Nicolas Rivron, PhD. Eng.

Group leader of the Laboratory of Synthetic Development

Institute of Molecular Biotechnology

Austrian Academy of Sciences

Vienna, Austria

Personal statement

Fish schools, ant colonies and bird flocks are complex systems that coordinate their collective behaviors to control the emergence and progression of patterns and functions. For example, fish in schools swim faster than individual animals are able to. This broad range of decentralized, adaptive behaviors based on local interactions is called self-organization. We explore how self-organization complements traditional hierarchical genetic (*e.g.*, HOX genes collinearity) and molecular (*e.g.*, morphogen gradients) processes to shape the mammalian organism.

The *blastocyst* is the early mammalian organism before implantation. It is a powerful model for self-organization because it is autonomous, adaptive, and small enough to be studied in great detail. Our lab created a unique model of the blastocyst, termed the *blastoid*, which is formed by the spontaneous organization of stem cells in a dish. Blastoids contain all three cell types that further develop into the complete organism (embryonic and extra-embryonic tissues), and implant when transferred *in utero*. Contrary to blastocysts, blastoids are versatile in that they facilitate the systematic modulation and analysis of the impact of cell numbers, states and communication mechanisms on development. Furthermore, blastoids are readily generated in large numbers, making them suitable for high-throughput genetic and drug screens.

We observe both blastocysts and blastoids to study how self-organization contributes to multicellular patterning and cellular decision making.

Positions

2019 – Present.
Group leader at the Institute of Molecular Biotechnology, Austrian Academy of Science.
Subject: Synthetic mammalian development.

2013 – 2019.
Junior group leader at the MERLN Institute for regenerative medicine.
Guest at the Hubrecht Institute for developmental biology and stem cell research.
Subject: Synthetic mammalian development.

2010 – 2013.
Postdoctoral researcher at the MIRA Institute for biomedical technologies.
Guest at the Hubrecht Institute for developmental and stem cell research.
Subject: Synthetic Embryology. Personal research line. Supervisor of two PhD students.

2006 – 2010.
PhD candidate at the MIRA Institute for Biomedical Technologies. University of Twente.
Thesis: Self-organized vascular networks for tissue regeneration.

2002 – 2004.
Research engineer at Medtronic Inc. U.S.A.
Heart Valve Research Division, California.
Material and Bioscience Center, Minnesota.
Subject: Polymers for cardiovascular medicine.

Education

2006 – 2010. University of Twente. The Netherlands. PhD.
Thesis: Vascular biology and tissue engineering.

1998 – 2003. Université de Technologie de Compiègne. France. Engineer and MSc.
Biology, polymer physics.

Publications

Corresponding author papers *

1: Embryonic signals perpetuate polar-like trophoblast stem cells and pattern the blastocyst axis. Frias-Aldeguer J, Kip M, Vivié J, Li L, Alemany A, Korving J, Darmis F, van Oudenaarden A, Geijsen N, **Rivron NC***. BioRxiv 510362; doi: https://doi.org/10.1101/510362
I conceived and directed the project. It was performed by my PhD student Javier Frias Aldeguer, in collaboration with the stem cell biologist Niels Geijsen and with the quantitative/theoretical biologist Alexander van Oudenaarden (Hubrecht Institute).

2: Chemically-defined induction of a primitive endoderm and epiblast-like niche supports post-implantation progression from blastoids. Vrij EJ*, Scholte op Reimer YS, Frias Aldeguer J, Misteli Guerreiro I, Kind J, Koo BK, van Blitterswijk CA, **Rivron NC***. BioRxiv 510396; doi: https://doi.org/10.1101/510396.
I conceived and directed the project. It was performed by my PhD student / Postdoc Erik Vrij, in collaboration with the stem cell biologist Bonk Young Koo (IMBA Institute) and with the epigeneticist Jop Kind (Hubrecht Institute).

3: **Rivron NC***, Pera MF*, Rossant J, Martinez-Arias A, Zernicka-Goetz M, Fu J, van den Brink SC, Bredenoord AL, Dondorp W, De Wert G, Hyun I, Munsie M, Isasi R. Debate ethics of embryo models from stem cells. Nature 2018.
Martin Pera (The Jackson Laboratory) and I led this consortium of stem cell biologists, ethicists, philosophers and lawyers to discuss the societal and ethical implications of stem cell-based embryo models.

4: **Rivron NC***, Vrij EJ, Frias-Aldeguer J, Boisset JC, Korving J, Truckenmuller RK, van Oudenaarden A, van Blitterswijk CA^, Geijsen N^. *In vitro* generation of blastocyst-like structures solely from stem cells. Nature 2018.
^ Equal contribution.
Commented on the BBC, CNN, Fortune and other medias.
I conceived, directed and performed the project, while in collaboration with the stem cell biologist Niels Geijsen and with the quantitative/theoretical biologist Alexander van Oudenaarden (Hubrecht Institute).

