Protist, Vol. 172, 125792, February 2021
http://www.elsevier.de/protis
Published online date 28 January 2021

Protist

FROM THE ARCHIVES

Protistology and Cell Biology at the Marine Arago Laboratory of Banyuls-sur-Mer (1961–2000): Personal Recollections

Marie-Odile Soyer-Gobillard

Sorbonne Université, Observatoire Océanologique-Laboratoire Arago de Banyuls-sur-Mer, 66650, France

Submitted January 2, 2021; Accepted January 20, 2021

Abstract

The history of protistology and the introduction of modern methods of unicell observations is described in a large maritime laboratory over a period of forty years by the initiator of this new team. The development of this team and the doctoral theses developed there are described as well as the major discoveries made. The Arago Laboratory, which was then in 1960 a field laboratory mainly devoted to the collection of biological material, becomes a research laboratory specializing in the study of the major fundamental problems which govern life: the organization and expression of the genome, mitotic processes and their nuclear and cytoplasmic components, cell cycle and its regulation as well as molecular phylogeny. The biological models chosen were essentially the dinoflagellate protists in their great variety: autotrophs, heterotrophs, myxotrophs and able of proliferating at sea, thus disrupting their cell cycle. Coupled with the techniques of biochemistry and molecular biology which it was in its infancy, the most advanced observation methods used electron and confocal microscopy often after use of ultra-cold cryopreparations, necessary to preserve the antigenic sites and allow the highlighting new proteins. The dinoflagellate model was then abandoned in favor of unicellular micro-eukaryotes allowing the development of environmental genomics.

Keywords: History of Protistology dinoflagellates; Banyuls marine station 1960–2000

Introduction

The Arago Laboratory was founded in 1881 by Henri de Lacaze-Duthiers (1821–1901), a French biologist, anatomist, and master of experimental zoology. The laboratory is located in Banyuls-sur-Mer (North Catalonia), on the Mediterranean coast. It is one of the three marine stations of the Sorbonne University (Paris), and it was also recognized as a National Observatory (OSU, Observatory of the Universe Sciences), then OOB (Observatoire Océanologique de Banyuls-sur-Mer) from January 1990.

When I arrived in Banyuls at the end of 1961, the Arago Laboratory was still a field marine laboratory, with a well-developed capacity for hosting visiting students and researchers. Permanent researchers were rare, as the Laboratory hosted a

https://doi.org/10.1016/j.protis.2021.125792

small handful of tight-knit scientists of diverse scientific backgrounds and interests. Its director at those times—from 1949 until 1964—Prof. Georges Petit (1892–1973), a brilliant zoologist and a cetacean specialist, was at the same time a confidant while striving to maintain his laboratory cohesiveness. His predecessor, as head of the Arago Laboratory, had been Prof. Edouard Chatton (1898–1947), a renowned protistologist and a corresponding member of the Academy of Sciences, and who had frequented this maritime station, in which he had made many discoveries since the early 1900s. As a pioneer of cell biology, a major discovery of Chatton was the formulation of the distinction between eukaryote Protists (with a nucleus limited by a membrane) and prokaryotes (without a nuclear membrane) (Chatton 1925; Soyer-Gobillard and Schrével 2020). The culminating point of his career was to be appointed Director of the Arago Laboratory, Professor at the Faculty of Sciences of Paris, Sorbonne University, succeeding in 1937 Prof. Octave Dubosq (1868–1943), a zoologist and protistologist himself, who directed this research center from 1923 to 1937. Unfortunately, when Georges Petit took over from Chatton after the latter's death in 1947, the Second World War (1939–1945) had resulted in damages to the Laboratory and its contents and so it was first necessary to rebuild and restructure what remained. In late 1961, as a young PhD student, just entering the CNRS (French National Center of Scientific Research) on a research training position, I was a pupil of the zoologist Prof. Pierre-Paul Grassé (1895–1985), Director of the Laboratory of Evolution of Organized Organisms in Paris (Faculty of Sciences, Sorbonne University), President of the French Academy of Sciences, and a recognized protistologist. Grassé had himself been a student of Octave Dubosq in Montpellier before the latter went to head the Arago Laboratory in 1927. To show his gratitude, in 1952, Pierre-Paul Grassé dedicated to him a new genus of protist, *Duboscquodinium,* a dinoflagellate.

So, I went to the Arago Laboratory to spend one month (in fact, I remained at the Arago Laboratory throughout my scientific career), the purpose of my stay was to collect marine planktonic protistological materials for my doctorate thesis (Soyer [-Gobillard] 1970) and to continue, using modern methods, the work elaborated by Chatton that had been carried out sixty years before on the free and parasitic dinoflagellates (Peridinians). Some of these protists have chloroplasts and are close to the plant kingdom, others do not, such as the *Noctiluca*, which is heterotrophic, bioluminescent, and carnivorous by eating its congeners and able to proliferate, provoking red tides in seawater. With my team, we used dinoflagellate protists as models to study the organization and expression of their genome. Also, their mitotic processes, nuclear and cytoplasmic components, cell cycle and its regulation as well as their molecular phylogeny were studied in my laboratory. At the same time, all the equipment necessary for such studies was installed, creating a new concept in a research center that was initially a field marine laboratory.

The Infancy of Cell Biology at the Arago Laboratory

When I arrived in Banyuls, only one protistologist, Dr. Jean Théodorides (a Parisian from the Grassé Laboratory), a specialist in Gregariniae (Sporozoa), was there for several months. It was he who taught me the first rudiments concerning planktonic marine protists, and my first scientific articles were dedicated to the description of several new species of Gregariniae, gastrointestinal parasites of pelagic copepods. But most of my scientific life was dedicated to dinoflagellate protists and particularly to their nuclear division and its major actors.

The Peridinians have a particular nuclear division, well described by Chatton in his doctorate thesis (Chatton 1920), but whose kinetics and components were poorly understood by then. I first used conventional cytology and cytochemistry methods, focusing on the best possible preservation of these particularly delicate cellular and nuclear structures. This preservation work extended to electron microscopy, observations being made in Paris, 900 km away from Banyuls-sur-Mer. Since the preparation of the observation grids took a considerable time, it seemed useful to be equipped in Banyuls with the basic equipment. A rudimentary but effective electron microscopy service was gradually put in place with the purchase of the first ultramicrotome. The knives intended to cut sections of biological material embedded in a resin were at that time made from glass bars and were subsequently latter replaced by diamond knives.

