent University, Ghent, Belgium and <sup>3</sup>Cell Death Research & Therapy Unit, Department for Molecular Cell Biology, Catholic University of Leuven, Leuven, Belgium  
\*Corresponding author: W Declercq, Department of Biomedical Molecular Biology, Faculty of Sciences, VIB, Ghent University, Technologiepark 927, Gent B-9052, Belgium. Tel: + 329 331 3760; Fax: + 329 331 3609; E-mail: [wim.declercq@dmbr.ugent.be](mailto:wim.declercq@dmbr.ugent.be)

### Ubiquitination Control of Cell Death Signaling

Pascal Meier (London, UK) found that etoposide can induce the formation of a RIP1-containing complex (the ripoptosome) that can signal to cell death. Combining etoposide with an IAP inhibitor (Smac mimetic) caused RIP1-dependent apoptosis or necrosis, depending on the cellular condition. John Silke (Victoria, Australia) presented that caspase and RIP1 inhibition prevented compound A (a potential anti-cancer drug)-induced cell death. RIP3 knockdown did not affect the cytotoxic effect, suggesting that RIP1 can act independently from RIP3. Phil Barker (Montreal, Quebec, Canada) reported that cIAPs are also involved NF- $\kappa$ B activation and in inflammation. Consequently, NOD2 prestimulation by MDP protects against DSS-induced colitis, but not in cIAP KO mice. Emmanuel Dejardin (Liège, Belgium) provided evidence that activated NIK promoted the formation of the TNF DISC and the activation of RIP1, which led to induction of cell death. Henning Walczak (London, UK) reported that Sharpin together with HOIL-1 and HOIP constitutes the linear ubiquitin chain assembly complex, which is recruited to receptor complexes by HOIP. Sharpin-deficient MEFs exhibited significantly reduced TNF-induced gene activation and were sensitized to TNF-induced apoptosis. Nele Vanlangenakker (Ghent, Belgium) showed that IAP depleted cells are greatly sensitized to TNF-induced necrosis but not to Fas-, poly(I:C)- or oxidative stress-induced necrosis. Similarly, lowering TAK1 levels or inhibiting its kinase activity sensitized cells to TNF-induced RIP1-dependent necrosis.

### Autophagy in Physiology and Pathology

Frank Madeo (Graz, Austria) reported that exogenous supply of spermidine prolongs the life span of several model organisms (yeast, nematodes, flies) in an autophagy-dependent way, and significantly reduces age-related oxidative protein damage in mice. Ana Maria Cuervo (New York, NY, USA) showed that chaperone-mediated autophagy (CMA) is important for the removal of oxidized-protein aggregates. Mice with increased levels of CMA were more resistant to cellular stress. Joel Beaudouin (Heidelberg, Germany) reported that caspase-3 activity appears abruptly in apoptotic cells, including the nucleus, just before formation of apoptotic bodies. In contrast, caspase-8 activity is mostly concentrated at the plasma membrane. Nieves Peltzer (Lausanne, Switzerland) showed that a caspase cleavage fragment of RasGAP protects cells against death in a Ras/PI3K/Akt-dependent manner. She found that keratinocytes from uncleavable RasGAP knock-in mice are more sensitive to UVB-induced apoptosis.

### Targeting Cell Death Processes in Cancer

Cosima Baldari (Siena, Italy) found that p66Shc, the longest of the three ShcA isoforms, can act as a negative regulator of survival signals from the T-cell and B-cell antigen receptors and as a promoter of apoptosis. Judy Liebermann (Boston, MA, USA) identified 44 RNA-binding proteins as novel granzyme B substrates. However, the functional