5: Computational profiling of U2OS cells using Cell Profiler and machine learning. Li L, Singh S, De Boer J, Carpenter A, **Rivron NC***^, Carlier A*^, ^Equal contribution.
Submitted.
I conceived and directed the project in collaboration with the computational biologists Aurelie Carlier (MERLN Institute), the bioengineer Jan de Boer (TuE Delft) and the computational biologist Anne Carpenter (Broad Institute). It was performed by my PhD student Linfeng Li.

6: Kicheva A*, **Rivron NC***. Creating to understand - Developmental biology meets engineering in Paris. Development. 2017.
Anna Kicheva (IST Austria) and I wrote this report of the conference ‘Engineering the embryo’ held in Pasteur Institute in 2017.

7: Vrij EJ, Rouwkema J, LaPointe VLS, van Blitterswijk CA, Truckenmüller R, **Rivron NC***.
Directed assembly and development of material-free tissues with complex architectures. Advanced Materials. 2016.
Commented in Science, the NRC Handelsblad, and other media.
I conceived and directed the project. It was performed by me and my PhD student Erik Vrij.

8: Vrij E, Espinoza S, Heilig M, Kolew A, Schneider AM, Truckenmuller RK, van Blitterswijk CA, **Rivron NC*.** 3D High Throughput Screening and featuring of Embryoid Bodies in thermoformed microwell plates.
Lab-on-a-chip. 2016.
I conceived and directed the project. It was performed by my PhD student Erik Vrij.

9: **Rivron NC***, Vrij EJ, Rouwkema J, Le Gac S, van den Berg A, Truckenmüller RK, van Blitterswijk CA. Tissue deformation spatially modulates VEGF signaling and angiogenesis.
PNAS. 2012.
I conceived and performed this project during my PhD training, under the guidance of tissue engineer Clemens van Blitterswijk (MIRA Institute).

10: **Rivron NC***, Raiss CC, Liu J, Nandakumar A, Sticht C, Gretz N, Truckenmüller R, Rouwkema J, van Blitterswijk CA. Sonic Hedgehog-activated engineered blood vessels enhance bone tissue formation.
PNAS. 2012.
Commented in F1000 and in Nature SciBX.
I conceived and performed this project during my PhD training, under the guidance of the tissue engineer Clemens van Blitterswijk (MIRA Institute).

11: **Rivron NC***, Rouwkema J, Truckenmüller R, Karperien M, De Boer J, Van Blitterswijk CA. Tissue assembly and organization: developmental mechanisms in microfabricated tissues.
Biomaterials. 2009.
Leading opinion paper.
I conceived and wrote this opinion paper during my PhD training.

12: **Rivron NC***, Liu J J, Rouwkema J, de Boer J, van Blitterswijk CA. Engineering vascularised tissues *in vitro*.
European Cells and Materials. 2008. Review article.
I conceived and wrote this review along with the molecular biologist Jun Liu (MIRA Institute), during my PhD training.

Co-author papers

1: Vértesy, A, Sahin, Z, Frias-Aldeguer, J, **Rivron, NC**, Geijsen, N, Oudenaarden A. Single-cell staging of meiotic entry identifies the precise sequence of gene expression waves and finds functional order in meiotic sex chromosome inactivation.
Submitted.
I participated to this work from the lab of Alexander van Oudenaarden (Hubrecht Institute) by helping with the single cell RNA sequencing and single molecule RNA FISH.

2: Basak O, Krieger TG, Muraro MJ, Wiebrands K, Stange DE, Frias-Aldeguer J, **Rivron NC**, van de Wetering M, van Es JH, van Oudenaarden A, Simons BD, Clevers H. Troy^{+} brain stem cells cycle through quiescence and regulate their number by sensing niche occupancy.
PNAS. 2017.
I participated to this collaboration between the laboratories of Hans Clevers (Hubrecht Institute) and Ben Simons (Gurdon Institute). I designed and performed, along with my PhD student Javier Frias Aldeguer, the 3D high content imaging of single molecule RNA FISH in whole-mount brain slices.

3: Leferink A, Schipper D, Arts E, Vrij E, **Rivron NC**, Karperien M, Mittmann K, van Blitterswijk C, Moroni L, Truckenmüller R. Engineered micro-objects as scaffolding elements in cellular building blocks for bottom-up tissue engineering approaches.
Advanced Materials. 2014.
I designed and directed the master's thesis of Anne Leferink, who pursued her PhD under the supervision of the microsystem engineer Roman Truckenmuller (MIRA Institute).

4: Potier E, **Rivron NC**, Van Blitterswijk CA, Ito K. Micro-aggregates do not influence bone marrow stromal cell chondrogenesis.
Journal of Tissue Engineering and Regenerative Medicine. 2014.
I contributed to design the project and provided the microwell arrays to the tissue engineer Esther Potier (Eindhoven University).