1967 was a decisive year because an international course in marine molecular biology was organized at the Arago Laboratory under the responsibility of a specialist in cell regeneration Prof. Marie Gontcharoff (Reims University), with the help of the renowned American cell biologist Prof. Daniel Mazia (1912–1996), a specialist of the mitotic apparatus (University of California, Berkeley) (Mazia and Gontcharoff 1964). The course participants were taught to use ultracen-

Figure 1. 1975. The first transmission electron microscope (Hitachi HU11A) in place at the Arago Laboratory (the first in a french marine station). Seating and working Marie-Odile Soyer-Gobillard. (©J. Lecomte, Bibliothèque du Laboratoire Arago/Sorbonne Université).

trifuges and scintillation counters to isolate, among other things, the sea urchin mitotic apparatus. Another important step, in 1975, was the "recovery" of a third-hand transmission electron microscope (TEM), Banyuls being the first French marine station to have such equipment at that time (Fig. 1). The TEM was first purchased by Prof. Pavans de Ceccatty (1927–2009), a famous histo-cytologist from the University of Lyon, it had been bought in second-hand by Prof. Combes from the neighbouring University of Perpignan, who, having been able to acquire new equipment, sold it to our Laboratory for 50,000 French francs, thanks to the support from the CNRS Life Sciences Department. The device, still of very good quality, performed the expected services and was replaced by a new TEM in 1982. Shortly after this, a first attempt was made to establish an external team in cellular and molecular biology, that of Dr Julio Pudles, a biochemist from the University of Orsay who decided to make long stays in Banyuls, Prof. André Berkaloff, then head of CNRS Life Sciences, financially supported this temporary establishment, our collaboration resulting in several publications (Coffe et al. 1982).

Thanks to my observations using TEM, I could deepen my knowledge on the division and condensation of chromatin in Blastodinides (parasitic, semi-heterotrophic dinoflagellates) and *Noctiluca* (free-living, heterotrophic bioluminescent dinoflagellate). I described, among others features in *Noctiluca*, its completely extraordinary nuclear membrane and its development during the morphogenesis of spores, the structure of its mouth, and its contractile tentacle. Also, for the first time, I described the first striated contractile myonemes of the animal kingdom (Soyer-Gobillard 1970), studied later by C. Métivier in her doctoral thesis on the *Noctiluca* motility, its structural organisation, ionic regulation, and cytosqueleton characterisation (Métivier and Soyer-Gobillard 1988). Jacques Soyer, then deputy director of the Arago Laboratory and I, defended our respective doctorate theses in Paris twenty-four hours apart on April 20 and 21, 1970. (Soyer and I were married from 1963 to 1983, this is why I signed my articles Marie-Odile Soyer and later Marie-Odile Soyer-Gobillard).

A Posthumous Article Signed with Chatton and Dedicated by André Lwoff (Fig. 2)

A few months later, in 1971, Prof. Pierre Drach (1906-1998), the director of the Arago Laboratory, an oceanographer and a crustacean specialist, arrived in my laboratory accompanied by Prof. André Lwoff (1902–1994), who had been awarded the 1965 Nobel Prize in Physiology or Medicine (Soyer-Gobillard and Schrével 2020). Lwoff himself was a former protistologist, pupil, and friend of Edouard Chatton (Soyer-Gobillard and Schrével 2003; Soyer-Gobillard 2019a, b). He had acquired the Mas Guillaume, a former fortress of Jacques 1er of Aragon (1208–1276), in Banyuls-sur-Mer as a holiday house. Lwoff asked me first to read my doctorate thesis and then to complete an unfinished manuscript of his master Chatton, of which he was the scientific heir. To Chatton's observations and marvellous drawings on the cycle of *Paradinium*, a parasitic plasmodial protist close to mycetozoans, I added my own observations along with the description of two new species (Chatton and Soyer [-Gobillard] 1973). Thus in 1973, twenty-six years after Chatton's death, I co-authored a posthumous article with him prefaced by André Lwoff (Fig. 2). It was the start of a long and fruitful collaboration that resulted in putting Chatton back in his rightful place in international protistology, alongside Lwoff (Soyer-Gobillard and Schrével 2020). As said by André Lwoff at the end of his Preface, «Edouard Chatton left a considerable number of

