Name	VASSILIS PACHNIS
Position	Senior Group Leader
Year joined *(Crick or founder institute)*	1991

Career History

1980: University of Athens Medical School (Athens, Greece) M.D.
1986: University of Pennsylvania, Philadelphia, PA (USA) Degree: Ph.D. (Genetics)
1986-1988: Howard Hughes Medical Institute Center for Neurobiology and Behavior, Columbia University, New York, NY (USA), Postdoctoral Fellow (Prof. Richard Axel's laboratory)
1988-1991: Department of Genetics and Development Columbia University, New York, NY (USA) Postdoctoral fellow (Dr. Frank Costantini's laboratory)

Major Awards, Honours and Prizes

Fellow of the Academy of Medical Sciences, UK (2000).
Janssen Award for Life Time Achievement in Neuroscience and Gastroenterology (2002)
EMBO Member (2007)
Fellow of the Royal Society (2018)

Membership of external committees, editorial boards, review panels, SABs etc

Lab Name	***Nervous System Development and Homeostasis Laboratory***

Research programme and achievements

Research in my laboratory focuses on understanding the genetic and molecular mechanisms that control the development and homeostasis of the vertebrate nervous system. During the last quinquennium, we addressed these questions in two parts of the nervous system, the forebrain and the enteric nervous system (ENS).

Our work demonstrated that the number of inhibitory neurons in the mouse cortex is regulated cell autonomously by activity-induced apoptosis during a critical postnatal window. This work provided novel insight into the mechanisms that determine the balance between excitation and inhibition in the mammalian forebrain (Denaxa et al. *Cell Rep* 2018).

We also explored the interaction of microbiota and the cell lineages of the ENS. In particular, we demonstrated for the first time that the cellular organisation of the ENS is highly dynamic and regulated by the postnatal colonisation of the gut by microbes (Kabouridis et al. *Neuron* 2015). In addition, we characterised molecular mechanisms underpinning the communication between ENS and the luminal environment of the gut. Collaborative work between my lab and the Stockinger lab demonstrated that central to the ENS-microbiota axis is the transcription factor AhR, which functions as a microbiota

and diet-activated biosensor of intestinal neural circuits, thus linking peristaltic activity to the luminal contents of the gut (Obata et al. *Nature* 2020).

We have extended our analysis of the molecular mechanisms that regulate the development of ENS lineages. Using *in vivo* genetic lineage tracing, we examined the developmental potential of mammalian ENS progenitors and characterised their transcriptomic landscape at early stages of ENS ontogenesis. In addition, clonogenic analysis of ENS progenitors led us to propose a new model for the spatial organisation of the mammalian ENS, which links developmental mechanisms employed for the assembly of intestinal neuroglia networks with their functional output (Lasrado et al. *Science*, 2017).

We have also explored the mechanisms that maintain the adult ENS at steady state. Using zebrafish as a model organism, we have demonstrated that enteric glial cells (EGCS), in addition to functioning as canonical glia that provide support and nourishment to enteric neurons and regulates neuronal activity, function as neural stem cells undergoing constitutive neurogenesis. This work reveals previously unappreciated similarities between enteric glia and neural stem cells in the brain and raise hopes that *in vivo* activation of the neurogenic potential of mammalian EGCs could restore the activity of intestinal neural circuits compromised by developmental deficits or disease (McCallum et al. *eLife* 2020).

Our work for the next quinquennium will aim at understanding the developmental basis of the functional units established between the intrinsic neural circuits of the gut and other gut tissues in adult life.

We have now generated a transcriptional landscape of mammalian ENS progenitors, enteric neurons at early stages of differentiation and enteric glia at all stages of development and in the adult. In parallel we have generated cellular and transcriptomic maps of adult zebrafish ENS. These experiments have generated rich datasets which identify a large number of candidate regulators of enteric neuron and glia differentiation and their assembly into functional circuits. We plan to employ hPSC-based *in vitro* models of enteric neurogenesis as well as genetic animal models (mouse and zebrafish) to dissect the molecular cascades that underpin ENS lineage differentiation. In addition to providing fundamental insight into the development of the peripheral nervous system, this work promises to identify molecular pathways implicated in the pathogenesis of neurogenic gut disorders.

