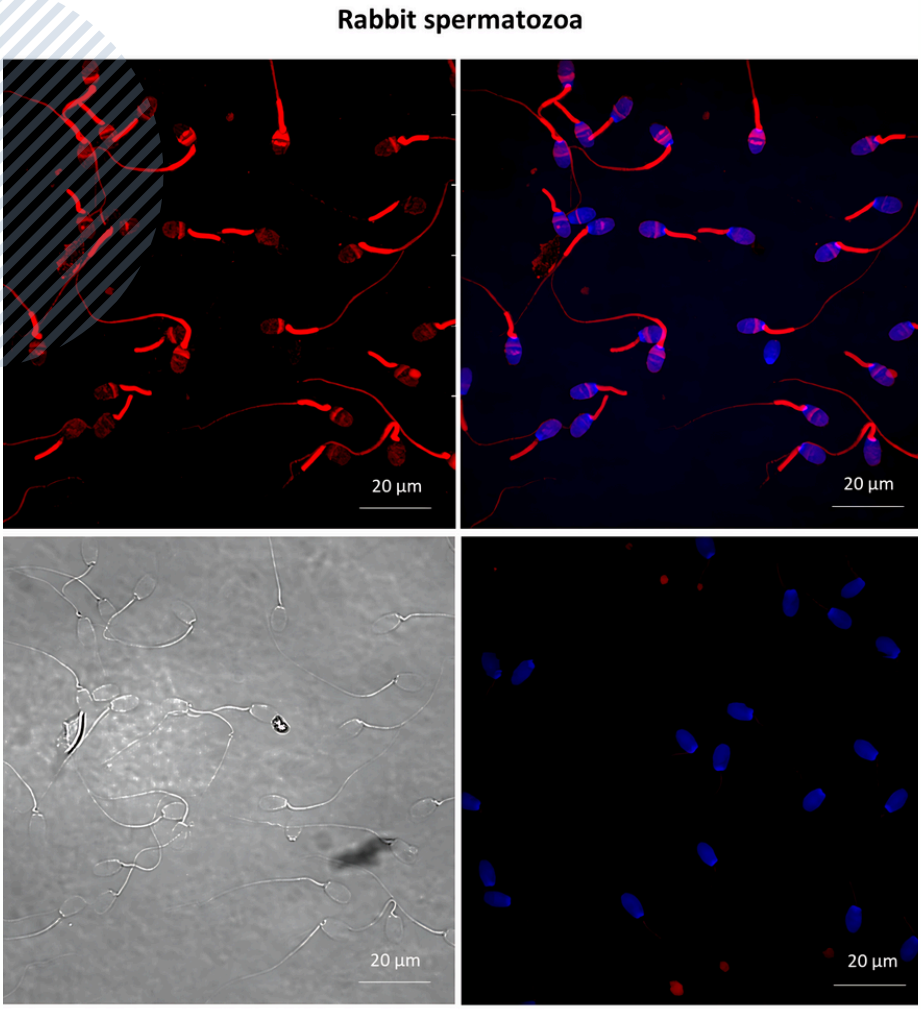
NEWSLETTER

SOCIEDADE PORTUGUESA DE BIOQUÍMICA

June 2024

Edition 3



Immunocytochemistry of rabbit spermatozoa targeting ATP11C

Patrícia Pinto-Pinho,
University of Aveiro

IN THIS EDITION

Editorial

3rd Edition

In this Edition:

In the third issue of the SPB Newsletter, we feature an interview with Cecília Arraiano from ITQB, who tells us about her academic career and research in the RNA world.

Paula Ludovico from the University of Minho and Dora Brites from the University of Lisbon, coordinators of the recently created thematic group on the "Biochemistry of Aging and Regeneration", share their insights on the secrets of aging.

Additionally, Margarida Fardilha from the University of Aveiro, the SPB Ambassador of the FEBS Education group, presents a short report on the FEBS Education and Training Conference.

