smaller *d*-orbital splitting — the shifting of each *d* orbital in a transition metal to a distinct energy value, caused by ligand binding — in iron(II) complexes than in ruthenium(II) or iridium(III) complexes with the same ligands[5]. Ruthenium(II) and iridium(III) complexes can therefore possess 'ligand-field' states in which the unoccupied *d* orbitals are higher in energy than the charge-transfer excited state. By contrast, in the ligand-field states of iron(II) complexes, these orbitals are lower in energy than the charge-transfer excited state. Such low-lying orbitals cause charge-transfer excited states[6] to deactivate on timescales of femtoseconds (1 fs is 10^{-15} s) to picoseconds (1 ps is 10^{-12} s), too fast for light-emission processes to be observed.

Fortunately, ligand-field states can be tuned by the electronic nature of the ligands. The combination of strong σ-donation (a process in which electron density is provided from ligands to the metal, along the ligand–metal bond axes) and strong π-back donation (in which *d*-orbital electron density from the metal populates 'anti-bonding' orbitals on the ligands) can induce large *d*-orbital splitting (a strong ligand field), moving the deactivating ligand-field states to higher energy values. This effect is primarily responsible for the extension of excited-state lifetimes of iron(II) MLCT complexes in the past few years[7] — enabling lifetimes of up to 26 ps to be achieved (ref. 8). Strong ligand fields are also facilitated when ligands adopt regular octahedral arrangements around metals[9].

Chábera and colleagues now report an iron(III) complex, $[Fe(btz)_3](PF_6)_3$ (Fig. 1), in which the judicious use of a special type of ligand (3,3′-dimethyl-1,1′-bis(*p*-tolyl)-4,4′-bis(1,2,3-triazol-5-ylidene); btz) achieves a similar extension of the excited-state lifetime. However, the combination of an iron(III) centre and three btz ligands reverses the direction of the charge-transfer excitation, generating a ligand-to-metal charge-transfer (LMCT) complex. The LMCT excited state emits light at room temperature, has a record lifetime of 100 ps, and potentially represents a paradigm shift in the design of inorganic charge-transfer complexes for excited-state photochemistry.

A unique feature of the $[Fe(btz)_3]^{3+}$ ion in the complex is that the iron(III) centre has a 'low-spin' electron configuration that leaves one electron unpaired; to put it another way, this configuration provides the electron vacancy needed to promote an LMCT transition. Such configurations featuring one unpaired electron are termed doublets. Chábera *et al.* studied the redox properties of $[Fe(btz)_3]^{3+}$, and found that its first reduction process (in which the complex accepts one electron) occurs at the metal centre, whereas the first oxidation process (the loss of a single electron) occurs in the btz ligands. This implies that, during the lowest-energy light-induced charge-transfer process of $[Fe(btz)_3]^{3+}$, the transferred electron terminates in an orbital on the metal ion.

This LMCT transition produces an excited state (known as ^{2}LMCT) that, like the ground state, is a doublet. The complex in the ^{2}LMCT state therefore retains its ground-state geometry, so that little energy is lost to molecular rearrangement processes resulting from the charge shift. This tallies with Chábera and co-workers' observation that the light emitted when the excited complex relaxes to its ground state is of only slightly lower energy than the light needed to excite the complex in the first place. The retention of ground-state geometry in the ^{2}LMCT state positions the molecule for spin-allowed photoluminescence — the doublet excited state relaxes to a doublet ground state with the emission of radiation, in a process that conserves electron spin.

Although the quantum yield for this photoluminescence (the amount of light emitted per photon absorbed) is extremely small, this is the first iron-based charge-transfer complex to be reported that photoluminesces at room temperature. Other photoluminescent LMCT complexes have been reported, but are rare, and detailed investigations of their excited-state dynamics are only beginning to emerge[10].