5: Fennema E, **Rivron NC**, Rouwkema J, van Blitterswijk C, de Boer J. Spheroid culture as a tool for creating 3D complex tissues.
Trends in Biotechnology. 2013.
Review. I wrote part of the manuscript.

6: Truckenmüller R, Giselbrecht S, Escalante-Marun M, Groenendijk M, Papenburg B, **Rivron NC**, Unadkat H, Saile V, Subramaniam V, van den Berg A, van Blitterswijk CA, Wessling M, de Boer J, Stamatialis D. Fabrication of cell container arrays with overlaid surface topographies.
Biomedical Microdevices. 2012.
I designed and perform the cell-based assays under the guidance of the microsystem engineer Roman Truckenmuller (MIRA Institute).

7: Truckenmüller R, Giselbrecht S, **Rivron NC**, Gottwald E, Saile V, van den Berg A, Wessling M, van Blitterswijk C. Thermoforming of film-based biomedical microdevices.
Advanced Materials. 2011.
Review. I wrote part of the manuscript.

8: Rouwkema J, **Rivron NC**, van Blitterswijk CA. Vascularization in tissue engineering.
Trends in Biotechnology. 2008. Review.
Review. I wrote part of the manuscript.

Invited lectures

- CSHL Stem cell biology. U.S.A. 2019. Self-organization in the blastocyst.
- ISSCR. Focus session on the ethics of human embryo models. U.S.A. 2019. What do blastoids teach us? From blastocyst development to the window of implantation.
- IMBA SY-Stem conference. Vienna, Austria. 2019. Stem cell-based embryology.
- EMBL Synthetic Morphogenesis conference. Heidelberg, Germany. 2019. Stem cell-based embryology.
- UCLA. Department of biological chemistry. Kathrin Plath. U.S.A. 2019. Synthetic development: From stem cells to embryos.

- RIKEN BDR Control and design of biosystems. Kobe, Japan. 2019. Stem cell-based embryology.
- DevStem meeting. Nantes University. France. 2019. Invited lecture. Synthetic development: from stem cells to embryos.
- Queen Mary Research Institute. Edinburgh. Scotland. 2019. Invited lecture. Synthetic development: from stem cells to embryos.
- Swiss Stem Cell Network. Basel. Switzerland. 2019. Invited lecture. Synthetic development: from stem cells to embryos.
- Erasmus MC Querido Honours College. Rotterdam, The Netherlands. 2019. Invited lecture. Synthetic development: ethical and societal implications.
- IMBA. Vienna, Austria. 2019. Invited lecture. Synthetic development: from stem cells to embryos.
- Erasmus University. Rotterdam, The Netherlands. 2019. Invited lecture. Synthetic development: from stem cells to embryos.
- DGRM. Dusseldorf, Germany. 2019. Invited lecture. Synthetic development: from stem cells to embryos.
- Scientific council of CRB Anim. Paris, France. 2018. Invited lecture. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.
- Stem Cell Center of Competence. Basel, Switzerland. 2018. Invited lecture. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.
- Belgium Society of Reproductive Medicine. Bruxelles. 2018. Invited lecture. Synthetic embryo: no sex needed.
- German Stem Cell Network. Heidelberg, Germany. 2018. Keynote. Synthetic development: ethical and societal implications.
- CiRA Center for iPSC research and applications. Kyoto, Japan. 2018. Invited lecture. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.
- Riken Center for Developmental Biology. Kobe, Japan. 2018. Invited lecture. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.
- EMBL Organoid conference. Heidelberg, Germany. 2018. Invited lecture. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.
- Cambridge University. Department of Physiology. Cambridge, UK. 2018. Invited lecture. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.

- Royan Institute for stem cells and embryology. Tehran, Iran. 2018. Invited lecture. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.
- Tissue Engineering Regenerative Medicine and regenerative medicine. Young Investigator forum. Kyoto, Japan. Invited lecture. 2018. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.
- Pilani Institute of Technology and Science. Pilani, India. 2018. Invited online lecture. What can stem cells do?
- Aachen University. Aachen, Germany. Invited lecture. In vitro generation of blastocyst-like structures from stem cells.
- The stem cell niche conference. Copenhagen, Danmark. 2018. Invited lecture. In vitro generation of blastocyst-like structures from stem cells.
- Cambridge University. Gurdon Institute. Cambridge, UK. 2018. Invited lecture. In vitro generation of blastocyst-like structures from stem cells.
- Medical Research Council. London, UK. 2018. Invited lecture. In vitro generation of blastocyst-like structures from stem cells.
- European Society for Human Reproduction. Workshop on In vitro modelling: from Embryo to gametes. Bilbao, Spain. 2018. Invited lecture. Blastoids: blastocyst-like structures generated solely from stem cells implant in utero.
- Dutch society for clinical embryology. Utrecht, The Netherlands. 2018. Invited lecture. The clinical potential of synthetic embryos.
- NRC Live. Amsterdam, The Netherlands. 2016. Battling the global health problems of contraception and infertility.
- Curie Institute. Paris, France. 2017. In vitro generation of blastocyst-like structures from stem cells.
- Stem cell and brain research institute. Lyon, France. 2017. Blastoid: synthetic blastocysts from stem cells.
- Pasteur Institute. Paris, France. 2016. In vitro generation of blastocyst-like structures from stem cells. Engineering the embryo conference.