We also highlight several upcoming events, including the National Congress of Biochemistry, various FEBS activities and open applications, and the IUBMB-FAOBMB-ComBio Conference.

We wish all members of the SPB community a wonderful summer holiday!

The SPB Directive Committee

Interview

Cecília Arraiano

Paula Ludovico (Universidade do Minho) spoke with Cecília Arraiano, Coordinating Investigator (Full Professor equivalent) at ITQB (Instituto de Tecnologia Química e Biológica António Xavier) from Universidade NOVA de Lisboa. Cecília Arraiano was a member of the SPB Board of Directors between 2012 and 2022.

Can you tell us about your scientific career?

I started the University at the Faculdade de Ciências da Universidade de Lisboa (FCUL), where I graduated in Biology, which at that time was a 5-year degree. I decided to do my thesis in Virology at the Laboratório Nacional de Investigação Veterinária (which is now at INIAV-Oeiras) supervised by Professor José Vigário and Professor Natércia Rodeia as the link to FCUL.

I was really privileged to have them as mentors! At the same time, I frequented courses and successfully finished the theoretical part as a Dietitian at Escola Superior de Tecnologia da Saúde de Lisboa.

I also took the English proficiency exam in the British Institute and studied German. I still managed to give private lessons to earn some money for myself. It was a very busy life, but at that age, it was joyful. And all without cell phones...

When I finished my degree, I wanted to go abroad not only to advance my studies but also as a life challenge. Therefore, I applied to Fulbright-Hays for a Fellowship to go to the USA. I had to take all the admission exams for American universities (GRE) and the TOEFL (for English), plus interviews, and passed with success. I ended up going to the Department of Genetics at University of Georgia (UGA) in Athens (near Atlanta), the USA second oldest public university - after Yale.

For some years, I was the only Portuguese there. Initially, I started having many disciplines, such as genetics, nucleic acids, biochemistry, which could lead to a master's or doctorate degree. After the prelims, I started working in Professor Sidney Kushner's laboratory, where I first studied DNA repair, but then I realized it wasn't quite what I liked, and changed to study RNA, which was not well known and attracted me. I implemented everything in the laboratory for RNA study, and at that time, nobody understood why we couldn't touch lab material without gloves and why everything had to be washed specially.

I started to have good results, going to conferences, speaking in societies, and everything started to go very well. When I finished my Ph.D. in Genetics I did a post-doc in the same University, but then I received an invitation to go to John´s Hopkins University in Baltimore to study RNA degradation in viruses. However, in my next vacation in Portugal, I had an interview with Prof. António Xavier, and he invited me to come to work at CTQB - now ITQB - Instituto de Tecnologia Química e Biológica António Xavier.

When I arrived, my laboratory was under construction, and they were still putting pipes... There were no computers, no phones to make a call. It was a shock... but now fortunately things are really different! Thanks to my small laptop computer brought from the USA, I was able to write my first project for JNICT (now FCT) and start developing the laboratory and getting students.

What motivated you to study RNA and how did you enter the RNA world?

I've always liked trying to discover things that I can't see, so I've always been curious about everything microscopic, and in this case, RNA goes even further down. I was in the USA when the RNA boom happened, and, at the time, almost nothing was known about RNA or what happened when cells were without RNases. What are these molecular machines that mature, perform the quality control and eventually degrade the RNAs? The RNAs that are not needed or are poorly made, the ribonucleases cut them and from the resulting pieces, like LEGO, they make other RNAs, it's like circular economy.

I tell students: DNA is the architect's plan, we're going to build this house and it's going to be like this, but then depending on the environmental conditions and the money available, the windows will have to be different and can't be as large as planned and have to face the sun, and so those who execute and alter DNA's orders - and these are the RNAs. In addition to these project managers there are also electricians, plumbers, certain specific tasks for certain RNAs. But there are also construction inspectors who are the small non-coding RNAs, which monitor if something is wrong. These small non-coding RNAs bind and communicate with others and then call ribonucleases and dismiss them when they no longer need them on the site. And this happens in all living beings.