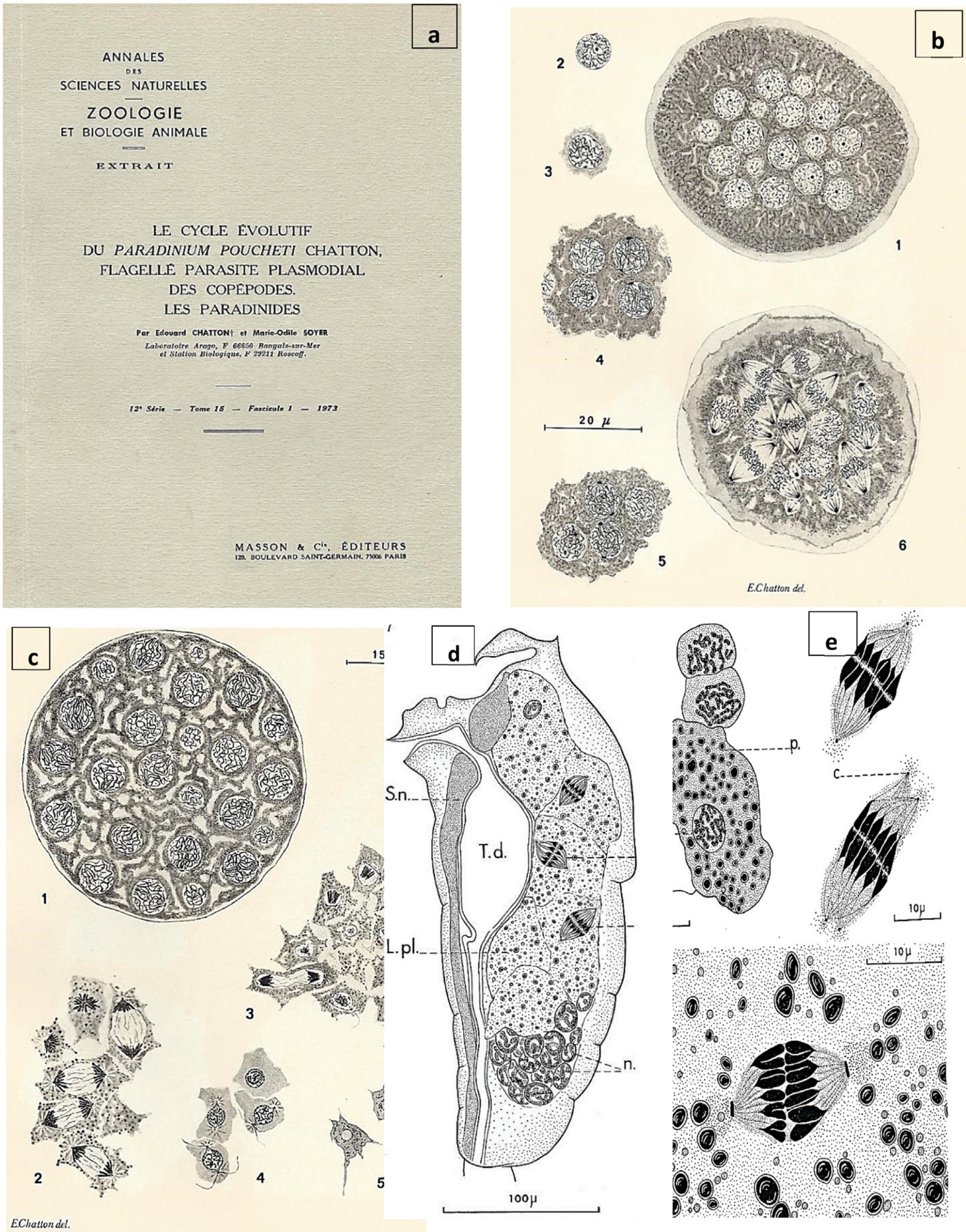


Figure 2. The genus *Paradinium* Chatton. In a posthumous publication (Fig. 2, a) the biological cycle of *Paradinium poucheti,* a protist close to Mycetozoa is described by Chatton: it multiplies in the form of a plasmodium in the general cavity of planktonic copepods before being ejected in the form of sporospheres (cysts) in which sporulation takes place (Fig. 2, b, c). In thin sections of the new species *Paradinium caulleryi* (Fig. 2, d), we can observe the large volume occupied by the plasmodium and the very large size of the nuclei (16 μm). In the second new species *Paradinium mesnilii* (Fig. 2, e), the nuclei are even larger (24 μm) with n = 6 chromosomes. Fig. 2 b, c: E.Chatton del., Fig. 2 d, e: M.O.Soyer- (Gobillard) del. ©Personal collection M.O.Soyer-Gobillard.

documents and materials untapped, but his name is unlikely to appear again as author of a publication. It is with great emotion that I present today the last memoir of the great protistologist, of whom I am honored to have been a pupil and a disciple».

In 1968, with Yves Bouligand (1935-2011), a specialist in cholesteric structures (or liquid crystals), I had begun to work on the structure of dinoflagellate chromosomes, which, in ultra-thin sections, have a unique arch-shaped appearance. Bouligand explained their twisted nature by a physical theory, likening them to so-called cholesteric liquid crystals (Bouligand et al. 1968). Several articles I published next from my thesis in *Chromosoma* (Berlin) attracted the attention of a young Finnish cytogeneticist, Olli Haapala, who asked to come and work with me at Banyuls-sur-Mer as part of his thesis on the ultrastructure of these dinoflagellate chromosomes. Hard work and several stays at the electron microscopy center on boulevard Raspail in Paris allowed Haapala and me to be the first to spread these chromosomes on water, to collect them on special grids for transmission electron microscopy, observe them and publish their coiled fibrillar organization in *Nature* in August 1973 (Haapala and Soyer [-Gobillard] 1973). This publication constituted an “accelerator enzyme” for the continuation of my work; shortly after, in 1974, I received the Paul Wintrebert Foundation Prize for these works. Despite the fruitful collaboration, I gave up working with Bouligand, preferring to devote myself to more functional research at the level of chromosomes, mitotic apparatus, nucleus, and cytoskeleton. In November 1974, Haapala defended his thesis at the University of Turku (Finland), which I attended. I took advantage of Russia’s proximity to give several seminars at the Institute of Cytology at the Leningrad Academy of Sciences in Saint Petersburg (Fig. 3) at the invitation of Prof. Georges Poljansky (1904–1993) and Igor Raikov (Raikov 1982), famous Russian protozoologists and specialists of ciliates.

Figure 3. Leningrad (Saint Petersbourg) 1974. Marie-Odile together with Professor Georges Poljanski, a famous Protozoologist of the Institute of Cytology of Academia of Sciences of Saint Petersburg (Leningrad), a specialist of Ciliate cytology and taxonomy. The Neva River and the Hermitage museum are in the background. ©Personal collection M.O.Soyer-Gobillard.

Expansion of the Team

In 1975, the cell biology team expanded with the recruitment of Michel Herzog, a PhD student, researcher at the CNRS; Françoise de Billy, a CNRS engineer; Paul Prévot, a PhD student from the DEA (Diplôme d’Études Approfondies) in Biological Oceanography; and later Yvonne Bhaud, a researcher at the CNRS (Bhaud et al. 1991). The team also benefited from the support of a technician from Paris 6 (Sorbonne) University, Marie Albert, who had already helped me during my thesis. In 1977, my team did not have any modern equipment, except the ultracentrifuge and the old transmission electron microscope. At the beginning of his thesis, Michel Herzog was helped with kindness by the advice of Prof. Paul Penon, a specialist on plant RNA polymerases, head of the LGDP (Laboratory of Genome and Plant Development) at the neighboring Perpignan University via Domitia (UPVD). With Herzog, we deepened the study of the organization and composition of dinoflagellate DNA (Herzog and Soyer 1983) which is generally always condensed, and he defended his thesis in Banyuls-sur-Mer in June 1983. An anecdote deserves to be told: The jury for Michel Herzog’s thesis included Prof. Guy Echalier, professor at Paris 6 University, specialist in the development of *Drosophila* cell lines established from embryos, and known for his “good words”. During his argumentation, he dared to say: “To obtain all these results, your Research Director, Marie-Odile, probably had to practice dinoflagellation!”

Michel Herzog then innovated by being the first to have sequenced the ribosomal genes of certain dinoflagellates with Luc Maroteaux (Herzog and Maroteaux 1986) while directing the thesis of Guy Lenaers, a pioneering work on the molecular phylogeny of dinoflagellates, at the time unknown (Lenaers et al. 1989) and of Montse Sala-Rovira from Barcelona, on the caracterization and cloning of non-histones nuclear basic proteins in the heterotrophic dinoflagellate *Crypthecodinium cohnii* Ehr (Sala-Rovira et al. 1991).

Herzog left the team when he was appointed Research Director at CNRS in 1990 for a professorship at the University of Grenoble, attracted by the *Arabidopsis* plant model. At the same time, my team developed another research component devoted to intracellular ecotoxicology: The impact of pollutants, such as heavy metals, organochlorine or organophosphorus pesticides, on our models of marine dinoflagellate protists (Prevot et al. 1993). After defending a thesis on these pioneering subjects, Paul Prévot, became a CNRS Research engineer, this research being supported by CNEXO (National Center of Exploration of the Oceans) and the French Ministry of Defense, extremely interested in the intracellular effect of defoliants (Agent Orange for example, of sinister memory for its use by the U.S. Armed Forces during the Vietnam War). These contracts made it possible, at a time when institutional credits had sharply decreased, to finance more basic research or to purchase equipment. The team's work was supported by engineers and technicians (P. Prévot, M. Albert, F. de Billy, M.-L. Géraud, D. Saint Hilaire, and later, a good photographer, J. Lecomte, and a designer, M.-J. Bodiou).

