

Melanoma Group



María S. Soengas

Group Leader

María S. Soengas was born in Agolada, Pontevedra (Spain), in 1968. She embarked upon her scientific career first as an undergraduate student at the *Universidad de La Coruña* and later at the *Universidad Autónoma de Madrid*, where she graduated in Molecular Biology. There she received her PhD with First Class Honours for her studies on molecular mechanisms of DNA replication at the laboratory of M. Salas, *Centro de Biología Molecular "Severo Ochoa"*.

In 1997 Soengas moved to the S. Lowe's group at the Cold Spring Harbor Laboratory, New York (USA), where she assessed the role of apoptosis as a tumour suppressor mechanism, with special focus on melanoma. She then joined the Department of Dermatology at the University of Michigan in 2002 to develop a basic research programme in Melanoma. Her group defined new molecular mechanisms underlying human melanoma initiation, progression and chemoresistance.

Since 2008, Soengas leads the Melanoma Group at the CNIO. The main objective of her team is to translate basic research in melanoma to the clinic by identifying novel markers of this disease and targets for drug development.

Soengas has been recipient of fellowships and awards from both the Human Frontiers in Science Programme and the Leukemia and Lymphoma Society of America. She has also received a Life Science Biomedical Scholar Award from the University of Michigan, the Diana Ashby Young Investigator Award from the Society for Melanoma Research as well as Career Development Awards from the American Dermatology Foundation, the Elsa V. Pardee Foundation and the V Foundation for Cancer Research. She has also been honoured with the *Premio M. Josefa Wonenburger* from the *Xunta de Galicia*.

Summary

Malignant melanoma is a paradigm of aggressive and chemoresistant cancers. The overall survival of patients with metastatic disease is typically limited to 6-10 months and has not significantly improved since the 1970s. The melanoma field is therefore in need of progression markers and improved anticancer strategies.

The long term goal of our Group is to identify molecular mechanisms involved in melanoma initiation, progression and drug resistance. Our experimental systems include a combination of genetic and functional analyses in cell lines, mouse models and human tissue biopsies. These studies are performed in the context of a multidisciplinary consortium of specialists in different areas of biology, chemistry, pharmacy, nanotechnology, molecular imaging, dermatopathology and clinical oncology. We also collaborate with biotechnology companies to translate our drug discovery results into the clinic.

Strategic Goals

- Identify mechanisms of suppression of melanoma initiation
- Define the contribution of stress response programmes (apoptosis, senescence and autophagy) to melanoma progression and metastasis
- Address mechanisms of melanoma chemoresistance using genetic and pharmacological approaches
- Develop new melanoma models for a more physiological analysis and validation of targeted therapies



Staff scientist: David Olmeda. **Post-doctoral fellows:** Agnieszka Checinska, María García (since November), Lisa Osterloh, Erica Riveiro (until June), David Sáenz and Damiá Tormo (until March). **Graduate students:** Direna Alonso, Metehan Cifdaloz (since October) and Eva Pérez. **Technicians:** Tonantzin G. Calvo and Estela Cañón.

Highlights

Melanoma is an increasingly common solid tumour accounting for 80% of skin cancer-related deaths. There is consequently a pressing need for new melanoma diagnostic markers and anticancer agents. Our Group's main objective is to identify the key causal mechanisms involved in melanoma initiation and maintenance to ultimately obtain a more rational approach to treatment. We are particularly interested in genes whose expression and function is differentially regulated in benign melanocytic lesions (nevi) and in malignant melanomas.

Mechanisms of suppression of melanoma initiation

Current statistics indicate that 1 in 58 individuals will develop melanoma during their lifetime. This rate would be considerably higher if potent tumour suppressors were not in place. Fair skinned individuals bear an average of 10 to 40 nevi (moles) which are constituted by melanocytes containing pro-oncogenic mutations in *BRAF* or *NRAS*. However, less than 1/1000 nevi develop into melanoma. Our research focuses on the identification of genetic and epigenetic factors that prevent the proliferation of nevus cells, and how these mechanisms of protection are disengaged to promote melanoma progression and chemoresistance.

We have previously identified new tumour suppressors in melanocytes associated with the endoplasmic reticulum. We have

also reported that the activation of stress programmes by *BRAF* and *RAS* oncogenes can also extend to human melanomas. In recent studies integrating cDNA arrays with functional assays, we have now identified a selective induction of Early Growth Response factors (EGR-1, -2, -4) in response to aberrant activation of *HRAS* and *NRAS*. EGR proteins have been described to have pro- or anti-oncogenic roles, depending on the cell type and environmental conditions; however, they have not yet been linked to the senescence of skin cells. Experiments are currently ongoing in our laboratory to define the mechanisms underlying the specific role of EGR proteins as melanoma suppressors.

