ASCIDIAN NEWS*

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A very big thank you to the many AN readers who sent in contributions and for letting me know how important AN continues to be! It is gratifying to know that it continues to be useful for so many.

There are **108** new publications listed at the end of this newsletter. Although the title of this newsletter is Ascidian News, I also include papers on the pelagic Tunicata: the pyrosomes, doliolids, salps and appendicularians, and the subphylum Cephalochordata: *Amphioxus* and *Branchiostoma*, because these publications include important work on phylogenetics and evolution of the primitive chordates.

Please keep in touch and continue to send me contributions for the next issue: abstracts of recent meetings, work in progress, student thesis abstracts. To be assured of your new publications being included, always send me the pdf.

***Ascidian News is not part of the scientific literature and should not be cited as such.**

NEWS AND VIEWS

1. It is almost time for the **Intl. Tunicata meeting, ITM 13, in Berlin, Germany, July 19-24.** Organizer Dr. Thomas Stach (thomas.stach@hu-berlin.de). Late registration may still be open. Ahead of the upcoming meeting, we launched **'Tunicata 101: Building Foundations for the Berlin Meeting,** a series of three Zoom lectures aimed at providing students and early-career researchers with a foundation in key areas of tunicate biology. The series features lectures by Prof. Marie Nydam on tunicate diversity and evolution and Prof. Takehiro Kusakabe on ascidian embryology and neurobiology. A final session by Prof. Thomas Stach is scheduled for June 4th. The lectures brought together participants from across the globe and provided an opportunity for learning, discussion, and community building in advance of the Berlin meeting. Recording of the lectures are available through the website https://www.itm-berlin-2026.de/ .

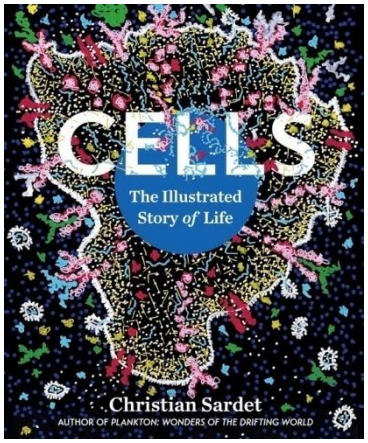
2. Society for the Study of Marine Bioinvasions: We're excited to announce that the next **International Congress on Marine Bioinvasions, ICMB XIII, will be held in Cape Town South Africa 5-7 October 2027,** at the Breakwater Lodge Hotel and Conference Centre! Join our mailing list: https://marinebioinvasions.info/contact/email-list. The website is https://marinebioinvasions.info/.

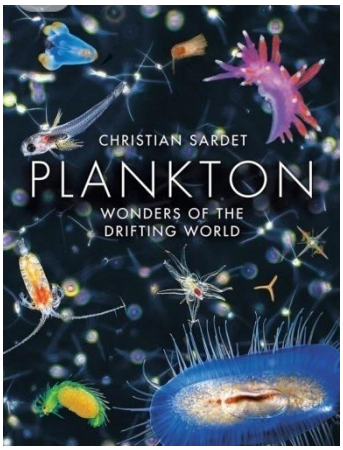
3. Farming sea squirts. At the recent SEAFOOD Expo Global/Seafood Processing Global in Barcelona was an exhibit by Smart Farm, whose specialist farming and harvesting system is

used in more than 20 countries around the world, primarily for growing mussels. The Smart Farm system comprises nets suspended up to 30 metres deep from long black pipes floating on the water surface, and a bespoke harvesting machine, which enables husbandry and harvesting to be done entirely in the water. This set-up also makes it easier to reduce the impact of fouling and predators. Prominent on the Smart Farm stand this year was a film featuring their SmartUnits being used for tunicate (sea squirt) farming in Norway. Pronofa ASA, which is based in both Norway and Sweden, has worked with Smart Farm to help perfect a system for growing and harvesting *Ciona intestinalis*, the vase tunicate. This species grows naturally along the coasts of Norway and Sweden and easily colonises the growing nets, producing up to 82kg of biomass per square metre. It is also fast growing, with a one-year harvest cycle.