Which notable achievements would you like to emphasize?

Our pioneer work on the structure and mechanism of action of RNase II was the first characterizing a ribonuclease from this family and resulted in the publication in Sept 2006 in Nature, and had a great impact because many groups all over the world were trying to achieve it. This was a stepping stone for international recognition. In 2008 I was elected member of European Molecular Biology Organization (EMBO), in 2009 elected Member of the Portuguese Academy of Sciences, in 2014 elected Fellow of the American Academy of Microbiology (the first in Portugal), in 2016 elected Fellow of European Academy of Microbiology.

In 2012-2015 I was elected President of the Portuguese Genetics Society and was the Ambassador of American Society of Microbiology in Portugal. In 2008 I received the Prize C. Pestana/Glaxo Smith-Klein, and in 2007 Prize for Oeiras Gold Medal for Services. In short, there was global recognition of myself and my research group.

In 2020 I was elected to Advisory Board of European Synthetic Biology Society (EUSynBioS). Since 2021 I belong to External Advisory of Egas Moniz Research Centre (CiiEM), Portugal. In 2021 I have integrated the Scientific Advisory Board of the Max Planck Research Unit for the Science of Pathogens, Berlin, for 2022-2028. The Director is Emmanuelle Charpentier - 2020 Nobel Prize for CRISPR-Cas9 genome editing. Federation of European Biochemistry Societies (FEBS) has a really important mission in Europe and beyond... I was first elected for the Advanced Courses Committee where I stayed several years. Prof. Claudina Rodrigues Pousada, with whom I became very close in many ways, encouraged me to apply to become Chair of the FEBS Working Group for Women in Science.

I won several elections for this role and therefore from 2012-2022 I was a member of the FEBS Executive Committee. In 2023 I received the "Diplôme d´Honneur" from FEBS for my work and for the service provided.

What scientific discovery has surprised you the most, and what is the greatest challenge you have faced or are still facing?

Let me begin with the greatest challenge I've been facing in the past two years, which is securing funding for my research group. Meaning, we still have some funds, but we are starting considering collaborations to conduct certain experiments because resources are limited. It's a new challenge in Portugal.

But my greatest challenge was when I first arrived in Portugal and had to establish a group that could compete internationally. I believe we succeeded, and now I'm faced with this unexpected challenge at a later stage in my career. Despite expectations, Portugal still has the same or even less funding than it did 20 years ago. As technology advances globally, there's a growing need for more comprehensive approaches, and although we've always networked, we need more support to continue.

The scientific discovery that brought me the most satisfaction or pleasure was being able to achieve in Portugal what American, French, and other international groups had been attempting for years, and we did it here, on our own, with local talent. Made in Portugal!

A more recent surprise, which also brought me immense joy, took place during the COVID-19 pandemics. Margarida Saramago an Investigator in my lab tried to see how we could help with our knowledge and other lab members joined forces and started studying the ribonucleases of the virus SARS-CoV-2.

We even obtained two patents because we found a strategy to hinder the virus's replication process. The virus carries a protein important for quality control in the virus's replication: NSP14, which is an exo- and ribonuclease with two domains. This protein with two domains is almost like science fiction.

One domain ensures fidelity in producing error-free virus RNAs. But the other domain, called methyltransferase, is absolutely surprising: the virus brings its own machinery to add a 5' cap to the viral RNAs in the cytoplasm, and the viral RNAs became alike the human RNAs. I tell the students that this is the Harry Potter cape. It makes its own RNAs invisible in our cells.

How does the virus know? It doesn't create this process out of nothing. It's like when someone enters a house they've never been to, without any prior information or map, and knows exactly where the safe is and brings the specific tool to open it. When I say this, the response is always "evolution," but evolution how? I believe there must be a mechanism of epitranscriptomics. I think there has to be a memory. Philosophically, it's very intriguing.