Given the unusual electronic structure of the btz ligand, it is difficult to know whether similar design principles to those underpinning $[Fe(btz)_3]^{3+}$ could be applied to other complexes of abundant transition metals to extend their excited-state lifetimes and turn on photoluminescence at room temperature. Nevertheless, Chábera *et al.* have provided a glimpse into fundamental and potentially useful aspects of LMCT excited states that are ripe for further exploration. A lifetime of 100 ps is already sufficient to drive interfacial electron-transfer chemistry at semiconductor surfaces, as required for solar-cell applications, so the future looks bright for iron-based charge-transfer complexes. ■

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1. Chábera, P. *et al. Nature* **543,** 695–699 (2017).
2. Hagfeldt, A., Boschloo, G., Sun, L., Kloo, L. & Pettersson, H. *Chem. Rev.* **110,** 6595–6663 (2010).
3. Shinar, J. (ed.) *Organic Light Emitting Devices: A Survey* (Springer, 2004).
4. Castellano, F. N. *Acc. Chem. Res.* **48,** 828–839 (2015).
5. Demas, J. N. & DeGraff, B. A. *Anal. Chem.* **63,** 829A–837A (1991).
6. Monat, J. E. & McCusker, J. K. *J. Am. Chem. Soc.* **122,** 4092–4097 (2000).
7. Harlang, T. C. B. *et al. Nature Chem.* **7,** 883–889 (2015).
8. Liu, L. *et al. Phys. Chem. Chem. Phys.* **18,** 12550–12556 (2016).
9. Hammarström, L. & Johansson, O. *Coord. Chem. Rev.* **254,** 2546–2559 (2010).
10. Adams, J. J. *et al. Inorg. Chem.* **54,** 11136–11149 (2015).

In Retrospect

Forty years of cellular clues from worms

The cell divisions that occur when a larva develops into an adult *Caenorhabditis elegans* worm were described in a cell-lineage map in 1977. The work has provided the foundation for many discoveries about developmental mechanisms.

PAUL W. STERNBERG

Forty years ago, John Sulston and Robert Horvitz[1] published a landmark paper that launched the nematode worm *Caenorhabditis elegans* on course to becoming a premier organism in which to study one of life's mysteries: how the cells of an embryo develop to form an adult animal. In the 1970s, the biggest questions of molecular biology were thought to have been answered, and the two authors joined Sydney Brenner in tackling the next big unsolved problem — how genes control development. Progress was being made at that time in understanding the life cycle of viruses called bacteriophages through genetics and elegant experimental logic, and *C. elegans* seemed perfect for taking a similar approach to investigating the development of a multicellular organism.

C. elegans are transparent, making them easy to observe using an optical technique, called Nomarski differential interference contrast microscopy, that highlights density differences to reveal subcellular detail, such as the nucleus and chromosomes at the mitotic stage of cell division. By means of this technique, Sulston and Horvitz made continuous observations of living worms, using hand drawings to record cell positions, divisions, migration, differentiation and, in some cases, programmed death, for each cell in the newly hatched worm until it reached adulthood two days later. This process was aided by the consistency of development that results from the genetic uniformity of the self-fertilizing *C. elegans* hermaphrodite and

