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Building the embryo of developmental biology in Uruguay

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Abbreviations: ANII, Agencia Nacional de Investigación e Innovación; CSIC, Comisión Sectorial de Investigación Científica; IIBCE, Instituto de Investigaciones Biológicas Clemente Estable; LASDB, Latin American Society for Developmental Biology; PEDECIBA, Programa de Desarrollo de las Ciencias Básicas; UCL, University College London.

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Abstract
In Uruguay, a country with a small population, and hence a small scientific community, there were no classical embryologists as such in the past. From the decade of 1950, however, a cumulus of favorable conditions gave rise to highly active and modern research groups in the fields of cytology and physiology, that eventually contributed to developmental biology. The advent of a long dictatorship between de 1970’s and 1980’s caused two things: a strong lag in local research and the migration of young investigators who learned abroad new disciplines and technologies. The coming back to democracy allowed for the return of some, now as solid researchers, and together with those who stayed, built a previously inexistent postgraduate training program and a globally-integrated academy that fostered diversity of research disciplines, including developmental biology. In the present work we highlight the key contribution of pioneer researchers and the significant role played by academic and funding national institutions in the growth and consolidation of developmental biology in our country.

Historic background: the egg
As a discipline, developmental biology is generally assumed to be the natural post-molecular biology derivative of classical embryology. And this is rather clearly so in developed countries, where the whole range of biological topics were being actively investigated around the 1970’s. But what happens with countries with a small population and a very low budget dedicated to science? Several disciplines would be underrepresented, and eventually inexistent, unless researchers from diverse areas would decide to foray into the unexplored areas. This appears to be the case of embryology in Uruguay. In addition to the work of some naturalists, most of the early-mid 20th century biology research in Uruguay was centered around biomedical research, largely led initially by the School of Medicine (Facultad de Medicina, Universidad de la República) and later also by the research institute founded by Clemente Estable (1894-1976), and that now carries his name (Instituto de Investigaciones Biológicas Clemente Estable, IIBCE). Anatomy and physiology research were greatly biased towards investigating the nervous system, probably in part related to the early formation of Estable at Nobel Prize winner Santiago Ramón y Cajal’s lab in
Madrid in the 1920’s; on the other hand, Estable had a great partner in Francisco Alberto Sáez, who was responsible for the establishment of an important cytogenetics school in Uruguay and the region, also influencing some of the work we describe below (Fig. 1A; for general accounts of these earlier years, please see Drets, 2013; Pellegrino et al., 2017).

The beginnings: from the embryo to a fast-growing larva
Not surprisingly, neuroscience research in Uruguay was largely at the cellular level, with a great development of cytology and electrophysiology. In the 1950’s the structural study of cells demanded the use of electron microscopy, and this is one of the technical developments that undoubtedly lead to some of the most important initial contributions of our country to the prospective disciplines of cell and developmental biology. In 1950, the institute led by Estable moved to a new building, where it is still located today, and with the support of The Rockefeller Foundation, acquired the first transmission electron microscope of Latin America. Estable invited Eduardo De Robertis (1913-1988; a recognized Argentinean researcher then living in the USA) to run the recently created Department of Cell Ultrastructure, which nucleated several researchers and was considered to be “the first Latin American school of electron microscopy” (Fig. 1B). De Robertis was interested in understanding the fine structure of vertebrate neurons, being the discoverer of microtubules in axons and synaptic vesicles. Some of his work in Uruguay included exquisite descriptions of the formation of the outer segment of mouse photoreceptors from the initial cilium to the development of the membranous stacks (De Robertis, 1956), a work that he continued as he returned to Buenos Aires. As a side note, De Robertis’ son Edward M. (1947), although born in the USA, was raised and graduated with honors as a medical doctor in Uruguay, before pursuing his research career abroad, to become a world-recognized developmental biologist leading the field of neural induction.