What message would you like to convey to young people dedicated to Biochemistry who want to continue researching?

Don't be afraid! At least we're still privileged compared to so many societies that are at war and in terrible conditions, societies that would love to have the opportunities we have.

I also leave the message I give to my students when they start in the lab: we have to be very curious. If you don't like it or if you're not curious enough, follow another path because this is not the life you should pursue.

You always have to think about fighting because every time we discover something, no matter how insignificant it may seem, we are the first to know. It's like planting a flag on the moon!

Even if you publish or not, it's ours, and we were the ones who managed to discover it. I think this should be a source of inspiration to keep going and wanting to know more. And to see how wonderful nature is. Nature is amazing!

We should also have the idea that whether it's now or further down the road, we are contributing to the good of humanity and to sustainability.

It's a profession where we usually don't work to be rich, although money is very necessary to have salaries, consumables and equipment, but we have an advantage that thousands and thousands of people don't have, which is doing what we love. Most people do what they have to do to survive, not necessarily what they like.

Often, we work hard, but there are certain days when we feel like we're engaged in a hobby, not a job.

> ***There's a great reward for this profession.***
> ***We don't have a routine!***
> ***We are forever young!***

News & Views

Secrets of ageing: Biological Processes, Models, and Rejuvenating Strategies

Paula Ludovico, School of Medicine. University of Minho
Dora Brites, Faculty of Pharmacy, University of Lisbon

Ageing is accompanied by a gradual decrease in physical and mental capacity, a growing risk of disease, morbidity, and mortality, which constitutes a major societal challenge.

In this global context, interest in ageing ceases to be a solely economic and philosophical issue and emerges as one of the most important and determining areas of scientific research which, due to its complex and multifactorial nature, requires different perspectives and approaches.

Early theories of ageing, such as the "**wear and tear**" proposed by August Weismann in the late 19th century, suggested that ageing was the result of the accumulation of damage to cellular structures over time, involving DNA mutations, protein oxidation, and cellular senescence (Figure 1).

In the mid-20th century, the discovery of genetic mutations that could extend lifespan led to the identification of several **longevity genes and pathways**, such as mTOR.

In the 1960s and 1970s, programmed theories of ageing emphasized genetic or biological reasons, regulated by specific biological clocks (e.g., **telomere shortening hypothesis**) (Figure 1).

With the advent of high-throughput "omics" technologies in the late 20th and early 21st centuries, there was a shift towards a more holistic and **systems-level understanding of ageing**, using the power of genomics transcriptomics, epigenomics, proteomics and metabolomics and involving the interplay of multiple molecular pathways and networks.