Molecular mechanisms of melanoma maintenance: identification of tumour markers

Melanoma cells accumulate a plethora of genetic and epigenetic defects in multiple signalling cascades. This knowledge is essential in our understanding of the mechanisms of melanoma progression but has not yet provided suitable molecular markers. Using comparative genomic hybridisation (GSK), spectral karyotyping (SKY) and histochemical analyses in tissue microarrays we found a strict melanoma-associated expression of chromatin binding factor DEK, when compared with minimum levels in nevus cells (Figure 1). Moreover, we demonstrated novel roles of DEK in the control of melanoma cell proliferation

and drug resistance. These results suggest that DEK may be a useful tumour marker and cooperate with other functional defects in apoptotic programmes recently reported by our Group.

Further comparative studies of benign and malignant melanocytic biopsies have uncovered a tumour-restricted accumulation of p62/sequestosome protein (Figure 1). In other cellular systems p62 acts as a link between autophagy and proteasome-dependent pathways. In melanoma cells however, targeted depletion of p62 does not block either of these processes but rather affects melanoma cell proliferation. Nevertheless, there is no available information thus far on the regulation and function of p62 in melanoma. We therefore expect that our studies will open new avenues of research into this aggressive disease.

Genetic and pharmacological analyses of survival signals in melanoma: identification of new therapeutic strategies

Given the persistent resistance of melanoma cells to standard chemotherapeutic agents, we performed a

screening to identify alternative cell death inducers. We focused on bioavailable anticancer agents (i.e. compounds administered systemically *in vivo* to control internal metastasis). Among the most potent and selective killers of melanoma cells identified were nanocomplexes of synthetic dsRNA and polycationic carriers (herein referred as BO-110). BO-110 was found to have a unique capacity of inducing massive changes in the transcriptome, mobilising the endosomal machinery and effectively engaging the apoptotic machinery in tumour cells. All these activities of BO-110 ultimately converge into an effective tumour self-digestion process, resulting in the inhibition of tumour growth in various animal models (Figure 2A-C). These results have attracted a great deal of attention in both the basic and clinical research settings.

The clinical relevance of our data led to the creation of *Bioncotech Therapeutics*, a small biotechnology company whose main objective is the clinical development of dsRNA-based anticancer therapies. Furthermore, to improve the uptake and targeted delivery of BO-110 to clinically relevant systems, we have established a Consortium between

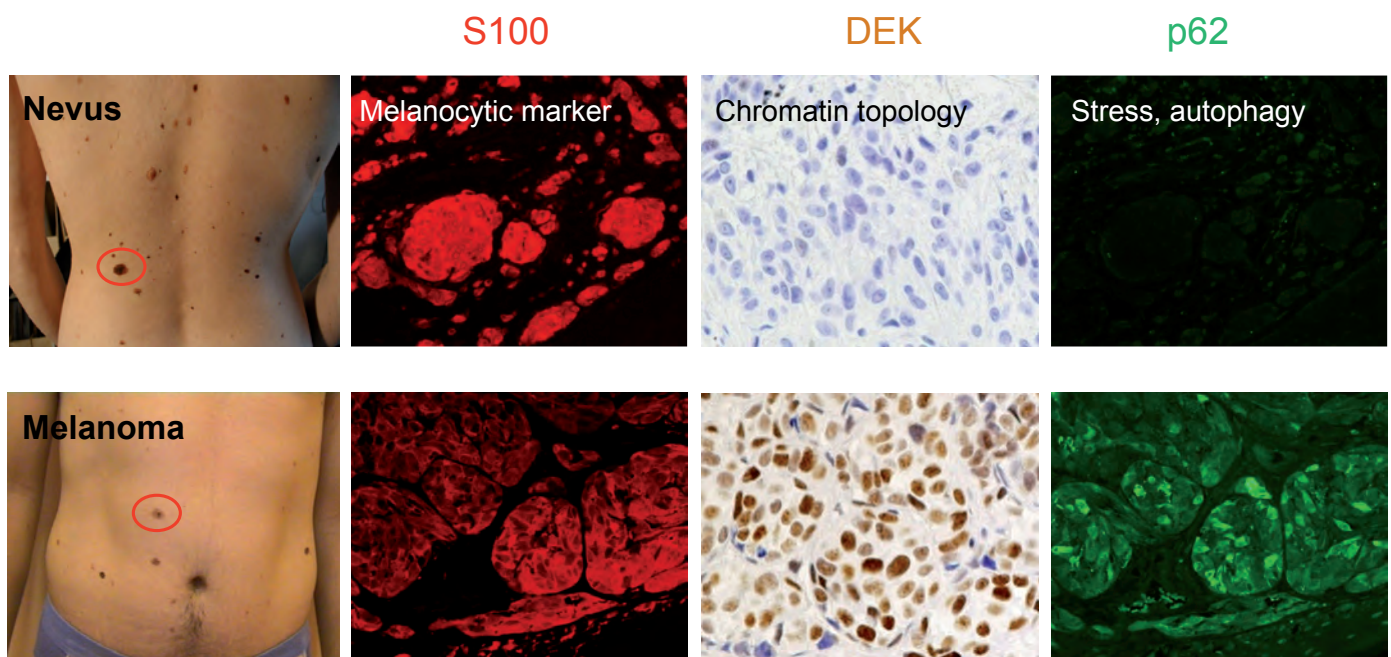


Figure 1: Putative new molecular diagnostic markers in melanoma. Macroscopically similar nevus and melanomas both positive for the S100 melanocytic marker (red signal) are distinguishable by the tumour-cell restricted expression of chromatin remodelling factor DEK (brown signal, immunohistochemistry) and p62/sequestosome (green staining, immunofluorescence).