An Unforgettable International Meeting in Banyuls: the Vth Meeting of ISEP (International Society for Evolutive Protistology)

The reputation of our team repeatedly allowed us to obtain the funds to organize several national, European or world congresses in the fields of protistology and cell biology. With Julio Pudles (University of Orsay) and Denise Paulin (Institut Pasteur), the French Cytoskeleton Club was created in 1982 which allowed researchers working in this field to meet, sometimes in Banyuls. The 5th Congress of the International Society for Evolutionary Protistology (ISEP) was held in 1983 (Margulis et al. 1984), with the active participation of Prof. André Lwoff and Prof. Alvin Pappenheimer (Harvard University). Pappenheimer was one of the most important biochemists and immunologists of that time, a specialist in growth factors in microorganisms. Also present was Prof. Lynn Margulis, founder of this International Society, a formulator of the symbiotic theory of the evolution of the first eukaryotic cell (Margulis et al. 2006). She was distinguished as the recipient of the American National Medal of Science, the highest scientific distinction in the United States, awarded by President Clinton in 1999. To resume this unforgettable meeting, what could be more precise than reproducing parts of the excellent foreword written by Lynn as an introduction to the book "Evolutionary Protistology: The Organism as Cell" (D.Reidel Publishing Company) reprinted from the proceedings of the meeting published in Origins of Life, 1983, **13** (3-4), pp 169-552. This foreword summarizes over several pages all the advances concerning the biology of Protists and their evolution at that time.

FOREWORD

Lynn Margulis † (1938-2011)

For the first time since its inception, at Boston University in June 1975[1], the Society for Evolutionary Protistology met in Europe. Under the direction of Marie-Odile Soyer-Gobillard and hosting some 70 people representing a dozen nations (Belgium, Canada, Denmark, England, France, W. Germany, The Netherlands, Poland, Scotland, Spain, Switzerland, U.S.A.) the meeting was held at Banyuls-sur-Mer in Catalunya. The 1983 ISEP met at the famed Laboratoire Arago on the Mediterranean Sea, most participants were housed in the Laboratory's newly refurbished Grand Hotel.

The previous meetings had emphasized single themes, e.g., (First) Boston, 1975 Evolution of Mitosis in Eukaryotic Microorganisms; (Second) Downsview Ontario, 1977 Criteria for Phylogeny in Protists. In spite of the fact that the third meeting, planned for Leeds, England in June of 1979, was never held some of the papers scheduled to be presented there were published in *BioSystems*, Volume 12, Numbers 1 and 2. The fourth meeting at Port Deposit, Maryland, 1981 called Conference on Cellular Evolution focused on the Evolution of Microtubules, Mitosis, Microfilaments and other Fibrillar Systems. The proceedings of this meeting were published in *BioSystems*, Volume 14, Numbers 3 and 4. This fifth meeting was planned around multiple themes: Experimental methods in studying evolution, uniformity and diversity in protistan structure, relationships between protistan phyla, relationship between nucleoid and cytoplasm in archaebacteria and nucleus and cytoplasm in eukaryotic cells, dinoflagellate chromosome organization and the origin of muticellularity. The papers from this 5th meeting are here (*Origins of Life* vol. **13**, p. 169-352 as the journal and the book) with the exception of contributions by Li-Jing Yang, D. Sigee, J. Dodge, P. Rizzo and Morris that deal with dinoflagellates. Those four promptly submitted papers appeared in *BioSystems* vol. **17**, 1984. The invited speaker at the meeting, Professor Guy Ourisson of the University of Strasbourg, introduced the protistologists to the power of organic geochemistry. He discussed studies of secondary metabolism in aiding the interpretation of phylogenies as well as the use of organic geochemical analysis in the interpretation of the fossil history of photosynthetic microbes and plants. Nobel Laureate, Andre Lwoff whose book of ciliate morphogenesis[2] and techniques of ciliate cortical staining (Chatton-Lwoff technique) has provided protistological inspiration since the 1940's, was in attendance and introduced Professor Ourisson.

As emphasized by John Corliss of the University of Maryland, the protists (*sensu lato*, by which he means the *protoctists*, eukaryotic organisms exclusive of members of the Kingdoms Animalia, Plantae and Fungi) comprise a far larger and diverse group of organisms than most realize. Corliss estimates that there are more than 110,000 species of protists comprising perhaps 40 major lineages or phyla. These organisms include the 'water molds' or so-called 'motile fungi' such as *Saprolegnia* and other oomycotes that are serving as excellent material to provide the basis for understanding of mitotic movement and sexuality. This was amply demonstrated by Professor I. B. Heath and his group (F. Murrin and L. MacKerracher). A general theory of the evolution of mitotic movements was presented by U.-P. Roos from Zürich.

The polyphyly of multicellularity was demonstrated by the work of Isabelle Desportes (Paris), in work on the bizarre life cycle of the *Paramarteilia* (myxosporidians, parasite of Polychaetes) in which cells develop inside other cells of the same organism.

The use of microtubular ultrastructural patterns to assess relatedness has become apparent to everyone. Both the taxonomy of heliozoans (Colette and Jean Febvre, Villefranche-sur-Mer) and the taxonomy of ciliates (Eugene Small, College Park, Maryland) are being extensively revised. The concept of *kinetid* (*cinetid*), the unit pattern of cell cortex which is comprised of the basal structure of microtubules and microfibrils surrounding the kinetosome, is becoming crucial in the explication of the phylogeny of members of the protoctists, independent of the presence of plastids. The importance of the heterotrophic portion of the cell was elegantly pointed out by P. Kivic and P. Walne (Tennessee) in a paper that showed members of the Euglenids and Kinetoplastids (the group to which *Trypanosoma* belongs) to be far more related than euglenoids and, for example, the chlorophyte green algae.

The importance of protists in the elucidation of fundamental cell problems was demonstrated by several speakers. The presence of striated fibers involved in cell calcium regulation and movement was shown by M. Melkonian (Münster) in his work on the prasinophyte *Tetraselmis* (= *Platymonas*).

Biomineralization, for example, the intracellular production of calcium carbonate tests, is optimally studied in the haptophytes (coccolithophorids), as shown by P. Westbroek from Leiden. A fascinating hypothesis that relates light perception and directed behavior in dinoflagellates was presented by J. Dodge (Surrey) in a paper that involves a strand of microtubules (originating at the base of the longitudinal undulipodium). This strand of microtubules passes over the eyespot (in two rather different species of dinoflagellates each with different eyespot organization). Dodge suspects that the microtubules have a direct role in the transmission of directional stimuli that bring about the phototropic response.