Other relevant contributions of this laboratory to the developmental biology field are pioneering ultrastructural studies on synaptonemal complexes of meiotic cells conducted by J. Roberto Sotelo (1915-1985), Rodolfo Wettstein (1942-2009) and Omar Trujillo-Cenóz (1933) (Fig. 1A). As an example, and initially as a collaboration with one of the founders of biological electron microscopy, Keith R. Porter (1912-1997) from
Rockefeller, Sotelo began an ultrastructural characterization of mammalian oocytes. This soon led to his report of the presence of synaptonemal complexes in these cells (Sotelo, 1959; Sotelo and Trujillo-Cenóz, 1960), only described on spermatocytes in previous years. In later studies, using micrographs from around a hundred ultrathin serial sections, Sotelo and Wettstein were capable of building 3D reconstructions of the entire meiotic nucleus and following the full trajectory of each bivalent from its insertion in the nuclear membrane (Sotelo and Wettstein, 1964; Wettstein and Sotelo, 1967). This approach confirmed the concept that synaptonemal complexes represent paired homologous chromosomes and allowed predicting their key role in the crossing over. Collectively, these studies gave rise to a new discipline, ultrastructural cytogenetics. The cell biology of meiosis work initiated by Sotelo and co-workers was continued by a collaborator of Wettstein, Ricardo Benavente (1954), first in Montevideo and now in Würzburg, Germany (Benavente and Wettstein, 1977; Spindler et al., 2019). Other remarkable contributions from this group are: studies on axon regeneration (Estable et al., 1957); ultrastructural analyses of embryonic development in the chick, including the first description of the presence and organization of primary cilia in the neuroepithelium of vertebrates (Sotelo and Trujillo-Cenóz, 1958); the innervation and synaptogenesis of the acoustic organ in chick embryos (Vázquez-Nin and Sotelo, 1968); the ultrastructure of the fly eye imaginal disk and differentiating retina (Melamed and Trujillo-Cenóz, 1975; Trujillo-Cenóz and Melamed, 1978).

Another technical development that eventually arrived in the early 1950’s was cell and tissue culture. In 1950, the then Professor of Histology and Embryology at Facultad de Medicina, Washington Buño (1909-1990) invited the famous Italian histologist and cytologist Giuseppe Levi (1872-1965), who among other things, helped Buño and his collaborator Horacio Goyena to install the first Tissue Culture Laboratory in Uruguay (Fig. 1C-E). Goyena directed this laboratory for nearly 25 years, and there he trained young researchers in this technique, which he used for research on the effect of cortisone on bone growth in vitro, isolating long bones from chick embryos (Buño and Goyena, 1955; Calcagno et al., 1970). It could be important to remember that Levi’s lab in Turin was the cradle of three future Nobel Prize winners: Salvatore Luria, Renato Dulbecco and Rita Levi-Montalcini. At the same Department of Histology and Embryology, María Antonieta Rebollo (1923) conducted research on different
anatomical and histochemical aspects of chick development, largely on the
neuromuscular system (see for example De Anda and Rebollo, 1968). A closely related
place where cell-developmental research was performed in those years was the School
of Humanities and Sciences (Facultad de Humanidades y Ciencias; from which the
current Facultad de Ciencias was separated in 1990). There, and since the 1950’s,
Gabriel Gerard (1919-2000) was Professor of the Department of Microscopic Anatomy
and Cytology (now Cell Biology). He was mainly a cell biologist, whose research was
focused on histochemical features of gametes and muscle cells (Castellano et al., 1969;
Castellano et al., 1973).
On the physiology side, it is of remark the study made by one our most internationally
recognized neuroscientists, Elio García-Austt (1919-2005), on the origin and
development of electrical activity of the chick embryo brain (Garcia-Austt, 1954).
Another physiologist, Roberto Caldeyro-Barcia (1921-1996), took a different path and
in a multidisciplinary collaboration (that at some stage included biochemists and
electrophysiologists, like García-Austt, but also very importantly, clinical physicians), he
established essential bases on human perinatology used worldwide today, and
deserving him being in the Nobel Prize shortlist in three occasions (Fig. 1F; see an
dexample of his many publications in Caldeyro-Bacaria and Poseiro, 1959, and a
complete biography in Beretta Curi, 2006).