Pronofa processes the tunicates into a product similar to traditional minced meat, under their food brand Purply. This mimics the taste, nutritional value and uses of minced meat, so can be incorporated in dishes such as lasagne as an alternative to meat, or turned into products like burgers, tacos and meatballs. See https://www.pressreader.com/uk/fish-farmer/20260514/281994679121681 for the complete article.

4. Christian Sardet (csardet@gmail.com) now retired from the Villefranche-sur-Mer Marine Station, contributed two papers on the history of art and science on the French Riviera and the Villefranche Marine Station. See the New Publications list at the end of this newsletter for the references. The papers are open access. “I am no more active in research but keep sharing about plankton and cells via conferences, exhibits, films and books. My new art&science book “Cells- The Illustrated Story of Life“ with 50 of my original drawings and 200 illustrations. It is now available in print and PDF forms for less than 30 dollars:
https://theexperimentpublishing.com/catalogs/fall-2025/cells/
www.planktonchronicles.org





5. Hero dog 'first in world' to detect invasive sea vomit threatening Britain waters.

Dr Texa Sim has trained her pup to sniff out the carpet-like sea squirt. A mountain search and rescue responder has trained her hero dog to sniff out invasive “sea vomit” in what is believed to be a world first. Dr Texa Sim’s two-year-old labrador, Uisge, can detect *Didemnum vexillum*, an invasive marine organism that has been found in UK waters.The species, commonly known as sea vomit, originates from Japan and is often transported across the world on the hulls of ships. This fast-spreading sea squirt can clone itself, forming thick, carpet-like colonies that

smother the seabed. If left unchecked, it can blanket entire areas, driving out native species and damaging biodiversity. It also poses serious problems for the fishing industry, particularly shellfish farms. https://www.express.co.uk/news/uk/2171131/hero-dog-first-world-detect

6. We are very saddened to read of the passing of **Todd Newberry**, who worked on and published a number of papers on ascidians including his doctoral thesis. From his obituary: "Todd Newberry, a professor emeritus of biology and a founding member of the faculty at the University of California, Santa Cruz, died on Feb. 9, 2026. He was 90 years old." You can read the lengthy and interesting obituary at https://news.ucsc.edu/2026/02/in-memoriam-todd-newberry/ which includes several excellent photos.

7. The conference book containing all the **abstracts for the 12th Intl. Conference on Marine Bioinvasion**s, which was held 7-9 Oct. 2025 in Madeira, is now available online. https://marinebioinvasions.info/user/pages/60.archives/ICMB-XII.pdf

8. Korea's Raw Delicacy Is a Billion-Dollar Pest Abroad. Korea is the only country in the world where people prepare the sea squirt ***Styela clava*** raw and enjoy it as sashimi. Abroad, however, the organism is known as a "pirate creature that clings to the bottom of ships." See the website for the entire article. https://en.sedaily.com/society/2026/04/04/koreas-raw-delicacy-is-a-billion-dollar-pest-abroad

9. New study reveals an unsuspected range of behaviors in the sea squirt *Ciona*.
"The sea squirt *Ciona intestinalis*, a small, sessile marine animal closely related to vertebrates, is found along the Norwegian coast where it lives attached to mooring ropes and other hard surfaces. While being a key part of marine ecosystems, sessile animals are often overlooked by researchers and the public. For that reason, researchers Oleg Tolstenkov and Sissel Norland from the Chatzigeorgiou research group at the Michael Sars Centre decided to investigate whether there was more than meets the eye to a sea squirt's daily routine."

"Although these animals look very simple and immobile, we found that they actually display a surprisingly rich range of behaviors", Tolstenkov said. "Honestly, we did not expect to find such a rich behavioral repertoire. We definitely underestimated those sea squirts!"

See the full article at: https://www.uib.no/en/michaelsarscentre/183065/new-study-reveals-unsuspected-range-behaviors-sea-squirt-ciona .