The uniqueness of the genetic organization of dinoflagellates was emphasized by several investigators (M. Herzog, M. O. Soyer-Gobillard, Banyuls-sur-Mer; Peter Rizzo, College Station; David Sigee, Manchester; and C. Galleron of Paris). Apparently the high quantity of hydroxymethyl uracil which replaces so much thymine in dinoflagellates appears in the DNA by means of a post-replicative mechanism. The peculiar characteristics of the dinoflagellates' nuclei strongly suggest that this group is monophyletic and has evolved independently of the other eukaryotes.

The tubulin proteins, especially beta-tubulin, comprising undulipodia are remarkably conserved in the great range of eukaryotes studied. On the other hand M. Little (Heidelberg), R. Ludueña (San Antonio) and their colleagues have shown that variations in alpha-tubulin provide fine tools for reconstructing the phylogeny of eukaryotic microbes and their relationships to animals and plants. Nonanimal alpha-tubulins, as determined by peptide digest studies, of the cytoplasm of a plant (rose) are nearly identical to the alpha-tubulins of the green algae and ciliates tested, and are very similar to cytoplasmic alpha-tubulins of the plasmodial slime mold *Physarum* and the heliozoan *Echinosphaerium*. These are in marked contrast to animal alpha-tubulins which closely resemble each other. These investigators including Andre Adoutte (Gif-sur-Yvette) hope to use tubulin sequence data and immunocytochemistry to solve the thorny problem of which protists were ancestral to animals and plants. Another approach to this classical problem came from C. Bardele (Tübingen) who showed that the details of the undulipodial necklace (membrane patterns, as revealed by freeze etching on the inside of cilia and sperm tails) show a close relatedness in all animals studied but are far more varying in protists. Perhaps by finding the protist pattern most like that of the metazoa, the extant lineage most closely related to the ancestral animals will eventually be identified.

Many ISEP members who participated in this meeting are also contributing to the *Handbook of Protoctists*. This handbook, edited by Lynn Margulis, John Corliss and David Chapman, is scheduled to be published in early 1985 by Jones and Bartlett Publishers. It will be one volume with chapters on each phylum in Kingdom Protoctista. P. Westbroek, P. Walne and P. Kivic, E. Cox, M. Melkonian, E. Small and D. Lynn, and D. Barr are some of these authors.

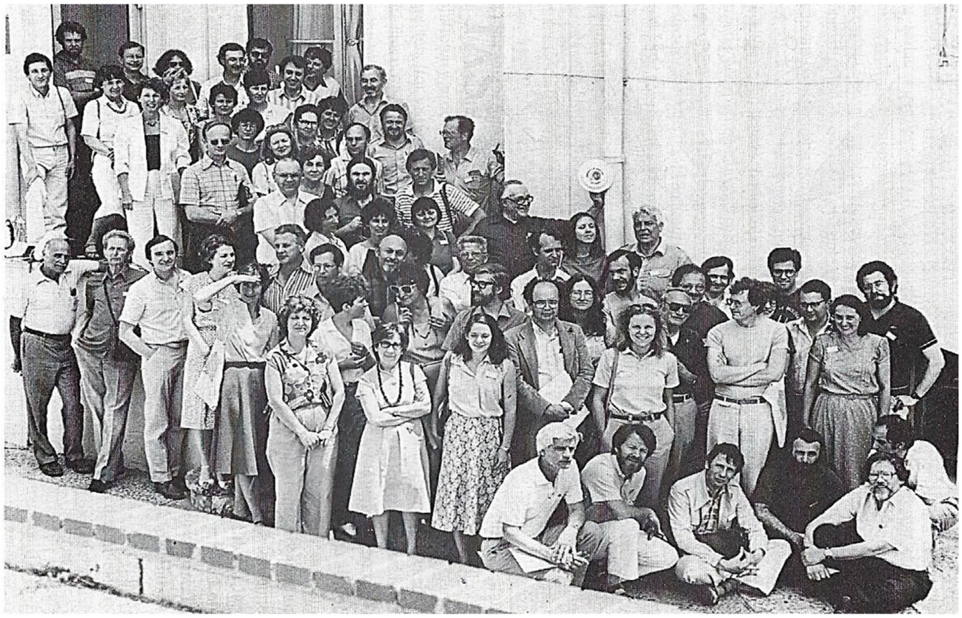
Due to the hard work of I. Brent Heath, the fledgling ISEP has achieved legal status as an international nonprofit scientific organization, registered in Canada. According to the by-laws a regular member of ISEP "shall be persons having an interest in the origin, evolution and phylogeny of eukaryotic organisms who have made an application to and have been accepted by the Secretary". The presidency of the Society has now passed from Christian Bardele to Professor Heath. The Secretary is Dr. Diana Lipscomb. It was decided after much discussion that the next biennial meeting will be held again in North America, at Ottawa June 10-14 1985, under the direction of Dr. Donald Barr. At that time Dr. Dennis Goode (University of Maryland), who was elected President Elect of ISEP at Banyuls, will begin his presidency.

The Banyuls meeting was beautifully organized, aid was forthcoming from several sources. Sources of money were: Centre National de la Recherche Scientifique (Secteur sciences de la vie), C.N.R.S. – PIRO (Programme interdisciplinaire pour la recherche oceanographique), Université Pierre et Marie Curie, Paris VI, Association Naturalia et Biologia (Paris). The food was remarkable and many participants greatly enjoyed their visit to Tautauvel, the small Catalunyan town. It was here, in the cave of Arago, that a fine fossil skull and other bones and teeth of *Homo erectus*, about 400,000 years old, have been found. These represent the oldest evidence for early man in Europe demonstrating the extreme desirability as living space of this magnificent, ancient part of the world.

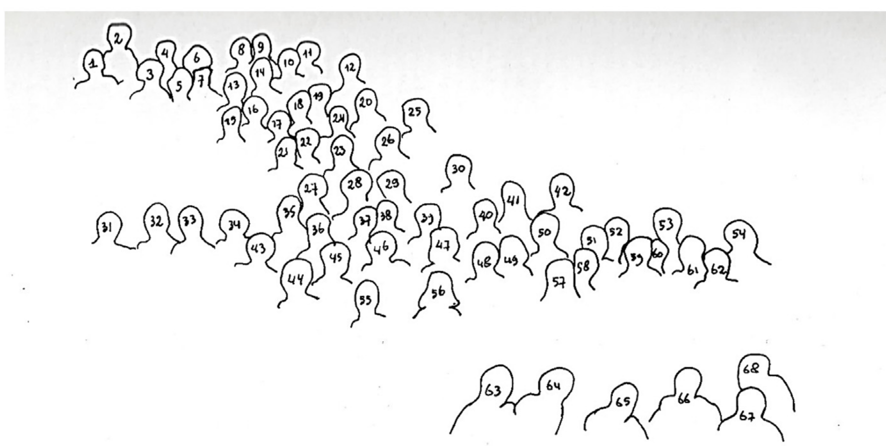
I thank I. B. Heath, B. Dexter Dyer, Donna Mehos, and Marie-Odile Soyer-Gobillard for aid in preparing this report.