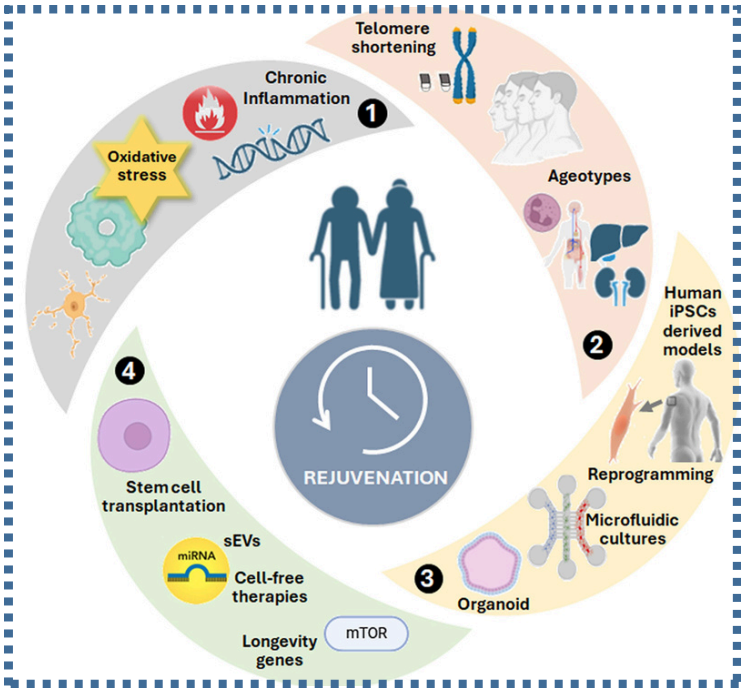


Figure 1: *Schematic representation of ageing drivers and regenerative strategies.*
(1) Ageing molecular and cellular processes;
(2) Ageing theories;
(3) Ageing models;
(4) Reversing ageing approaches. sEVs, small extracellular vesicles.

Created with the help of Biorender.com.

Lately, twelve hallmarks of ageing were proposed: genomic instability, telomere attrition, epigenetic alterations, loss of proteostasis, disabled macroautophagy, dysregulated nutrient-sensing, mitochondrial dysfunction, cellular senescence, stem cell exhaustion, altered intercellular communication, dysbiosis and **chronic inflammation.**

More recently, cutting-edge molecular technologies, longitudinal studies and multiomic data identified the existence of **pro-ageing and pro-youthful factors**, as well as distinct ageing phenotypes, termed "**ageotypes**" (metabolic, immune, hepatic, and nephrotic) [1].

Ageotypes represent unique ageing trajectories that individuals may follow based on various factors, such as genetics, lifestyle, and environmental exposures. Ageotypes can change over time and a person may have more than one ageotype. Though ageotypes might help people in taking preventive measures (diet, regular exercise, or medicines) and hold promise for personalized medicine and targeted interventions, the understanding on why different cells, organs or systems are particularly vulnerable to ageing effects requires a long way of research once there are no tests to assess one's ageotype.

It is crucial to identify **epigenetic predictors** (dynamic changes in DNA methylation and histone modification, non-coding RNA, and chromatin remodeling) as gold standard biomarkers for the estimation of the ageotype. In-depth research on the **secreted miRNAs** and their target genes in ageing may also impact as biomarkers of ageing.

Metabolic processes by providing substrates, producing proinflammatory cytokines and generating reactive oxygen species, can influence the epigenetic landscape.

A better comprehension of the metabolic foundations of ageing and age-related diseases can facilitate the development of personalized interventions toward specific ageotypes. Impaired **DNA repair mechanisms and accumulation of DNA damage** are drivers of genomic instability and influence an individual's propensity for ageing-related changes and the development of ageotypes.

Lastly, the combination of multiple omics with the artificial intelligence has revealed **organ-specific clocks** and sex-specific (female life expectancy is longer than males), which may enable early risk identification and the application of preventive strategies soon [2].

Dissection of the underlying mechanisms of ageing benefit from the use of human models based on 2D/3D systems and reprogramming technologies. **Induced pluripotent stem cells (iPSCs)**, transdifferentiated cells and organoids have been used to investigate ageing.

Ways to induce an ageing phenotype in these rejuvenated cells are long-term culture, culture in aged extracellular matrix (ECM), telomere shortening and gene edition by CRISPR/Cas9 [3]. In contrast, **direct cellular conversion** largely retains ageing-associated features of the original somatic cell but poses scaling-up challenges.

Considering the 2D ageing models, they are easy to perform and analyze, but do not recapitulate physiological cell distribution and interaction. In this context, efforts were developed to construct **3D models** comprising different cell types and ECM components in a **microfluidic system** to mimic the interrelated paracrine signaling, or **organoids** originated from progenitor cells differentiated from iPSCs, cultured in suspension, or embedded in ECM.

Organoids are a good model to study cell type dependent ageing mechanisms, inter-individual variability, and patient specific drug testing.