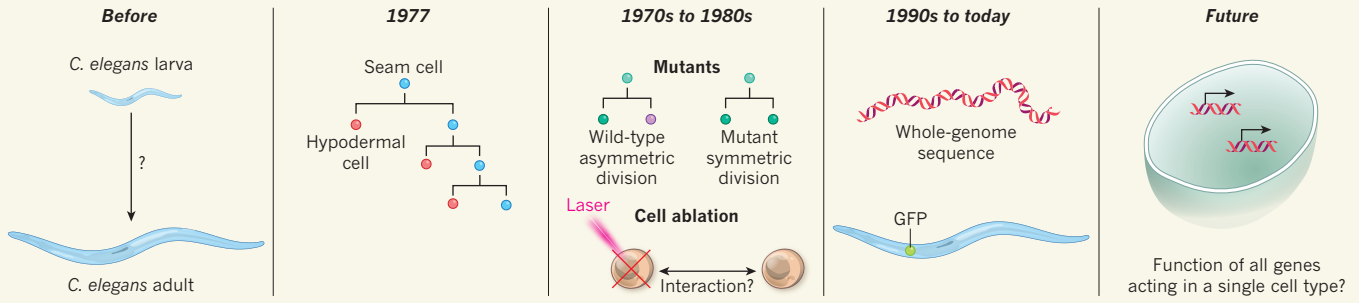

Figure 1 | Understanding development at a cellular level in the worm *Caenorhabditis elegans*. How a larva develops into an adult animal at the cellular level was unknown until Sulston and Horvitz[1] reported the cell lineage of post-embryonic development in *C. elegans* in 1977. The authors recorded the cell divisions in the worm, creating lineage trees such as that for the V1 cell (a section of which is shown), which gives rise to hypodermal cells and seam cells. Experiments in the 1970s and '80s investigated the regulation of worm development by analysing mutants that have abnormal patterns of cellular division, including mutants that undergo symmetric rather than asymmetric divisions, and by laser-based cell-ablation studies to identify the role of interactions between cells. Since the 1990s, the publication of the complete *C. elegans* genome sequence[17] and the use of green fluorescent protein (GFP) to monitor specific cells have further aided studies of *C. elegans* development. Perhaps in the future, our understanding of worm development will reach the level of knowing the function of all genes in a single cell type.

its invariant pattern of cell divisions.

The authors assembled a cellular lineage — an enormous cellular 'family tree' of all the cells in the worm. Sulston and Horvitz's work, and other studies[2,3], described how the newly hatched larvae containing 550 cells develop into an adult hermaphrodite that has 959 cells, or an adult male with 1,031 cells. In 2015, a cell division in the head of the male[4] was reported that Sulston and Horvitz had not spotted.

An obvious initial conclusion about the mechanisms driving worm development was that the fate of a cell probably depends on its ancestry, suggesting that daughter cells are different because of unequal inheritance of cellular components during cell division. Another insight came from the observations of similar cell-division patterns in different sections of the lineage. For example, the V1 cell divides in a repetitive pattern to form cells on the worm's outer surface. In part of the V1 lineage, cell division occurs to form an anterior hypodermal cell, which does not undergo further division, and a posterior seam cell. The posterior seam cell divides to form another anterior hypodermal cell and a posterior seam cell, and this division pattern is repeated (Fig. 1). This pattern also occurs for other cell lineages, including V2, V3, V4, V5 and V6, suggesting that a small number of subprograms might be used more than once to drive development. Deviations from this pattern, such as the formation of a neuronal precursor cell in part of the V5 cell lineage, hinted that each subprogram was also controlled by a specific genetic module.

The worm's invariant cell lineage offers an advantage for genetic studies because any change will stand out in stark contrast to the usual pattern. On the basis of the wild-type development, potential mechanisms of developmental control could be predicted, and these predictions suggested that mutants with specific types of abnormal development could be found. Indeed, many such mutants were identified by the authors and by their directly or indirectly trained scientific 'descendants', leading to the discovery of a staggering array of key developmental-control genes, and launching a thousand careers, including my own.

Cell-lineage mutants were identified rapidly, and many of these illuminated core developmental processes. For example, the *unc-86* gene was found to have a role in mother–daughter identity in neuronal lineages and provided hints about how daughter cells become different from their mother cells[5]. Mutations[6] of the *lin-12* gene, which encodes Notch-family receptor proteins, affect cell fates in small groups of multipotent cells (those able to form more than one cell type), providing insights into how a cell's neighbours specify its fate.

"The Wnt signalling pathway demonstrates the interplay between intrinsic and extrinsic factors in* C. elegans *development."