As communicated recently by Michael Melkonian, present at this meeting: «Yes, of course, I remember the ISEP meeting in Banyuls in 1983 very well, it was a "magical" meeting and I was a young, ascending scientist then. The weather was fine, the science (and the wine) excellent and bioluminescence in the harbor conspicuous. Tom Cavalier-Smith went bird watching around noon in the summer heat and a group photograph was taken in front of a ship (I saw the photograph recently in the Internet). I remember well my encounter with André Lwoff at the ISEP meeting in Banyuls. In some ways our families had a similar history because both originally emigrated from Imperial Russia » (Fig. 4).

I am honored that all of my work on the structure and functioning of dinoflagellate chromosomes, their maintenance by divalent cations and structural RNAs, earned me the 1988 Trégouboff Prize from the French Académie des Sciences. In 1989, Éric Perret, a new PhD student arrived, supported by a scholarship from the Montpellier Biology-Health doctoral School. His thesis was devoted to the study of cell division in the dinoflagellate *Crypthecodinium cohnii.* A harvest of results followed from this work, in particular, the discovery of several proteins playing a major role in mitotic mechanics, some of which having been preserved from dinoflagellates to humans (Perret et al. 1993, 1995). A little bit before, I managed the demonstration of the co-localization of two DNAs (B- and Z-DNA) by electro-immunolocalization allowing to explain the functioning of these compacted chromosomes (Soyer-Gobillard et al. 1990). From 1985 to 1990, this department succeeded in acquiring the equipment necessary to develop in particular electro-immunocytochemistry techniques. Thanks to the technique of vitrification of biological material at ultra-low temperatures (at the temperature of liquid helium, i.e. $-269\,^{\circ}C$), the team could precisely localize proteins for which the genes had been isolated and the antibodies produced. Thanks to this cryopreservation technique, (cryo-electron microscopy was developed by the Swiss Prof. Jacques Dubochet and earned him the 2017 Nobel Prize in Physics) implemented for the first time in a marine station, the antigenic sites were ideally preserved. A confocal microscope completed this equipment, and a third-generation transmission electron microscope was also acquired, the second having been sold to the Corsican University of Corte. In 1991, Michèle Barbier, a new PhD student arrived, whose work, supported by an IFREMER (French Research Institute for Sea Exploitation) grant, was co-directed by André Picard, great specialist of the molecular regulation of the cell cycle and me: this was to study the specific molecules regulating the cell cycle in dinoflagellate unicellular eukaryotes. This work led to the defense of her thesis in 1996 in which a homolog of the MPF factor (M-Phase Promoting Factor) was demonstrated in a dinoflagellate as well as a homolog of cyclin B, and their controlling role during the cell cycle (Barbier et al. 1995). With Michèle Barbier and Muriel Audit, we also made an incursion into the yeast *Saccharomyces pombe*, demonstrating the presence and immunolocalizing the unique cyclin B (p56cdc13) that controls the course of its life cycle (Audit et al. 1996). Several years earlier, Catherine Rausch de Traubenberg had started, also in my laboratory, a study on the specific populations of symbiotic bacteria associated with dinoflagellates. Thanks to an IFREMER grant, she was able to continue this work at IFREMER Nantes and defended a distinguished thesis about the interactions between the toxic dinoflagellate *Prorocentrum lima* Ehr. and its bacterial microbiota. Her thesis was co-supervised by Dr Patrick Lassus (IFREMER Nantes) and myself (Rausch de Traubenberg et al. 1995a,b). Meanwhile, Jérôme Ausseil, who arrived a little later and was supported by a grant



a



b

1 Michel Herzog	12 John Dodge	23 David Patterson	34 Michelle Laval-Peuto	45 M.C. Tellez	57 Susan Lenk
2 Paul Prévot	13 Christine Métivier	24 Claude Grevet	35 Jean Genermont	46 Monique Cachon	58 Morris Alexander
3 Colette Galeron	14 Marie Albert	25 Peter Westbroek	36 Denis Searcy	47 Tom Cavalier-Smith	59 Melvyn Little
4 F.J.R. (Max) Taylor	15 Guy Brugerolle	26 Urs Peter Roos	37 Guy Ourisson	48 Richard Luduena	60 Diana Lipscomb
5 Marie-Odile Soyer	16 Pilar Garcia	27 Colette Demar	38 Josette Rouvière-Yaniv	49 Faye Murrin	61 David Lloyd
6 Annie Boillot	17 Isabelle Desportes	28 Veronique Machelon	39 Jean Cachon	50 Gunther Bahnweg	62 Lynn Margulis
7 Yvonne Bhaud	18 M. Dynner	29 Patricia Walne	40 Brent Heath	51 Michael Melkonian	63 Jerome Motta
8 Peter Rizzo	19 Martha Estrada	30 John Corliss	41 Betsy Dexter-Dyer	52 S.T. Moss	64 Dennis Goode
9 Peter Kivic	20 Øjvind. Moestrup	31 Alvin Pappenheimer	42 Robert Allen	53 John Heisler	65 Vincent Demoulin
10 David Sigee	21 Jean-Pierre Mignot	32 André Lwoff	43 Gisele Bardele	54 André Adoutte	66 Eugene Small
11 Anton Bopman	22 Marcelle Lefort-Tran	33 Alain Bilbaud	44 Mrs Motta	55 Lisa McKerracher	67 Jean Febvre

Figure 4. Banyuls-sur-Mer 1983. Participants of the Vth ISEP (International Society of Evolutionary Protistology) meeting. Among the personalities, to be remarked : André Lwoff and Alvin Pappenheimer (32, 31), Lynn Margulis and André Adoutte (62, 54), Marie-Odile Soyer-(Gobillard) and Max Taylor (5, 4), Michael Melkonian (51).(©J. Lecomte, Bibliothèque du Laboratoire Arago/Sorbonne Université), © Origins of Life and Evolution of Biospheres, 13, 1984, P.IV, D.Reidel Publishing Company, The Netherlands.

Table 1. Doctorate Theses carried out into the team « Genome and cell cycle of Unicell Eucaryotes » Laboratory Arago-Banyuls-sur-Mer from 1970 through 2000.