The diaspora and the return to democracy: after pupation, a bright butterfly emerges
From the beginning of the 1970’s and especially with the coup d’etat of 1973, until
1985, the country was under a dictatorship that set up the diaspora of most of the
researchers that worked on embryology, as well as other related disciplines. Many of
them ended up living in France, Spain, Germany, Sweden, Mexico and other countries
where they could follow their studies and/or establish in laboratories that were
interested in similar topics. Uruguay returned to democracy in 1985, and one year
later a revolutionary idea born from researchers living in the country and abroad, was
made a reality: PEDECIBA (the Program for the Development of Basic Sciences). This
transversal program, which spans all academic institutions where scientific research is
conducted and aims largely at the formation of graduate researchers. Throughout the
first 20 years it was possible to recover more than a hundred Uruguayan researchers
whether living abroad or in Uruguay but kept away from labs. Among these researchers, and the courses that they organized, developmental biology was present for the first time with this name. The first regular course on Developmental Biology, still running today, was organized by Cristina Arruti (1944-2018), a former collaborator of Horacio Goyena. After brief passages through the labs of the famous developmental biologists Étienne Wolff and Louis Gallien in the early 1970's, Arruti went back to Paris to work on eye development at Yves Courtuois’ lab, where she obtained her Doctorate. There, she discovered a factor secreted by the retina able to affect the differentiation state of lens cells, which turned out to be the basic fibroblast growth factor, or FGF2 (Arruti and Courtois, 1978). Returning to Uruguay in the 1980's, Arruti rescued and directed Goyena’s Tissue Culture Laboratory first at Facultad de Medicina, and since 1999 established at Facultad de Ciencias (where she was Professor of Cell Biology until retirement in 2015). During this time, she supervised many young researchers who still today work on different aspects of eye and nervous system development, in Uruguay and abroad (see for example Cirillo et al., 1990; de Maria and Arruti, 1995; Zolessi and Arruti, 2001). A hallmark regarding developmental biology teaching was the invitation she made to Claudio D. Stern (1954), another Uruguayan-born and very influential developmental biologist of today (now in UK), who gave a highly motivating experimental course in Montevideo in 1996.

Ricardo Ehrlich (1948) also returned by that time from an exile in France, where he obtained a Doctorate in Physical Science at University Louis Pasteur, Strasbourg. While preparing his return, his interaction and exchange with other Uruguayan researchers in France, including Arruti, was an important stimulus for him to start a research line on the molecular bases of development in cestode worms. This topic allowed him to link his interests on basic molecular biology with issues of interest for the country, like the fight against hydatidosis. This happened a short time after the discovery of the homeobox in 1983, and in the middle of a whole revolution caused by Drosophila molecular genetics on developmental biology. At that time, molecular studies on flatworms were directed to the highly attractive model organism planaria, but Ehrlich did not hesitate to look into parasitic flatworms instead. The study of the molecular basis of Echinococcus granulosus development allowed for the formation of a
significant number of researchers, some of whom continue to work on the
developmental biology of parasites and also formed new generations that have
contributed to the knowledge of fundamental issues, like the determination of the
anterior-posterior axis in these species (see for example Esteves et al., 1993; Martínez
et al., 1997). He also started the first undergraduate and graduate courses on
molecular biology in Uruguay. The first students of these courses are now working as
teachers or researchers throughout the country. In addition, Ehrlich promoted the
temporary stay, and doctoral thesis completion, of Guillermo Oliver (now in the USA),
who he co-tutored with E. M. De Robertis (Oliver et al., 1992). Oliver's passage through
the country strongly influenced young people at the lab, awakening their interest on a
then newborn subject, evo-devo.