Work in Progress

1. From **Alison Murray**, Division of Earth and Ecosystem Sciences, DRI, Reno, NV, USA. Alison.Murray@dri.edu

Antarctic Peninsula ascidian-focused research expedition. Between Feb 19-March 26th, our interdisciplinary team embarked from Punta Areas, Chile on a 35-day expedition on the R/V Sikuliaq to study Antarctic ascidians, their natural products and host-associated microbiomes. In particular, this work focuses on the palmerolide A-producing ascidian,*Synoicum adareanum*. Palmerolide A is a bioactive compound with anti-cancer potential produced by a bacterium that lives in association with this polyclinid ascidian. We are now studying the biogeographical extent of the palmerolide-microbe association, comparative 'omics with several ascidian morphotypes and species, and working to understand the potential ecophysiological roles of palmerolide A in this ascidian-microbe association. Activities on board included SCUBA diving, remotely operated vehicle (ROV) explorations, microscopy, microbial cultivation and molecular biology efforts (natural product genetic tracking and DNA sequencing). The homepage for the project is https://www.dri.edu/life-in-antarctica-a2mp/ where you can find a link to follow along the journey through our daily story map. A more extensive description of the project was posted by the Univ. of S. Florida: https://phys.org/news/2026-06-antarctica-advances-potential-melanoma-treatment.html

Meetings Abstracts

1. XXVI Meetng of the Italian Associaton of Developmental and Comparatve Immunobiology (IADCI).

a) Immune priming in the colonial ascidian *Botryllus schlosseri*. Peronato, Anna; Franchi, Nicola; Drago, Laura; Domenichi, Simone; Venier, Paola; Bortoletto, Enrico; Ballarin, Loriano. Dept. of Biol., Univ. of Padova, Padova, Italy.

Immune priming is the capability of the innate immune system to use the information on the molecular structure of nonself, acquired in the course of a past experience, to give rise to an enhanced immune response in the case of a second exposure to related or unrelated molecule/cell. It can be considered a short-term immune memory, highly differing from the long-term, lymphocyte-based, immune memory of jawed vertebrates. In the last decades, immune priming has been described in a variety of invertebrates. Nevertheless, studies on immune priming rely on a few parameters or the expression of a low number of genes involved in immune responses, such as those codifying pattern recognition receptors or compment factors. In addition, the molecular mechanisms at the basis of the phenomenon are, in many cases, far from being understood. In the attempt to demonstrate the presence of immune priming in colonial ascidians, we microinjected colonies of B. schlosseri with bacteria or LPS from Escherichia coli and compared the obtained response, in terms of transcription of immune-related genes, to that obtained following a second challenge. The responses in various phases of the colonial blastogenetic cycle were also considered. Results indicate the capability of B. schlosseri to enhance the transcription of immune-related genes after a first challenge with nonself and pave the way for future analysis of the mechanisms underlying the process.

b) Effects of polyethylene nanoplastics on immune- and stress-related genes of ascidians. Sabbadin, Giacomo1; Sunanaga, Takeshi2; Fujiwara, Shigeki2; Ballarin, Loriano1. 1Dept. of Biol., Univ. of Padova, Padova, Italy; 2Fac. of Sci. and Technol., Kochi Univ., Japan.

The increasing levels of plastic pollution, particularly in the form of micro- and nanoplastics (NPs), represents a growing threat to aquatic ecosystems, as these particles persist in the environment and interact with multiple biological compartments. Despite their ecological relevance, the effects of these anthropogenic stressors on immunity, development, and physiological resilience remain poorly understood in sessile filter-feeding invertebrates such as ascidians, which can directly uptake NPs from the surrounding environment during feeding. This work aims to study the effects of NP pollution on the expression of immunity- and stress-related genes in two species of ascidians Polyandrocarpa misakiensis and Ciona robusta. Animals were collected in the coastal area near the Usa Marine Biological Institute of Kochi University (Japan). Ascidians were exposed, in the laboratory, to NPs for a week at a 0.5 µg/mL concentration. After exposure, the gut and pharynx of Ciona as well as the buds and zooids of Polyandrocarpa were separately collected and the expression of the genes of interest was analyzed through qRT-PCR. Results show how NPs exposure can alter the expression of some of these genes; results indicate also a different effect depending on the type of tissue analyzed.