consequences of the cleavage of these targets are not clear. Marion MacFarlane (Leicester, UK) reported that although most transformed cell lines are sensitive to TRAIL, most primary tumors are TRAIL-resistant. She found that tumor cells are sensitized to apoptosis by combining chemotherapeutics with selective triggering of TRAIL-R1. Peter Krammer (Heidelberg, Germany) showed that annexin 1 suppressed not only the secretion of inflammatory cytokines but also the expression of co-stimulatory surface molecules of dendritic cells (DC) by antagonizing Toll-like receptor signaling pathways. Kris Gevaert (Ghent, Belgium) combined metabolic proteome labeling with targeted proteomics in a time-kinetic setup to distinguish physiologically relevant protease substrates from bystanders. They identified 18 efficiently cleaved human granzyme B substrates. Alexander Pintzas (Athens, Greece) reported that MYC, RAS, K-RAS and BRAF can sensitize for TRAIL-induced apoptosis due to overexpression or redistribution of TRAIL-R1 and 2. The mechanisms of this sensitization involve RAF/MAPK and PI3K pathways. Tom Cotter (Cork, Ireland) found that FGF protects the retina from apoptosis by generation of low levels of hydrogen peroxide by Nox2 and Nox4, leading to the inactivation of PP2a, a negative regulator of the Akt pathway. Sven Horke (Mainz, Germany) presented that paraoxonase-2, which is frequently overexpressed in various tumors, prevented mitochondrial-mediated cell death and caspase activation using CHOP-dependent and -independent mechanisms. Catharina Svanborg (Lund, Sweden) reported that *in vitro* and *in vivo* experiments indicate that HAMLET, a complex consisting of partially unfolded  $\alpha$ -lactalbumin and oleic acid, is cytotoxic for tumor cells and immature cells, but not for differentiated cells.

### The ECDO Honorary Lecture

Guido Kroemer (Villejuif, France) reported that immunogenic cancer cell death, elicited by radiotherapy and some chemotherapeutic agents, is crucial for successful tumor therapy. The released or surface-exposed DAMPS (damage associated molecular patterns) activate receptors expressed by dendritic cells (TLRs, P2X7 purinergic receptors) to allow presentation of tumor antigens to T cells, which results in an effective anti-tumor immune response. These findings have important consequences for anti-tumor therapeutic strategies.

### Caspase-independent Cell Death

Martin Krönke (Cologne, Germany) showed that ROS induction by TNFR1, TLR2, TLR4 and TLR9 depends on Nox1 and 2, and riboflavin kinase. TNF-induced cytochrome c release, PARP cleavage and cell death was remarkably reduced in Nox-deficient HeLa cells. Francis Chan (Worcester, MA, USA) identified that phosphorylation at S161 was important for the necrosis-inducing activity of RIP1. Interestingly, a mouse RIP3 S204A mutant was able to block TNF-induced necrosis, whereas the phosphorylation-mimicking S204D mutant led to RIP1-independent cell death. Chan also showed that knockdown of CYLD prevents TNF-induced necrosis, whereas A20 knockdown promotes cell death. Cristina Claudia Mihalache (Bern, Switzerland) presented

that neutrophils demonstrating vacuolization undergo rapid cell death depending on RIP1 and papain family protease(s), but not on caspases. Kelly Jean Thomas (Grand Junction, CO, USA) reported that the apoptotic resistance of highly tumorigenic cell lines is correlated with impeded mitochondrial fission and mitophagy.

### ***In Vivo* Cell Death in Health and Disease**

Edward Mocarski (Atlanta, GA, USA) reported that RIP3 controls virus-induced necrosis as well as death receptor-induced necroptosis. However, unlike necroptosis, the viral death pathway is RIP1-independent. In addition, he found that the embryonic lethality of caspase 8 deficiency was rescued in a RIP3-deficient background. Shigekazu Nagata (Kyoto, Japan) previously identified MFG-E8 and Tim4, which are important for PS-mediated engulfment of dying cells. However, in contrast to MFG-E8 deficient macrophages, Tim4-deficient macrophages cannot engulf dead cells. He reported that living PS-exposing cells were not engulfed. This suggests the existence of additional 'eat me' signals or efficient 'don't eat me' signals. Caetano Reis e Sousa

(London, UK) showed that the necrotic DAMPs-sensing C-type lectin DNGR-1 regulates CD8<sup>+</sup> T-cell cross-priming by cross-presenting dead cell-associated antigens, both in mice and humans. Abhishek Garg (Leuven, Belgium) found that hypericin-mediated photodynamic therapy induced surface exposure of calreticulin, leading to immunogenic cell death, was dependent on PERK, PI3K and BAX/BAK.

### **Conflict of Interest**

The authors declare no conflict of interest.

**Acknowledgements.** We would like to thank Veronique Vandevoorde and Jelle Verspurten for the excellent administrative organization of the meeting in Ghent in all its aspects. Furthermore we are indebted to the people of the Vandenebeele and Agostinis units for their practical help. We also thank the city of Ghent, Ghent University and VIB for their contribution to an interesting opening session. Especially we would like to thank the Faculty of Sciences and FWO for substantial financial support. Last, but not the least, we thank the members of the scientific committee, speakers, poster presenters and participants for making an interesting and vivid meeting possible.