In addition to PEDECIBA, the return to democracy also brought the opportunity for the
University to create an ambitious program for the development of research, the CSIC
(Comisión Sectorial de Investigación Científica), which among many other things,
provided funds for research grants. The government also modestly supported research
through different programs along this time, until the creation in 2006 of the National
Agency for Research and Innovation (ANII, Agencia Nacional de Investigación e
Innovación), allowing for a much greater investment on research grants and graduate
fellowships. The same year witnessed another breakthrough, the foundation of the
Institut Pasteur Montevideo; academically linked to its homonymous in Paris and
funded by the Uruguayan government, is based on open platforms equipped with
modern and high-cost technology, some of which was not available before.

It is necessary to highlight the influence of the developmental biologists in neighboring
countries in the initial, and especially recent, development of the discipline in Uruguay.
A strong will to scientifically collaborate was re-ignited in the countries of the Southern
Cone since the return to democracy, and regarding Developmental Biology in Uruguay,
there was a great influence of Argentinian and Chilean researchers. As an example, we
remark the initial organization of a regional course by Roberto Mayor (now at UCL) and
Miguel Allende in Santiago de Chile in January, 1999. That course, repeated biennially
later, was tremendously influential for all participants, to the point that it can be
probably recognized as the seed of the current Latin American Society for
Developmental Biology (LASDB), founded in 2003. In 2012, the LASDB meeting took
place in Montevideo, preceded by an impressive international training course centered on evo-devo (Fig. 2). These courses, LASDB, and the contacts they generated have undoubtedly fostered developmental biology in Uruguay, like in other regional countries. In addition to this, several other more specific international events related to development have taken place in Uruguay in the last years: a series of Symposia on Neural Development and Plasticity (2008, 2012); the First Meeting of the Latin American Zebrafish Network (2010); two Symposia on Annual Fishes (2010, 2015); and the First Latin American Worm Meeting (2017).

A present looking into the future: the flight

As a consequence of the creation of the above mentioned academic and funding agencies and the progressive ingression of the local scientific community into the global network, including international collaborations and training of young researchers, at present there are several research groups working on different areas of developmental biology, distributed among several institutions. In addition to work on “classical” model organisms, such as rodents, chick, zebrafish, D. melanogaster, C. elegans and A. thaliana many of them incorporated only in the last 10-15 years, local developmental biology has shown a special strength in not so traditional, but equally, or even more important species, such as parasitic worms or electric and annual fishes. As examples of this, we mention below some recent contributions from different currently active groups.

On the large field of neural development, we find research on vertebrates, such as spinal cord neurogenesis and regeneration in turtles and rats (Fabbiani et al., 2018; Fabbiani et al., 2020), neurogenesis and late development of the olfactory/visual systems in annual fishes (Torres-Pérez et al., 2017), neurogenesis in the electric fish Gymnotus omarorum (Olivera-Pasilio et al., 2017), the innervation plasticity of the rat uterus (Brauer, 2016; Richeri et al., 2020), developmental plasticity of the visual pathway in mice (Vierci et al., 2016), the role of oxidative stress-response genes in neural development in rats and zebrafish (López et al., 2018), the role of cell polarity in neurulation and retinal differentiation in chick and zebrafish (Aparicio et al., 2018; Lepanto et al., 2016; see also Aparicio et al. in this issue), and synaptogenesis and different aspects of neural development in Drosophila (Cantera et al., 2014; Pazos
On reproductive aspects of development, other groups are working on mammalian spermatogenesis and meiosis from a molecular point of view (Trovero et al., 2020), sex differentiation and gonadal development in fishes (Vizziano-Cantonnet et al., 2016) and on the neuroendocrine regulation of ovarian innervation in rats (Chávez-Genaro and Anesetti, 2018). On different aspects of molecular developmental biology in invertebrates, we highlight the already mentioned continuing research on parasitic worms such as cestodes (Chalar et al., 2016; Montagne et al., 2019; Preza et al., 2018), and in Drosophila (Bolatto et al., 2015). The annual fishes are a particular case, where several groups at Facultad de Ciencias are contributing to understand the particular developmental stages, the molecular regulation of the diapauses and the functional genomics of the early stages and sexual determination of members of the genus Austrolebias (Arezo et al., 2014; Arezo et al., 2017). In association with the Center for Genome Regulation (University of Chile), they are currently finishing the sequencing and assembly of the genome and basic transcriptome of one of these species, A. charrua.