c) MAPK signaling integrates immune and stress responses during colonial blastogenesis in Botryllus schlosseri. Drago, Laura1; Frisoni, Francesca2; Santovito, Gianfranco1; Ballarin,Loriano1. 1Dept. of Biol., Univ. of Padova, Padova, Italy; 2Dept. of Biol., Univ. of Konstanz, Konstanz, Germany.

Mitogen-activated protein kinase (MAPK) pathways are evolutionarily conserved signaling cascades that regulate key cellular processes such as proliferation, differentiation, and stress responses. In the ascidian Botryllus schlosseri, the colony consists of repeated functional units called zooids, which are continuously replaced through blastogenesis, a cyclical developmental process requiring immune-mediated clearance of aging cells. We identified two MAPK genes, bsmapk-1 and bsmapk-8, homologous to vertebrate erk and jnk, and examined their transcriptional dynamics throughout the blastogenetic cycle. The two genes showed distinct temporal patterns and differential responsiveness to immune stimulation and environmental stressors, suggesting pathway-specific roles. Functional inhibition of ERK and JNK signalling impaired hemocyte phagocytosis, altered the expression of genes involved in oxidative stress defense (sod, gpx-5) and stress granule dynamics (tiar, ttp), and disrupted normal blastogenesis, resulting in reduced zooid growth and developmental arrest. These findings demonstrate that MAPK pathways act as critical integrators of immune function, stress responses, and developmental renewal, ensuring the coordinated turnover and maintenance of colonial modules in a non-vertebrate chordate.

2. Seventh Scientific Retreat, Dept. of Biology, Univ. of Padova, Italy, 12-13 May, 2026.

a) Identifying the biosynthetic origins of tunichromes, an enigmatic group of natural products. András Zeke[1], Ilaria Battisti[2], Annj Zamuner[3], Laura Drago[1], Paola Brun[4], Monica Dettin[3], Laura Cendron[1], Elisabetta Schievano[5], Loriano Ballarin[1], Giorgio Arrigoni[2], Lucia

Manni[1]. [1]Dept. of Biol., [2]Dept. of Biomedical Sci., [3]Dept. of Industrial Engineering, [4]Dept. of Molecular Medicine, [5]Dept. of Chemical Sci.,Univ. of Padua, Italy.
Introduction: Tunichromes are poorly understood marine natural products identifiedin the hemolymph cells of various tunicate species. These short, highly modified oligopeptides contain DOPA and dehydroDOPA moieties. These features make themoxidatively unstable and prone to covalent crosslinking but also render them excellentmetal chelators.
Methods: We applied a combination of bioinformatics, tissue sampling, proteomics and gene expression analyses.
Results: We present compelling evidence that tunichromes arise from much longer,genomically encoded, ribosomally produced and post-translationally modified precursor proteins (RiPPs). While RiPPs are common in bacteria and also occur in plants and fungi, they are almost unknown in the animal kingdom. A deeper exploration of tunicate genomes and transcriptomes also reveals a wealth of novel precursor proteins belonging to a large number of hitherto unidentified products.Surprisingly, antimicrobial peptides are often produced by the same precursors, strongly suggesting tunichromes to be defensive compounds against microbes. This is also supported by their expression patterns. What is more, even closely related species tend to produce different products, as shown by rapid evolutionary shuffling of sequences, gene duplications, de novo evolution of precursors, as well as proteomic analyses.
Conclusions: Our findings suggest a new, untapped source of marine natural products for antimicrobial discovery.