Grants and Awards

2019 - HFSP Young Investigator grant. Main applicant in collaboration with Kyogo Kawaguchi (Riken Institute) and Shantanu Singh (Broad Institute).
2018 – Weijerhorst grant. Co-applicant (declined).
2017 - KNMF personal grant. KIT, Germany.
2016 - ERC advanced grant of Clemens van Blitterswijk (major contribution in conception).
2015 – LINK regional investment.
2014 - KNMF personal grant for microfabrication. Karlsruhe Institute of Technology, Germany.
2013 - Fellow of the Translational Adult Stem Cell Breakthrough programme. ZonMW, The Netherlands.
2012 - Young Scientist Award. World Biomaterial Conference, China.
2010 – Start-up package. University of Twente.
2009 - Conference award. EMBO Conference Series on Morphogenesis and Dynamics of Multicellular Systems, Heidelberg, Germany.
2009 - Conference award. 35th North-East BioEnginering conference, Boston, U.S.A.

Patents

- Blastoid, cell line-based artificial blastocyst P97269EP00. 2013/04/16.
- Self-assembling tissue modules WO/2009/154466. 2008/06/20.
- Implantable medical devices having recesses US20060184235 A1. 2003/09/04.

Book chapter

Rivron NC, Rouwkema J, Truckenmüller RK, van Blitterswijk CA. Cell and Organ Printing. Springer. 2010. What should we print? Emerging principles to rationally design tissues prone to self-organization.

Teaching and mentoring

Post-graduate student trainee

2019 - present. Jinwoo Liu.

2019 - present. Harunobu Kagawa.

2018 – 2019. Delia Koning. Metabolic activity and stem cell differentiation.

2016 – 2017. Erik Jacob Vrij. 3D high content screening for primitive endoderm differentiation.

Graduate Student trainees

2016 - 2018. Linfeng Li. Computational approaches to morphological profiling of stem cells.

2013 - 2019. Javier Frias-Aldeguer. The ground state of trophoblast stem cells.

2011 - 2016. Erik Jacob Vrij. 3D high content and high throughput imaging.

Undergraduate Student Trainees

2016. Maarten Kip.

2015. Franck Darmis.

2012. Anne Dirkse.

2011. Anne Leferink.

2010. Christian Raiss.

2010. Erik Jacob Vrij.

2009. Vedashree Ramakrishna.

Lecturing

2018. Lectures and practicals in Stem cell biology, regenerative medicine, Maastricht University.

2017. Lectures and practicals in Stem cell biology, regenerative medicine, Maastricht University.

2011-2014. Lectures in Biomedical engineering, University of Twente.

2013. Guest lecture in Regenerative medicine, UMC Utrecht.

Practice-based learning

2018. PRA1001. Biological methods. Maastricht University.

2018. PRA1001. Research methods. Maastricht University.

2017. PRA1001. Research methods. Maastricht University.

Collaborators (current)

Alexander van Oudenaarden. Director of the Hubrecht Institute.
Alexander develops experimental, computational, and theoretical approaches to quantitatively understand decision-making in single cells. With his critical help, my lab adapted the available methods (i.e. single cell sequencing protocol, whole-mount single molecule FISH) and used computational approaches to identify and assess the changes occurring upon blastoid development and compartment communication.

Anne Carpenter. Director of the Imaging platform at the Broad Institute.
Anne created the high content screen, open-source software *Cell Profiler,* which uses a series of image-processing and machine-learning modules to quantify and mine the information present in cellular images. With her help, my lab developed pipelines to extract quantitative data from 2D and 3D images, and measure single cell phenotypes (i.e. single molecule FISH, cell/nuclei/organelles morphologies in blastoid, blastocyst and cultured stem cells). Linked with single cell transcriptomic data, this informs us of morphogenetic processes within blastoids. Following the visit to her lab of Linfeng Li, my PhD student, I will visit Anne's laboratory to further strengthen our collaborative work.

Reviewer

Peer-reviewing:
Nature, Plos Biology, FASEB, Lab-on-a-chip, Nature communication, Nature scientific reports, Frontiers, Advanced Biosystems, Biomaterials.

Grants:
ERC grant. Veni grant.