Concluding remarks
We have attempted here a summarized account of the “non-canonical” advent of developmental biology in Uruguay, not directly from embryology, but rather from other disciplines such as cytology and physiology, later reinforced by the return of researchers from exile. The schools they initiated, added to the interaction with regional and international developmental biologists, led to the several research groups we find today. As these groups grow, and students graduate or return from training abroad, a sustained diversification and strengthening of Uruguayan developmental biology is expected in the years to come.

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References

DE ANDA G, REBOLLO MA (1968). Histochemistry of the neuromuscular spindles in the chicken during development. *Acta Histochem* 31: 287–295.

APARICIO G, ARRUTI C, ZOLESSI FR (2018). MARCKS phosphorylation by PKC strongly impairs cell polarity in the chick neural plate. *genesis* 56: e23104.

AREZO MJ, PAPA N, GUTTIERREZ V, GARCÍA G, BEROIS N (2014). Sex determination in annual fishes: Searching for the master sex-determining gene in Austrolebias charrua (Cyprinodontiformes, Rivulidae). *Genet Mol Biol* 37: 364–374.

AREZO MJ, PAPA NG, BEROIS N, CLIVIO G, MONTAGNE J, DE LA PIEDRA S (2017). Annual killifish adaptations to ephemeral environments: Diapause I in two Austrolebias species. *Dev Dyn* 246: 848–857.

ARRUTI C, COURTOIS Y (1978). Morphological changes and growth stimulation of Bovine Epithelial Lens cells by a retinal extract in vitro. *Exp Cell Res* 117: 283–292.

BENAVENTE R, WETTSTEIN R (1977). An ultrastructural cytogenetic study on the evolution of sex chromosomes during the spermatogenesis of Lycosa malitiosa (Arachnida). *Chromosoma* 64: 255–277.

BERETTA CURI A (2006). *Roberto Caldeyro Barcia: el mandato de una vocación*. PEDECIBA/Ediciones Trilce, Montevideo.

BOLATTO C, PARADA C, REVELLO F, ZUÑIGA A, CABRERA P, CAMBIAZO V (2015). Spatial and temporal distribution of Patched-related protein in the Drosophila embryo. *Gene Expr Patterns* 19: 120–128.

BRAUER M (2016). Plasticity in Uterine Innervation: State of the Art. *Curr Protein Pept Sci* 18: 108–119.

BUÑO W, GOYENA H (1955). Effect of Cortisone upon Growth in vitro of Femur of the Chick Embryo. *Proc Soc Exp Biol Med* 89: 622–625.

CALCAGNO M, GOYENA H, ARRAMBIDE E, ARRUTI DE URSE C (1970). Action of cortisone and cortisol upon biosynthesis of chondroitin sulfate in femur in vitro cultures of chick embryo. *Exp Cell Res* 63: 131–137.

CALDEYRO-BARCIA R, POSEIRO JJ (1959). OXYTOCIN AND CONTRACTILITY OF THE PREGNANT HUMAN UTERUS. *Ann N Y Acad Sci* 75: 813–830.

CAMPOS MUÑOZ A (2013). Eduardo de Robertis. En el centenario de su nacimiento (1913-2013). *Actual Medica* 790: 167–170.
CANTERA R, FERREIRO MJ, ARANSAY AM, BARRIO R (2014). Global gene expression shift during the transition from early neural development to late neuronal differentiation in Drosophila melanogaster. *PLoS One* 9.