The *lin-4* gene[7] was shown to affect the relative timing of developmental events, and was later identified[8] as encoding the first known microRNA, an evolutionarily conserved non-coding RNA molecule that affects gene expression. The control of region-specific cell identity was revealed by mutations[9] in the gene *mab-5*. And mutants in the *lin-17* gene, which encodes a receptor for the intercellular signalling protein Wnt, was found to have some identical daughter cells that should have been different[10], thereby revealing mechanisms of control of asymmetric divisions. The normal developmental roles of the cancer-promoting proteins epidermal growth factor (EGF) and Ras were also found by analysing mutations[11] that altered cell lineages.

Another approach to investigating *C. elegans* development was by cell ablation using a focused laser microbeam. The concept of development regulated by cell-intrinsic mechanisms was challenged by the results of such ablation experiments[12,13], in which the destruction of some cells changed the fate of their neighbouring cells. This idea was also supported by cases of indeterminate development, in which a pair of cells assumed positions at random and their fate depended on their position[1–3].

The pendulum swung towards the need for cell interactions in development when some of the cell-lineage mutants were found to affect signalling pathways between cells regulated by the proteins Notch, Wnt and EGF (ref. 14). Yet the pendulum swung back towards cell-intrinsic mechanisms when Wnt signalling was shown to be required in a parental cell that divides along an anterior–posterior axis to generate daughter cells that have distinct fates[15]. Wnt signalling in the direct parent or grandparent of a cell can affect its fate, as was shown by the finding that four cellular 'cousins' have distinct states that depend on the level of Wnt signalling received from their parents and grandparents[16]. This concept fits beautifully with the modularity of *C. elegans* lineages, because most cells undergo only one to three divisions. Thus, the role of the Wnt signalling pathway demonstrates the interplay between intrinsic and extrinsic factors in *C. elegans* development.

C. elegans was the first animal to be completely described at the cellular level in terms of its anatomy, development and neuronal connections, and also the first to have its genome sequenced[17]. We could, in principle, discover the effect of every gene on every cell in the worm. The use of green fluorescent protein as a marker has greatly aided the ability to identify and monitor cells. Next-generation DNA sequencing can define gene expression in individual cells with exquisite temporal resolution[18], and, along with ongoing efforts to knock out each gene in the *C. elegans* genome, we can

hope that we might discover the function of all genes in a single cell type. Such progress would help to answer fundamental questions about the relationship between an organism's genetic make-up (its genotype) and its physical form (phenotype). Extension of such understanding to a parent cell and its two progeny, coupled with cell-biological experiments, would exploit the potential unlocked by Sulston and Horvitz to reveal how the lineage history of a cell influences its fate. ■

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1. Sulston, J. E. & Horvitz, H. R. *Dev. Biol.* **56,** 110–156 (1977).
2. Kimble, J. & Hirsh, D. *Dev. Biol.* **70,** 396–417 (1979).
3. Sulston, J. E., Albertson, D. G. & Thomson, J. N. *Dev. Biol.* **78,** 542–576 (1980).
4. Sammut, M. *et al. Nature* **526,** 385–390 (2015).
5. Chalfie, M., Horvitz, H. R. & Sulston, J. E. *Cell* **24,** 59–69 (1981).
6. Greenwald, I. S., Sternberg, P. W. & Horvitz, H. R. *Cell* **34,** 435–444 (1983).
7. Ambros, V. & Horvitz, H. R. *Science* **26,** 409–416 (1984).
8. Lee, R. C., Feinbaum, R. L. & Ambros, V. *Cell* **75,** 843–854 (1993).
9. Kenyon, C. *Cell* **46,** 477–487 (1986).
10. Sternberg, P. W. & Horvitz, H. R. *Dev. Biol.* **130,** 67–73 (1988).
11. Horvitz, H. R. & Sternberg, P. W. *Nature* **351,** 535–541 (1991).
12. Sulston, J. E. & White, J. G. *Dev. Biol.* **78,** 577–597 (1980).
13. Kimble, J. *Dev. Biol.* **87,** 286–300 (1981).
14. Sawa, H., Lobel, L. & Horvitz, H. R. *Genes Dev.* **10,** 2189–2197 (1996).
15. Lin, R., Hill, R. J. & Priess, J. R. *Cell* **92,** 229–239 (1998).
16. Zacharias, A. L., Walton, T., Preston, E. & Murray, J. I. *PLoS Genet.* **11,** e1005585 (2015).
17. The C. elegans Sequencing Consortium. *Science* **282,** 2012–2018 (1998).
18. Schwarz, E. M., Kato, M. & Sternberg, P. W. *Proc. Natl Acad. Sci. USA* **109,** 16246–16251 (2012).