the CNIO, *Bioncotech Therapeutics*, the departments of Dermatology and Pathology at the *Hospital 12 de Octubre* and experts in nanotechnology from the *Universidad de Alcalá de Henares* as well as drug pharmacology at the *Universidad de Valencia*. The initial results of this multidisciplinary group evaluation are a

series of carbosilane-based dendrimers (Figure 2D) with an enhanced ability to specifically deliver dsRNA to melanoma cells. We anticipate that results from our ongoing preclinical studies will allow implementation of the first-in-class nanoparticles customised for targeted killing of melanoma cells *in vivo*.

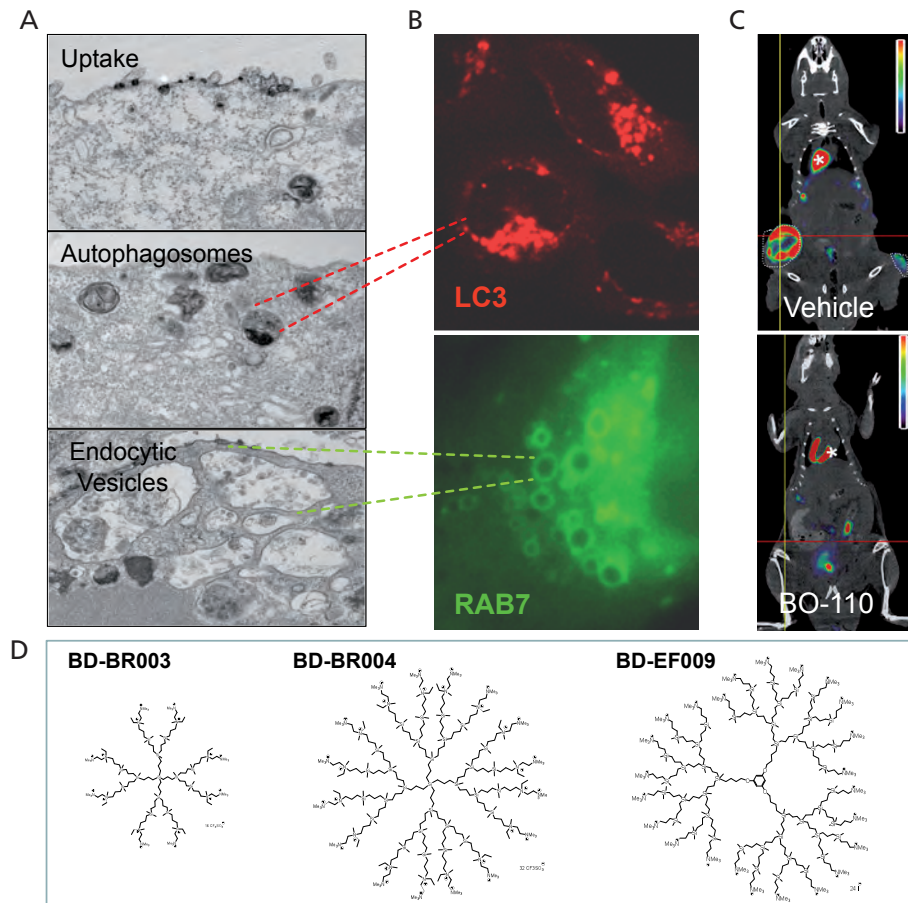


Figure 2: Antimelanoma activity of dsRNA nanocomplexes (BO-110). Cellular uptake, generation of autophagosomes and large endocytic vesicles visualised by electron microscopy (A) or immunofluorescence (B) against the indicated proteins. Antitumour activity of BO-110 monitored by PET-CT in Tyr:NRAS^{Q61K}; INK4a/ARF^{-/-} mice (C). Schematic representation of dendrimeric structures with enhanced selective delivery of dsRNA to melanoma cells (D).

Publications

Alonso-Curbelo D, Soengas MS (2010). Self-killing of melanoma cells by cytosolic delivery of dsRNA: Wiring innate immunity for a coordinated mobilization of endosomes, autophagosomes and the apoptotic machinery in tumor cells. *Autophagy* 6, 148-150.

Riveiro-Falkenbach E, Soengas MS (2010). Control of tumorigenesis and chemoresistance by the DEK oncogene. *Clin Cancer Res* 16, 2932-2938.

Patent

Tormo D, Soengas MS (2010). Procedure for the identification of dsRNA inducers and their use as anticancer agents (OEPM-04/07/2009). PCT European Patent Application P20100595993. Licensed to *Bioncotech Therapeutics*.

Awards and Recognition

Elected Board Member, Melanoma Research Foundation Steering Committee, USA

Valencia IDEA Award in Biotechnology and Biomedicine, Ayuntamiento de Valencia, Spain