CASTELLANO MA, GERMINO NI, BEROIS DE HARO N, GERARD G (1969). Histochemical demonstration of l-amino acid-tetrazolium reductase. *Histochemie* 18: 277–280.

CASTELLANO MA, TÓRTORA JL, GERMINO NI, RAMA F, OHANIAN C (1973). The effects of isonicotinic acid hydrazide on the early chick embryo. *Development* 29.

CHALAR C, MARTÍNEZ C, BRAUER MM, EHRLICH R, MARÍN M (2016). Eghbx2, a homeobox gene involved in the maturation of calcified structures in Echinococcus granulosus. *Gene Reports* 3: 39–46.

CHÁVEZ-GENARO R, ANESETTI G (2018). First ovarian response to gonadotrophin stimulation in rats exposed to neonatal androgen excess. *J Mol Histol*.

CIRILLO A, ARRUTI C, COURTOIS Y, JEANNY JC (1990). Localization of basic fibroblast growth factor binding sites in the chick embryonic neural retina. *Differentiation* 45: 161–167.

DRETS ME (2013). Francisco Alberto Sáez - Primer citogenetista de América Latina.

DIRAC, Facultad de Ciencias, Montevideo.

ESTABLE C, ACOSTA-FERREIRA W, SOTELO JR (1957). An electron microscope study of the regenerating nerve fibers. *Zeitschrift für Zellforsch und Mikroskopische Anat* 46: 387–399.

ESTEVES A, DALLAGIOVANNA B, EHRLICH R (1993). A developmentally regulated gene of Echinococcus granulosus codes for a 15.5-kilodalton polypeptide related to fatty acid binding proteins. *Mol Biochem Parasitol* 58: 215–222.

FABBIANI G, REALI C, VALENTÍN-KAHAN A, REHERMANN MI, FAGETTI J, FALCO MV, RUSSO RE (2020). Connexin signaling is involved in the reactivation of a latent stem cell niche after spinal cord injury. *J Neurosci* 40: 2246–2258.

FABBIANI G, REHERMANN MI, ALDECOSEA C, TRUJILLO-CENÓZ O, RUSSO RE (2018). Emergence of serotonergic neurons after spinal cord injury in turtles. *Front Neural Circuits* 12.

GARCIA-AUSTT E (1954). Development of Electrical Activity in Cerebral Hemispheres of the Chick Embryo. *Proc Soc Exp Biol Med* 86: 348–352.

LEPANTO P, DAVISON C, CASANOVA G, BADANO JL, ZOLESSI FR (2016).
Characterization of primary cilia during the differentiation of retinal ganglion cells in the zebrafish. *Neural Dev* 11: 10.

LÓPEZ L, ZULUAGA MJ, LAGOS P, AGRATI D, BEDÓ G (2018). The Expression of Hypoxia-Induced Gene 1 (Higd1a) in the Central Nervous System of Male and Female Rats Differs According to Age. *J Mol Neurosci* 66: 462–473.

DE MARIA A, ARRUTI C (1995). α-crystallin polypeptides in developing chicken lens cells. *Exp Eye Res* 61: 181–187.

MARTÍNEZ C, CHALAR C, GONZÁLEZ J, EHRLLICH R (1997). The homeobox-containing gene EgHbx3 from Echinococcus granulosus is expressed in the stalk of protoscoleces. *Int J Parasitol* 27: 1379–1381.

MELAMED J, TRUJILLO-CENÓZ O (1975). The fine structure of the eye imaginal disks in muscoid flies. *J Ultrastruct Res* 51: 79–93.

MONTAGNE J, PREZA M, CASTILLO E, BREHM K, KOZIOL U (2019). Divergent Axin and GSK-3 paralogs in the beta-catenin destruction complexes of tapeworms. *Dev Genes Evol* 229: 89–102.

OLIVER G, VISPO M, MAILHOS A, MARTÍNEZ C, SOSA-PINEDA B, FIELITZ W, EHRLLICH R (1992). Homeoboxes in flatworms. *Gene* 121: 337–342.