b) Perspectives in the study of anthropogenic stressors on marineinvertebrates.
Sabbadin Giacomo, Domenichi Simone, Ballarin Loriano, Manni Lucia. Dept. of Biol., University of Padova. giacomo.sabbadin.1@studenti.unipd.it

Marine ecosystems are threatened by climate change and pollutants. While much research focuses on vertebrates, invertebrates are less studied despite their fundamental ecological roles. In the context of various research projects, we aim to study the effects of some anthropogenic stressors, i.e. heat waves, underwater noise, and nanoplastics, in sessile filter-feeding invertebrates such as the ascidian *Botryllusschlosseri*, a model organism, bred and studied for years at the Department of Biology,University of Padova. We are studying the effects of these anthropogenic stressors on physiological responses (immunity and stress responses, filter feeding activity) and on the colonial blastogenetic cycle and regenerative capabilities. Results obtained so far suggest that these stressors negatively affect both physiological and asexual reproduction/regeneration parameters. In the future, we will investigate the above-affected processes and study the combined effects of the stressors, both in lab and inthe field.

Thesis Abstracts

1. INVESTIGATION OF DIDEMNID SPECIES ALONG THE TURKISH COASTLINES UTILIZING GENETIC, ENVIRONMENTAL, AND MORPHOLOGICAL DATA. İrem Bekdemir, M.S. degree, Marine Biology and Fisheries, Middle East Technical University, Turkey. Supervisor: Assoc. Prof. Dr. Arzu Karahan. arzukar@metu.edu.tr

The **Didemnidae** family is the most speciose and taxonomically challenging group within the class Ascidiacea (Tunicata), because of the small size of zooids and the lack of diagnostic external characters. This thesis presents the **first comprehensive and integrative**

assessment of the biogeographic distribution and diversity of the Didemnidae family along the Turkish coastline. An integrative approach combining **morphological analyses** (including spicule morphology) with **molecular analyses** based on the mitochondrial cytochrome *c* oxidase subunit I (COI) gene was applied, together with **population genetic analyses** of *Didemnum pseudovexillum*.

A total of 524 **samples** were collected from 20 stations across the Black Sea, Marmara Sea, Aegean Sea, and Mediterranean Sea. Species delimitation analyses using **ASAP, PTP**, and **Bayesian phylogenetic inference** identified **22 species-level lineages**, of which **17 represent putative new species** for the region. Additionally, **four known species** were genetically recorded, three of them for the first time in Turkish waters. Genetic distance analyses were conducted to evaluate interspecific divergence, and neutrality tests were applied to assess the phylogeographic structure of *D. pseudovexillum*.

No didemnids were recorded from the Black Sea, whereas the **Mediterranean Sea showed the highest species richness**. A positive relationship was detected between species richness and salinity. Phylogeographic analyses of *D. Pseudovexillum* suggest long-term demographic stability. Southeastern populations showed significant genetic structuring, whereas western populations exhibited higher connectivity. Overall, the results highlight the importance of integrative taxonomic approaches for accurately documenting Didemnidae biodiversity.
Keywords:COI, physicochemical parameters, morphology, species delimitation, phylogeography. January 2026, 134 pages

2. DIVERSITY, POPULATION STRUCTURE, AND HOLOBIONT ORGANIZATION OF BOTRYLLID ASCIDIANS WITH A FOCUS ON *BOTRYLLUS SCHLOSSERI* IN RELATION TO ENVIRONMENTAL PARAMETERS ALONG TURKISH COASTS. Tohumcu, Begüm Ece, Ph.D., Dept. of Marine Sciences and Fisheries, Middle East Technical University, Turkey. Supervisor: Assoc. Prof. Dr. Arzu Karahan arzukar@metu.edu.tr

Botryllid ascidians, especially *Botryllus schlosseri*, are used as model organisms for studies on immune system evolution, developmental biology, and whole-body regeneration. Ecologically, botryllid ascidians, as sessile filter feeders, serve as efficient bioindicators in coastal ecosystems and exist as holobionts that harbor diverse microbial communities within. In addition to their ecological and evolutionary significance, detailed information regarding botryllid diversity and the genetic composition of *B. schl*osseri populations in Turkish coastal waters has remained limited locally.