Names	Universities	Dates	Titles
M.O. SOYER-(GOBILLARD) (CNRS)	Paris 6	1970	Cytology and division in two dinoflagellates: *Noctiluca* Suriray, freeliving genus and *Blastodinium*Chatton, parasitic genus.
O. K. HAAPALA (Finlandia)	Turku	1974	Dinoflagellate chromosome structure.
M. HERZOG (CNRS)	Paris 6	1983	Chromosomes, chromatin and DNA of Dinoflagellates.
P. PREVOT (Grants CNEXO, IFREMER)	Paris 6	1985	Marine Dinoflagellates *P. micans* and *C. cohnii*, target organisms of toxic pollutant impact study (cadmium, selenium, parathion, malathion).
Ch. METIVIER (Scholarship Vocation)	Paris 6	1986	Motivity in the evoluted Dinoflagellate *Noctiluca miliaris S*. Structural organisation, ionic regulation, cytosqueleton characterisation.
G. LENAERS (Scholarship Doctoral Formation Biology-Health)	Montpellier	1990	Structure and evolution of ribosomal 24-26S RNA of protists. Application to the Dinoflagellate phylogeny.
M. SALA ROVIRA	Barcelona-Paris 6	1991	Caracterization and cloning of non-histones nuclear basic proteins in the Dinoflagellate *Crypthecodinium cohnii* Ehr.
C. RAUSCH DE TRAUBENBERG	Nantes, IFREMER	1993	Interaction between a Dinoflagellate and its bacterial associated microflore : role of bacteria in toxicity of *Prorocentrum lima* Ehr.
E. PERRET (Scholarship Doctoral Formation Biology-Health)	Montpellier	1993	Study of cell division in a primitive eukaryote *Crypthecodinium cohnii*: microtubular dynamics, identification of antigens immunologically related with human centrosome.
M. BARBIER (Scholarship IFREMER)	Paris 6	1996	Régulation of cell cycle in unicell Eucaryote Dinoflagellates.
J. AUSSEIL (Scholarship Ligue against cancer 66)	Paris 6	1999	Proteins of cell division in Dinoflagellates : Identification of nuclear and cytoplasmic motive Proteins
A. NAVARRETE AGUILERA	Barcelone	1999	Caracterizacion ecofisiologioca y bioquimica de los tapetes microbioanos del delta del Ebro.

from the League against Cancer 66, was working on the identification of cell division proteins in dinoflagellates, in particular, nuclear motor proteins and cytoplasmic proteins. Not only did he isolate them, made antibodies and immunolocalized them on an ultrastructural scale, but he also sought for the interrelationships of these motor proteins, such as actin and a new P80 protein, specific for dinoflagellates, two essential partners. His work culminated in a thesis in June 1999 and numerous publications (Ausseil et al.1999) and Table 1.

In 1996, Hervé Moreau (1958–2020), a cellular and molecular biologist, joined my team "Genome and Cell Cycle of Unicellar Eukayotes" with a doctoral student, Delphine Guillebaud and a CNRS engineer Evelyne Derelle. In 2000, he was attracted by another single-celled model from the chlorophytes (Prasinophyceae), *Ostreococcus tauri* Courties and Chrétiennot-Dinet 1995, the smallest known chlorophyllous eukaryote protist, whose genome was soon sequenced (Derelle et al. 2006), paving the way for a new science, environmental genomics.

Despite the end of the use of the interesting model of protist dinoflagellates at the Arago Laboratory, contemporary with my retirement in 2000,

Figure 5. Banyuls-sur-Mer 1990. The team of Dr Marie-Odile Soyer-Gobillard, Director, CNRS researcher (seated). Next from left to right, P. Prévot, CNRS engeener, Ch. Métivier and E. Perret, PhD. students from Paris 6 and Montpellier Universities, Dr Y. Bhaud, CNRS researcher, M.L. Géraud-Escande, CNRS engeener, M. Sala-Rovira, PhD. student from Barcelona University, M. Albert†, Technician, Paris6 University, G. Lenaers, PhD. Student from Montpellier University, Dr M. Herzog, CNRS researcher. (©J. Lecomte, Bibliothèque du Laboratoire Arago/Sorbonne Université).

these protists continue to be studied in many laboratories around the world, in terms of the fundamental themes that had been developed during these decades in my team: organization and expression of the genome, mitotic processes and their nuclear and cytoplasmic components, cell cycle and its regulation, and molecular phylogeny. I continue to defend this marvelous and original model (Soyer-Gobillard and Dolan 2015; Soyer-Gobillard 2019a,b) (Fig. 5).

Conflicts of interest

I have no conflicts of interest to declare.

Acknowledgements

This article is an homage to Prof. Jacques Soyer (1938-2019), Head of the Arago Laboratory from 1976 to1989, for his permanent support in the introduction of the new concepts of cell biology for a Laboratory of Marine Biology and Oceanography and for the creation of the infrastructures which resulted from it. The author thanks Prof. Michael Melkonian (Max Planck Institute for Plant Breeding Research, Cologne, Germany), Dr Michael Dolan (University of Massachusetts-Amherst, Sona Dolan Memorial Microscopical Observatory of Belchertown, MA, USA), Prof. Ricardo Guerrero (University of Barcelona, Barcelona, Spain), and Mrs. Mercè Piqueras (Science Writer, Barcelona, Spain) for critical proofreading of the manuscript. The author also acknowledges Dr Guy Jacques (CNRS, Sorbonne University) which was first at the origin of the recollections expressed here. I would like to thank my supervisory body, the CNRS (French National Center of Scientific Research) for its unfailing support throughout my scientific career as well as Sorbonne University where my Laboratory was housed. Without these organizations, our research would not have been possible.

References

Audit M, Barbier M, Soyer-Gobillard MO, Albert M, Geraud ML, Nicolas G, Lenaers G (1996) Cyclin B (p56cdc13) localization in the yeast *Schizosaccharomyces pombe:* An ultrastructural and immunocytochemical study. Biol Cell **86**:1–10

Ausseil J, Soyer-Gobillard MO, Geraud ML, Baines I, Preston T, Bhaud Y, Moreau H (1999) Characterization of p80, a novel nuclear and cytoplasmic protein in dinoflagellates. Protist **150**:197–211

Barbier M, Albert M, Geraud ML, Bhaud Y, Picard A, Soyer-Gobillard MO (1995) Cell cycle regulation of the primitive dinoflagellate *Crypthecodinium cohnii* B. : evidence for the presence of a homologue of cyclin B. Biol Cell **84**:35–42

Bhaud Y, Salmon JM, Soyer-Gobillard MO (1991) The complex cell cycle of the dinoflagellate protoctist *Crypthecodinium cohnii* B. as studied *in vivo* and by cytofluorimetry. J Cell Sci **100**:675–682