OLIVERA-PASILIO V, LASSERRE M, CASTELLÓ ME (2017). Cell proliferation, migration, and neurogenesis in the adult brain of the pulse type weakly electric fish, Gymnotus omarorum. *Front Neurosci* 11.

PAZOS OBREGÓN F, PALAZZO M, SOTO P, GUERBEROFF G, YANKILEVICH P, CANTERA R (2019). An improved catalogue of putative synaptic genes defined exclusively by temporal transcription profiles through an ensemble machine learning approach. *BMC Genomics* 20.

PELLEGRINO V, KLIMAVICIUS S, VIQUE MI, VARELA G (Eds) (2017). *Científicos uruguayos: destacadas contribuciones al conocimiento biológico durante los siglos xix y xx*. Grupo Magro Editores, Montevideo.

PREZA M, MONTAGNE J, COSTÁBILE A, IRIARTE A, CASTILLO E, KOZIOL U (2018). Analysis of classical neurotransmitter markers in tapeworms: Evidence for extensive loss of neurotransmitter pathways. *Int J Parasitol* 48: 979–992.

RICHÉR I, VIERCI G, MARTÍNEZ GF, LATORRE MP, CHALAR C, BRAUER MM (2020). Neuropilin-1 receptor in the rapid and selective estrogen estrogen-induced
neurovascular remodeling of rat uterus. *Cell Tissue Res.*

DE ROBERTIS E (1956). Morphogenesis of the retinal rods; an electron microscope study. *J Biophys Biochem Cytol* 2: 209–218.

SOTELO JR (1959). An electron microscope study on the cytoplasmic and nuclear components of rat primary oocytes. *Zeitschrift für Zellforsch und Mikroskopische Anat* 50: 749–765.

SOTELO JR, TRUJILLO-CENÓZ O (1960). Electron microscope study on spermatogenesis - Chromosome morphogenesis at the onset of meiosis (cyte I) and nuclear structure of early and late spermatids. *Zeitschrift für Zellforsch und Mikroskopische Anat* 51: 243–277.

SOTELO JR, TRUJILLO-CENÓZ O (1958). Electron microscope study on the development of ciliary components of the neural epithelium of the chick embryo. *Zeitschrift für Zellforsch und Mikroskopische Anat* 49: 1–12.

SOTELO JR, WETTSTEIN R (1964). Electron microscope study on meiosis - The sex chromosome in spermatocytes, spermatids and oocytes of Gryllus argentinus. *Chromosoma* 15: 389–415.

SPINDLER MC, FILBECK S, STIGLOHER C, BENAVENTE R (2019). Quantitative basis of meiotic chromosome synapsis analyzed by electron tomography. *Sci Rep* 9.

TORRES-PÉREZ M, ROSILLO JC, BERROSTEGUIETA I, OLIVERA-BRAVO S, CASANOVA G, GARCÍA-VERDUGO JM, FERNÁNDEZ AS (2017). Stem cells distribution, cellular proliferation and migration in the adult Austrolebias charrua brain. *Brain Res* 1673: 11–22.

TROVERO MF, RODRÍGUEZ-CASURIAGA R, ROMEO C, SANTIÑAQUE FF, FRANÇOIS M, FOLLE GA, BENAVENTE R, SOTELO-SILVEIRA JR, GEISINGER A (2020). Revealing stage-specific expression patterns of long noncoding RNAs along mouse spermatogenesis. *RNA Biol* 17: 350–365.

TRUJILLO-CENÓZ O, MELAMED J (1978). Development of photoreceptor patterns in the compound eyes of muscoid flies. *J Ultrastruct Res* 64: 46–62.

VÁZQUEZ-NIN GH, SOTELO JR (1968). Electron microscope study of the developing nerve terminals in the acoustic organs of the chick embryo. *Zeitschrift für Zellforsch und Mikroskopische Anat* 92: 325–338.