Our group was the first to identify signalling queues which originate from the gut mesenchyme and promote the development of the mammalian ENS. We have obtained evidence that, in a reciprocal manner, ENS lineages also regulated the morphogenesis, differentiation and homeostasis of surrounding gut tissues. We plan to characterise such ENS-derived signals and their upstream regulators, and their effect on surrounding tissues at different developmental stages and in adult animals. Of particular interest is our recent discovery that homeostatic immune signals acting on EGCs regulate the inflammatory state of a wide spectrum of non-neuroectodermal cell types of the gut, including fibroblasts, mesothelial cells and macrophages. We will identify the molecular mechanisms underpinning such interactions and their role in tissue maintenance and repair following injury or disease.

Finally, we will examine how changes in the maternal environment, such as dysbiosis, infections, altered diet and stress, influence the development of the ENS in mammalian embryos. We will determine whether adverse maternal environment alters the differentiation and organisation of the ENS during embryogenesis and early postnatal life and how such changes may predispose to gastrointestinal disorders in later life.

Research outputs

Kabouridis SP, Lasrado R, McCallum S, Chng SH, Snippert H, Clevers H, Pettersson S and Pachnis V. (2015) *Microbiota controls the homeostasis of glial cells in the gut lamina propria.* Neuron 85: 289-295.DOI: 10.1016/j.neuron.2014.12.037
This paper demonstrates for the first time that the cellular organisation of the mammalian enteric nervous system is highly dynamic and is influenced by the postnatal colonisation of the gut by microbiota.

Lasrado R, Boesmans W, Kleinjung J, Pin C, Bell D, Bhaw L, McCallum S, Zong H, Clevers H, Vanden Berghe P, Pachnis V. (2017) *Lineage-dependent Spatial and Functional Organization of the Mammalian Enteric Nervous System*. Science 356:722-726. DOI: 10.1126/science.aaam7511
In this paper we use genetic lineage tracing and clonal analysis to characterise mammalian enteric nervous system progenitors, define differentiation trajectories for enteric neurons and glia during development and propose a new model for the 3-D organisation of the enteric nervous system.

Denaxa M, Neves G, Burrone J and Pachnis V. (2018) *Modulation of Apoptosis Controls Inhibitory Interneuron Number in the Cortex*. Cell Rep 22:1710-1721. DOI: 10.1016/j.celrep.2018.01.064
Here, we provide evidence that the extent of cortical interneuron apoptosis during the critical early postnatal period is plastic and cell-type specific and can be reduced in a cell-autonomous manner by acute increases in neuronal activity.

Obata Y, Castaño Á, Boeing S, Bon-Frauches AC, Fung C, Fallesen T, Gomez de Agüero M, Yilmaz B, Lopes R, Huseynova A, Horswell S, Rao Maradana M, Boesmans W, Vanden Berghe P, Murray AJ, Stockinger B, Macpherson AJ and Pachnis V. (2020) *Neuronal programming by microbiota regulates intestinal physiology*. Nature 578:284-289. DOI: 10.1038/s41586-020-1975-8
In this paper we explore the molecular mechanisms used by enteric neurons to monitor the luminal environment of the gut. In particular, we demonstrate that the transcription factor AhR functions as a biosensor of intestinal neural circuits, linking their functional output to the microbial environment of the gut lumen.

McCallum S, Obata Y, Fourli E, Boeing S, Peddie CJ, Xu Q, Horsewell S, Kelsh R, Collinson L, Wilkinson D, Pin C, Pachnis V* and Heanue T*. (2020) *Enteric glia as a source of neural progenitors in zebrafish*. ELife 9:e56086. DOI: 10.7554/eLife.56086.
Here we identify the stem cells of the vertebrate enteric nervous system. We demonstrate that enteric glia in the gut of zebrafish have a double character: they function as canonical glial cells, but are also capable of proliferating under physiological conditions giving rise to progeny that differentiate to enteric neurons.