In this thesis context, the diversity of botryllid ascidians and the population structure of *B. schlosseri* along the Turkish coastline, including the Black Sea, Sea of Marmara, Aegean Sea, and Eastern Mediterranean, were studied. Specimens were located in between 2023 and 2024 from 26 stations selected as representative of diverse hydrographic conditions and microhabitats. Phenotypic variability was evaluated through morphotype analyses, whereas mitochondrial COI sequences and microsatellite markers were employed to assess genetic diversity and population structure. Host-associated microbial communities were characterized through 16S and 18S rRNA amplicon sequencing, and environmental variables were incorporated to define their holobiont system components.

The findings indicate significant environmental variability among basins and microhabitats. Eight species (*Botrylloides niger*, *Botrylloides israeliensis*, *Botryllus humilis*, *Botryllus schlosseri*, *Botryllus gaiae*, *Botrylloides* sp. (Saudi Arabia, ON053356)), *Symplegma brakenhielmi*, and *Polyclinum constellatum*) were reported, along with an additional botryllid

lineage identified at the Konacık station that could not be aligned with existing reference databases. *B. schlosseri* was the sole botryllid species exhibiting a complete north-south distribution, albeit with an uneven presence. The research additionally documents the first record of *B.gaiae* in Turkish coastal waters. Population analyses revealed significant genetic diversity in *B. schlosseri*, characterized by 83 distinct COI haplotypes and pronounced spatial heterogeneity. A total of 68 morphotypes were recorded, and the variation among morphotypes was predominantly independent of mitochondrial genetic structure. Holobiont analyses revealed distinct differences between tissue-associated and seawater-derived microbial communities, identified multiple potential bacterial symbionts, and uncovered a highly specialized tissue-associated eukaryotic component predominantly comprised of the obligate parasite *Lankesteria*.

This thesis serves as the first comprehensive assessent of botryllid ascidian diversity across Turkish waters by synthesizing environmental, genetic, morphological, and microbial data, thereby providing essential baseline information for further phylogenetic, ecological, and taxonomic research. January 2026, 233 pages

Keywords:*Botryllus schlosseri*, population genetics, diversity, eDNA, Turkish coastlines.

3. TRANSCRIPTIONAL REGULATION OF THE TRANSCRIPTION FACTOR LHX3 IN THE NOTOCHORD: INSIGHTS FROM THE SIMPLE CHORDATE *CIONA*. Naya R. Haddad, MS thesis, Dept. of Biology, New York University, April 2026. Mentors: Dr. Lenny Joel Negrón-Piñeiro and Anna Di Gregorio. adg13@nyu.edu

During the early development of all chordates, including humans, the notochord provides critical structural support to the developing embryo and plays an essential role in patterning the neural tube, as well as in the development of the endoderm, paraxial mesoderm, and their associated structures.

Disruptions in notochord development can result in severe congenital malformations, while in adults, notochord-derived remnants are implicated in intervertebral disc disorders and chordoma, a rare but aggressive bone-derived tumor. Despite the importance of the notochord, the *cis*-regulatory mechanisms controlling notochord gene expression remain poorly understood. This study aims to shed light on the *cis*-regulatory strategies governing early notochord development by analyzing in detail the *cis*-regulatory module (CRM, or enhancer) associated with the evolutionarily conserved transcription factor (TF) Lhx3 in the simple chordate Ciona. Using a combination of *cis*-regulatory analysis, functional assays, and transcription factor binding site identification, we sought to uncover how the expression of Lhx3 is activated, and to position this TF within the gene regulatory network (GRN) that controls notochord morphogenesis in *Ciona*. The ultimate goal of this study is to advance our understanding of the regulatory strategies that ensure proper notochord gene expression across chordates, and aid both diagnosis and prevention of congenital malformations and tumorigenesis originating from notochord cells.

NEW PUBLICATIONS

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