nature cell biology

# Milestones in light microscopy

**A supplement contextualizes key advances in light microscopy over 400 years, while the Tara expedition sets sail to explore oceanic ecosystems at the microscopic level.**

Light microscopy is central to biological research; for example, all but two of the papers in this issue contain microscopy data. It is easy to overlook that the bread and butter microscopy tools and techniques of the cell biologist are built on breakthrough technological advances that fundamentally changed the way cells and molecules can be examined. Indeed, cell biological data have undergone a revolution from largely non-quantitative representative observations in fixed cells to high-throughput quantitative data in live cells. In recognition of the key role played by light microscopy in modern cell biology, we decided to focus the ninth in our series of 'Milestones' on this technology: we are pleased to present *Milestones in Light Microscopy*, developed in collaboration with *Nature Methods* and *Nature Reviews Molecular Cell Biology*. The supplement celebrates historical advances in imaging, from the first microscope to recent techniques that reach beyond the diffraction limit of light. As in previous Milestones, this collection highlights key achievements within a field in a series of short summaries of the original research papers, with the aim of presenting each development in the context of its relevance to biologists at the time. A panel of 23 leading experts aided us in extracting the most influential publications. Given the proliferation of significant advances and our desire to contain the supplement in a non-encyclopedic form, the selection was in this case particularly difficult, involving considerable debate among advisors and editors. Consequently, some may feel that certain advances or papers were overlooked.

The historical observations made by Hooke and van Leeuwenhoek in the 17$^{th}$ century, mark the start of imaging in biology and allowed the former to coin the term 'cell'. In the late 1800s and early 1900s, histological dyes were developed that allowed cellular structures to be observed. However, the more detailed architecture of unstained transparent biological samples cannot be seen with the normal bright field microscope and the development of contrast enhancing techniques — phase contrast and differential-interference contrast (DIC) — was vital in enabling the study of these. Abbe's theory on the resolution limit remains fundamental to modern lens design. The first fluorescence microscopes from 1911 relied on autofluorescent samples, but soon fluorescent labels were developed and, once Albert Coons managed to tag these to antibodies, the stage was set for the fluorescence microscope to become the invaluable tool it is today. The confocal microscope provided increased contrast and resolution, and enabled researches to look deeper into tissues, while the more recent TIRF (total internal reflection) microscopy gives a detailed view of processes at the cell edge. No fewer than three Milestones discuss green fluorescent protein and its derivatives, illustrating the extraordinary impact that the fluorescent proteins have had on biological research, as also marked by last year's Nobel prize in Chemistry (*Nature Cell Biol.* **10,** 1233; 2008). Not only can the dynamic behaviour of proteins and cells be studied in their intact environment, but with fluorescent protein-based biosensors and photoactivatable probes, signal transduction events can be directly visualized and populations of molecules followed over time. The 'F-techniques', fluorescence correlation spectroscopy (FCS), fluorescence recovery after photobleaching (FRAP) and Förster resonance energy transfer (FRET), have opened the door for sophisticated analyses of protein movement and interactions. These are but a few of the advances discussed in the supplement. In the present era of rapidly advancing imaging technologies, new developments are hard to predict. Although the Milestones highlight some emerging technologies, such as light sheet microscopy and 'super-resolution' techniques, we have left developments since 2006 for a future series. A hint of what is on the horizon is instead given by Jason Swedlow in one of two specially commissioned commentaries. In the other, Dave Piston discusses the impact of technological development, as much of the hardware (such as lasers, computers and cameras) that was invented for purposes other than microscopy was nonetheless essential for advances in the field.

Milestones and commentaries are freely accessible online, and a selection of related articles from the participating journals is freely available for six months. The website is complemented by a library of relevant papers from the *Nature* journals. We would like to acknowledge the support from EXFO, Invitrogen Molecular Probes and Carl Zeiss, and to warmly thank our advisors for their input. We hope that the Milestones supplement will be a useful educational tool and an inspiration. For anyone who has looked down a microscope (or at the attached computer screen these days) the appeal of light microscopy is obvious — it's simply enthralling to see what's going on in living cells in real time!

## Another milestone exploration

Microscopes were also in the arsenal of early biological explorers such as Alexander von Humboldt and Charles Darwin. It is all too rare in these busy times of grant deadlines, publication pressures and teaching responsibilities for a cell biologist to contemplate journeys of discovery into unknown territories far from their familiar research. While such endeavours can take the form of exotic research topics or approaches, on rare occasions someone dares to go one step further: Eric Karsenti of EMBL has gathered a group of enthusiasts (representing a total of 50 laboratories from 15 different countries) to launch an ambitious three year quest to examine how global warming affects plankton and thus, ultimately, global oxygen production. Not only is this a dramatic departure for Karsenti from his long-standing research interests in the cytoskeleton to genomics and geobiology, but it is a dramatic departure from the setting of his laboratory. Tara, an elegant 36 m long two master, is an oceanographic research ship re-equipped with advanced imaging equipment, including state of the art confocal microscopes, for the purpose of collecting and studying biological samples from ecologically relevant locations. Whereas the very first microscopic organisms van Leeuwenhoek set eyes on were sweet water protist, the microecology of the oceans remains largely uncharted — a major knowledge gap given its important role in fragile oceanic ecology and, indeed, climate regulation.

It is only too fitting to launch this epic plan in the year of Darwin's anniversary. The expedition is making a splash through daily webcasts and extensive media coverage, which will serve to inspire budding researchers by showcasing the excitement of cell biology that addresses a pressing cause in a dramatic setting and relevant context. Our hope is that some truly unexpected discoveries will emerge from the exploration of territories that remain as uncharted as at the time of the Beagle expedition, at least at the microscopic level. We salute captain Karsenti and wish him *bon voyage*.