Bouligand Y, Soyer MO, Puiseux-Dao S (1968) La structure fibrillaire et l'orientation des chromosomes chez les Dinoflagellés. Chromosoma (Berlin) **24**:251–287

Chatton E (1920) Les Péridiniens parasites. Morphologie, reproduction, éthologie. Arch Zool exp et gén **59**:1–475

Chatton E (1925) *Pansporella perplexa*, Amoebien à spores protégées parasites de Daphnies. Réflexions sur la biologie et la phylogénie des Protozoaires. Ann Sci Nat **8**:5–84

Chatton E, Soyer-Gobillard MO (1973) Le cycle évolutif du *Paradinium poucheti* Chatton, flagellé parasite plasmodial des Copépodes. Les Paradinides. Ann Zool (Ann Sci nat) Paris **15**:27–60

Coffe G, Rolla FH, Soyer MO, Pudles J (1982) Parthenogenetic activation of sea-urchin egg induces a cyclical variation of the cytoplasmic resistance to hexylene glycol-triton X-100 treatment. Exp Cell Res **137**:63–72

Derelle E, Ferraz C, Rombauts S, Rouze P, Worden AZ, Partensky F, Robbens S, Degroeve S, Echeynie S, Cooke R, Saeys Y, Wuyts J, Jabbari K, Bowler C, Ball S, Ral JP, Bouget F, Piganeau G, de Baets B, Picard A, Delseny M, Demaille JJ, Van de Peer Y, Moreau H (2006) Genome analysis of the smallest free-living eukaryote *Ostreococcus tauri* unveils unique genome heterogeneity. Proc Natl Acad Sci USA **103**:11647–11652

Haapala OK, Soyer-Gobillard MO (1973) Structure of dinoflagellate chromosomes. Nature (New Biol London) **244**:195–197

Herzog M, Maroteaux L (1986) Dinoflagellate 17S ribosomal sequence inferred from the gene sequence: Evolutionary implications. Proc Natl Acad Sci USA **83**:8644–8648

Herzog M, Soyer-Gobillard MO (1983) The native structure of dinoflagellate chromosomes and their stabilization by Ca^{2+} and Mg^{2+} cations. Eur J Cell Biol **30**:33–41

Lenaers G, Maroteaux L, Michot B, Herzog M (1989) Dinoflagellates in evolution. A molecular phylogeny analysis of large-subunit ribosomal RNA. J Mol Evol **29**:40–51

Margulis L, Soyer-Gobillard MO, Corliss J (eds) Evolutionary Protistology. The Organism as Cell. D. Reidel, Dordrecht, Holland, 352 p.

Margulis L, Chapman M, Guerrero R, Hall J (2006) The last eukaryotic common ancestor (leca): acquisition of cytoskeletal motility from aerotolerant spirochetes in the proterozoic eon. Proc Natl Acad Sci USA **103**:13080–13085

Mazia D, Gontcharoff M (1964) The mitotic behavior of chromosomes in Echinoderm eggs following incorporation of bromodeoxyuridine. Exp Cell Res **35**:14–25

Métivier C, Soyer-Gobillard MO (1988) Organization of cytoskeleton during the tentacle contraction and cytostome movement in the dinoflagellate *Noctiluca scintillans* Mc Cartney. Cell Tissue Res **251**:359–370

Perret E, Davoust J, Albert M, Geraud ML, Soyer-Gobillard MO (1993) Microtubule organization during the cell cycle of a primitive eukaryote dinoflagellate. J Cell Sci **104**:639–651

Perret E, Moudjou M, Geraud ML, Derancourt J, Soyer-Gobillard MO, Bornens M (1995) Identification of an HSP70-related protein associated with the centrosome from dinoflagellates to human cells. J Cell Sci **108**:711–725

Prevot P, Perret E, Jupin H, Soyer-Gobillard MO (1993) Use of fluorescence induction measurement for parathion toxicity studies in a marine dinoflagellate *Prorocentrum micans* Ehr: a possible model for biodetection. Ecotoxicol Environ Safety (USA) **25**:360–371

Raikov IB (1982) The Protozoan Nucleus. Morphology and Evolution. In Alfert M, Beermann W, Goldstein L, Porter KR, Sitte P (eds) Cell Biology Monographs **vol. 9**. Springer-Verlag, Wien – New York, 474 p

Rausch de Traubenberg C, Geraud ML, Soyer-Gobillard MO, Emdadi D (1995a) The toxic dinoflagellate Prorocentrum lima and its associated bacteria. I. An ultrastructural study. Europ J Protistol **31**:318–326

Rausch de Traubenberg C, Soyer-Gobillard MO, Geraud ML, Albert M (1995b) The toxic dinoflagellate *Prorocentrum lima* and its associated bacteria. II. Immunolocalization of okadaic acid in axenic and non axenic cultures. Europ J Protistol **31**:383–388

Sala-Rovira M, Geraud ML, Caput D, Jacques F, Soyer-Gobillard MO, Vernet G, Herzog M (1991) Molecular cloning and immunolocalization of two variants of the major basic nuclear protein (hcc) from the histone-less eukaryote *Crypthecodinium cohnii* (pyrrhophyta). Chromosoma (Berlin) **100**:510–518

Soyer [-Gobillard] MO (1970) La cytologie et la division chez deux types de Dinoflagellés*: Noctiluca* Suriray (genre libre) et *Blastodinium* Chatton (genre parasite). Thèse Doc. Etat Fac Sci Paris , 240 p 80 pl

Soyer-Gobillard MO (2019a) André LWOFF (1902-1994), Nobel Prize (1965): The man, the scientist, the friend, the artist. Arch Appl Med **1**:1–10

Soyer-Gobillard MO (2019b) Dinoflagellates. In Schmidt TM (ed) Encyclopedia of Microbiology **vol. 2**, Eukaryotic Microbes 4th edn Elsevier, pp 28–49

Soyer-Gobillard MO, Dolan M (2015) Chromosomes of protists: the crucible of evolution. Int Microbiol **18**:209–216

Soyer-Gobillard MO, Schrével J (2003) André Lwoff (1902-1994), Nobel prize of medicine, as protistologist. Protist **154**:455–468

Soyer-Gobillard MO, Schrével J (2020) The Discoveries and Artistic Talents of ÉDOUARD CHATTON and ANDRÉ LWOFF, Famous Biologists. Cambridge Scholars Publishing (CSP), 248 p

Soyer-Gobillard MO, Geraud ML, Coulaud D, Barray M, Theveny B, Revet B, Delain E (1990) Location of b- and z-DNA in the chromosomes of a primitive eukaryote dinoflagellate. J Cell Biol **111**:293–308