VIERCI G, PANNUNZIO B, BORNIA N, ROSSI FM (2016). H3 and H4 lysine acetylation
correlates with developmental and experimentally induced adult experience-dependent plasticity in the mouse visual cortex. *J Exp Neurosci* 2016: 49–64.

VIZZIANO-CANTONNET D, DI LANDRO S, LASALLE A, MARTÍNEZ A, MAZZONI TS, QUAGIO-GRASSIOTTO I (2016). Identification of the molecular sex-differentiation period in the siberian sturgeon. *Mol Reprod Dev* 83: 19–36.

WETTSTEIN R, SOTELO JR (1967). Electron microscope serial reconstruction of spermatocyte nuclei at pachytene. *J Microsc* 6: 557–576.

ZOLESSI FR, ARRUTI C (2001). Sustained phosphorylation of MARCKS in differentiating neurogenic regions during chick embryo development. *Brain Res Dev Brain Res* 130: 257–267.
Fig. 1. The beginnings. (A) Part of the scientific staff of the II BCE ca. 1956: 1, Raúl Vaz Ferreira; 2, Oscar Vincent; 3, Eduardo Pagani; 4, Alvaro Díaz Maynard; 5, Aída Pintos; 6, Amalia Laguardia; 7, Margarita Pérez; 8, Nadir Brum; 9, José Sas; 10, Omar Trujillo-Cenóz; 11, Br. Amatti; 12, Aníbal Alvarez; 13, Elsa Trinkle; 14, Gloria Martínez; 15, Adela Wittemberger; 16, Mercedes Rovira; 17, Nilsa Iriondo; 18, Hugo Freire; 19, Tomasa Rodríguez; 20, José María Martínez; 21, Walter Acosta Ferreira; 22, Héctor
Franco Raffo; 23, Máximo E. Drets; 24, Nubia F. de San Germán; 25, María Isabel Ardaó; 26, Clemente Estable; 27, Francisco Alberto Sáez; 28, J. Roberto Sotelo. (B) Eduardo De Robertis, in front of an electron microscope. (C) Giuseppe Levi, hiking. (D) Washington Buño. (E) School of Medicine (Facultad de Medicina) building in Montevideo, seen from the north-east corner where, from the first floor up is still located the Department of Histology and Embryology. The Tissue Culture Laboratory, leaded first by Horacio Goyena and later by Cristina Arruti, occupied the top of the tower. (F) Roberto Caldeyro Barcia at the Electronics Department. A, IIBCE archive, key and names taken from Drets, 2013; B, from Campos Muñoz, 2013; C, from www.nico.ottolenghi.unito.it; D, drawing by M. Picón, from the Dean office, Facultad de Medicina, Universidad de la República; F, from Beretta Curi, 2006.
Fig. 2. The LASDB-SDB International Course in 2012: A Systems Biology Approach to Understanding Mechanisms of Organismal Evolution. All the students and most of the faculty seated at the entrance stairs of the Institut Pasteur Montevideo. For space reasons, we name here only the faculty and collaborators; in parentheses, country of instructors from abroad: 21, Gonzalo Aparicio; 22, Heather Marlow (Germany); 23, Daniel Prieto; 28, Brigitte Galliot (Switzerland); 29, Soledad Astrada; 30, Andrea Toledo; 34, Nicolás Papa; 36, Katia Del Río-Tsonis (USA); 37, Nipam Patel (USA); 38, Alejandro Sánchez-Alvarado (USA); 39, Billie Swalla (USA); 40, Ida Chow (USA); 41, Flavio Zolessi; 42, Panagiotis Tsonis (USA); 43, Detlev Arendt (Germany); 44, Rolando Rivera-Pomar (Argentina); 45; Nibia Berois; 46, María José Arezo. Faculty members missing in the photo: Luis Acerenza; Siobhan Brady (USA); María E. Castelló; Estela Castillo; Yolanda P. Cruz (USA); Walter J. Gehring (Switzerland); Enrique P. Lessa; Claudio D. Stern (UK); Andrea Streit (UK).