MARINE CONSERVATION

How to heal an ocean

Marine protected areas are being implemented at an accelerating pace, and hold promise for restoring damaged ecosystems. But glaring shortfalls in staffing and funding often lead to suboptimal outcomes. SEE ARTICLE P.665

BORIS WORM

Humans have arrived at an interesting juncture in their relationship with the ocean. After using (and overusing) it for centuries as a free food source, highway and dumping ground[1], there is a growing political resolve to reverse some of the damage that has been inflicted[2]. Marine protected areas (MPAs; Fig. 1) represent a cornerstone of our global strategy to heal compromised ecosystems[2,3], but their success has been varied and uneven in practice[4–6]. On page 665, Gill *et al.*[7] expose some of the reasons behind this variation, identifying global capacity gaps — shortfalls in staffing, funding and scientific monitoring — that have been opened by the rapid, but woefully underfunded, expansion of protected areas worldwide.

On the surface, the extraordinary growth of MPAs looks like an environmental success story. Since the 1960s, global coverage of these areas has been growing exponentially at a rate of more than 8% per year (Fig. 2). The past decade, in particular, has seen a dramatic expansion of extremely large MPAs in some of the remotest corners of our planet, and especially of strongly protected areas that ban commercial extraction of natural resources such as fish, minerals or oil, and where recreational or subsistence fishing is light.

Later this year, the world's largest conservation area of any kind will come into force, protecting 1.55 million square kilometres of the Ross Sea in Antarctica. This means that nine of the ten largest protected areas on Earth will be marine. Still, the combined coverage of designated and implemented MPAs currently accounts for just 4% of total ocean area (Fig. 2), compared with 15% on land. Several international agreements have committed coastal nations to reach higher to correct this imbalance, aiming at 10% MPA coverage by 2020, as ratified under the Convention on Biological Diversity and reaffirmed by the United Nations Sustainable Development Goals.

Despite such commitments, further MPA expansion can be controversial, and the utility and effectiveness of protected areas are sometimes questioned. Like parks on land, MPAs can flourish or fail. Previous analyses[4–6] that sought to disentangle some of the complex reasons for this have focused largely on biophysical aspects, such as the size, age, connectivity or remoteness of a particular area, all of which proved influential. Social factors influencing the effectiveness of management or governance have been more difficult to quantify. Gill *et al.* provide a new perspective by focusing squarely on the role of people in MPA effectiveness[7].

The authors compiled an impressive database of management features in 433 MPAs around the world, and matched it with fish population data in 218 MPAs, thus providing a 'deep dive' into the relationship between socioeconomic factors and biological outcomes. The management data included a variety of indicators, ranging from budget constraints to the inclusion of stakeholders in decision-making processes.

The aggregate results were sobering, with 79% of MPAs in the global sample not meeting even half of the thresholds for adequate

JIANGANG LUO/MARINE PHOTOBANK

Figure 1 | A diver monitors marine life at the Dry Tortugas Ecological Reserve in Florida. Gill *et al.*[7] report that the biggest factors in the success of marine protected areas are adequate staffing and funding.