However, they show deficient cell maturation, the presence of a necrotic core and low reproducibility given the limited vascularization. Lately, incorporation of functional vasculature throughout microfluidic-trapped embedded 3D organoids allowed long-term cultures, maturation, and functional connections, enabling age stratification and development of personalized senolytics.

Putative rejuvenation (age reversal event) strategies encompass the dilution and/or removal of pro-ageing factors or by the introduction of beneficial youthful factors.

Partial cell reprogramming is also a promising and exciting technique to maintain cellular identity while inducing cellular rejuvenation (Yamanaka factors or exposure to chemical cocktails), but side events, low efficiency and lack of specificity have hampered its translational application [4].

Nevertheless, lifespan extension after **stem cell transplantation** was shown by several studies. Moreover, the use of cell-free therapies, based on the administration of cell-derived **small extracellular vesicles (sEVs)**, or sEVs engineered with miRNAs, also counteracted pre-existing ageing and cell senescence, but production challenges should be adequately overcome in the future.

In conclusion, ageing is a complex, dynamic, and time-dependent process associated with cell damage accumulation, tissue/organ structure/function deterioration, and presence of chronic diseases.

There are individual specifications and not all tissues "age" at the same rate, though they selectively influence the multiorgan ageing network.

Therefore, a better knowledge on such individual processes will contribute to emerging specific regenerative and rejuvenation interventions targeting ageing-related mechanisms as priority goals in biomedical research to extend the human lifespan while preserving health.

REFERENCES:

[1] Grammatikopoulou, M.G., et al., Ageotypes revisited: The brain and central nervous system dysfunction as a major nutritional and lifestyle target for healthy aging. Maturitas, 2023. 170: p. 51-57.

[2] Prattichizzo, F., et al., Organ-specific biological clocks: Ageotyping for personalized anti-aging medicine. Ageing Res Rev, 2024. 96: p. 102253.

[3] Pitrez, P.R., et al., Cellular reprogramming as a tool to model human aging in a dish. Nat Commun, 2024. 15(1): p. 1816.

[4] Yucel, A.D. and V.N. Gladyshev, The long and winding road of reprogramming-induced rejuvenation. Nat Commun, 2024. 15(1): p. 1941.

FEBS activities

FEBS EDUCATION AND TRAINING CONFERENCE

Turkey, March 20th-23rd, 2024

Report by Margarida Fardilha, University of Aveiro

The **FEBS EDUCATION AND TRAINING CONFERENCE**, which took place in Turkey from March 20-23, was the first Molecular Life Sciences Education conference in Europe organized by FEBS.

Fig. 1. FEBS Education and Training Conference

It was attended by ~200 people from all over the world (Fig 1), including professors and senior and junior scientists to share their expertise and learn from each other. Portugal was very well represented with 8 participants from different parts of the country (Fig 2).

The conference included a variety of activities, plenary lectures; practical workshops; meet the expert; certification opportunities; oral and poster presentations and networking opportunities. The Portuguese team played an important role with lectures and workshops.

Participants will always remember the exceptional quality of the lectures and workshops, as well as the valuable networking opportunities.

Fig. 2. FEBS Education and Training Conference

Calendar & Events

Until 01/07

FEBS Excellence Award application
LINK

3 months before

FEBS Short-Term Fellowships
LINK

Until 01/09

FEBS Advanced Courses 2025 applications
LINK

22 - 26/09

Biomolecular Horizons 2024 (IUBMB-FAOBMB-ComBio)
Melbourne, Australia
LINK

25 - 27/09

RNA Therapeutics Symposium: Unlocking the RNA's Potential in Medicine
Porto
LINK

Until 30/09

FEBS 60th Anniversary activities
FEBS Letters Writing Contest: "Blueprints for the scientific society of tomorrow"
LINK

24-26/10

XXII SPB National Congress of Biochemistry
Aveiro
LINK

Open

FEBS National Lectures and FEBS 3+ Meeting applications (2024 and 2025)
LINK