Sophisticated lessons from simple organisms: appreciating the value of curiosity-driven research

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ABSTRACT
Long before scientists learned to grow miniature organs from human cells in a dish, biological research was performed using organisms that were accessible: garden peas, sea urchins collected at low tide, newt eggs, and flies circling rotten fruit. These organisms help us to understand the world around us, attracting and inspiring each new generation of scientists with the promise of mystery and discovery. Subsequent studies repeatedly showed that fundamental biological mechanisms discovered in such simple organisms are conserved in more complex organisms, including humans. Yet, biologists are increasingly being tasked with problem solving rather than being allowed to follow the path of discovery. Here, we provide examples of important lessons learned from research using selected non-vertebrate organisms. We argue that, for the purpose of understanding human disease, simple organisms cannot and should not be replaced solely by human-cell-based culture systems. Rather, these organisms serve as powerful discovery tools for new knowledge that may subsequently be tested for conservation in human-cell-based culture systems. In this way, curiosity-driven biological research in simple organisms has and will continue to pay huge dividends in both the short and long run for improving the human condition.

KEY WORDS: C. elegans, Drosophila, Invertebrates, Sea urchin, Yeast

Introduction
The emphases and directions of medical practice are influenced not only by scientific evidence but also by factors such as financial interests and societal trends. For example, interventions such as cupping, the application of suction cups to the skin, have gained popularity not due to scientific evidence (Lee et al., 2011), but based on popular figures promoting the practice. In contrast, impactful progress in medicine invariably follows major advances in biological understanding due to scientific discovery. Examples extend from Pasteur’s recognition of the infectious basis of many diseases to the recent discovery of CRISPR/Cas9-based genome editing (see Box 1 for a glossary of terms) (McNutt, 2015; Porter, 1961). Such advances in our understanding of biology have driven revolutionary advances in medicine, in addition to the incremental and unsteady progress that medicine typically makes. Despite this history of success, science is increasingly being tasked to focus on problem solving rather than discovery (Fang and Casadevall, 2010; Hand et al., 2013; Minogue and Wolinsky, 2010). It is therefore with a sense of urgency that we write this article to defend the winning strategy of ‘discovery first’.

Our understanding of human diseases and the ability to treat them hinges on a foundation of knowledge about basic biology. This knowledge has been gained largely by research using organisms that are experimentally tractable in ways that humans or human cells can never be. Those who do not know the history of yeast, flies and other non-vertebrate organisms and their contributions to biomedicine may consider such studies as unworthy of participation or of funding. Given the growing excitement about human stem-cell-based cell culture systems, particularly organoids (Box 1), some may argue that organisms that do not recapitulate all of the complexity of humans will cease to be useful in the near future. This line of thought is not only incorrect but could hamper scientific progress.

In this article, we highlight the continuing value of curiosity-driven research, with a focus on widely used non-vertebrate experimental organisms. These are often referred to as ‘model organisms’ (Box 1) to highlight their utility in discovering and understanding fundamental biological principles that also apply to other organisms, particularly humans, and to mechanisms of disease. Although we use this term for the remainder of the article, we wish to emphasize that research using such organisms need not be motivated solely for the purpose of modeling human biology or human disease.

We will illustrate the utility of each model organism with seminal historical examples, rather than providing a comprehensive overview. We hope to motivate the reader to look into additional contributions. Indeed, it is hard to think of an aspect of biology that has not benefited from studies in model organisms, be it behavior (e.g. Kravitz and Fernandez, 2015), aging (e.g. Kenyon, 2011) or memory (e.g. Dubnau and Tully, 1998). Highlighting past achievements will not necessarily encourage future work. Therefore, we also discuss potential future breakthroughs that might come from curiosity-driven research in model organisms. This is not to say that we disagree with the well-supported contention that no one can predict at the time of discovery how applicable findings from basic research will turn out to be. For example, John S. Dexter, investigating mutant notched wings of fruit flies (Dexter, 1914), could not have predicted the prominent role of the Notch cell surface receptor in cancer (Nowell and Radtke, 2017; Ranganathan et al., 2011). We do not have such foresight either. Nonetheless, it is worth pondering what more we can learn from model organisms, particularly in the era of human stem cells, organoids and facile genome editing using CRISPR/Cas9-based technologies.

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The historical impact of model organism research

Biological research was historically driven by curiosity about the natural world. The four model organisms discussed below – yeast, fruit fly, worm and sea urchin – came into use for practical reasons, such as the ease of rearing or their natural abundance. We describe examples of their contributions and discuss their attributes that have enabled investigators to ask questions that would be impractical or impossible to address using mammalian experimental systems.

Yeast

We owe a great deal of our understanding of eukaryotic biology to two species of yeast: *Saccharomyces cerevisiae* (budding yeast) and *Schizosaccharomyces pombe* (fission yeast). They have been standout model organisms largely because of their simple life cycles and suitability for large-scale genetic analyses as described below. The relevance of such simple cells to the biology of complex organisms might not be obvious, but, as we discover time and time again, fundamental biological mechanisms are largely the same in simple and more complex organisms, as nicely illustrated by two influential studies (Table 1).

The first study asked a simple but fundamental question: which genes control how one cell becomes two? Leland Hartwell and his colleagues recognized that the way budding yeast grew and divided would allow the identification of mutants that are defective in the cell division cycle by simply observing the cells (Hartwell et al., 1970, 1973). The researchers took advantage of a useful feature of yeast. Because yeast can grow at different temperatures, one can isolate conditional mutants (Box 1) in which a gene product is active at a low temperature but becomes inactive at a high temperature. These temperature-sensitive conditional mutants could be grown and propagated at the low temperature and their defects examined after a shift to the high temperature. Because yeast cells are small and easy to grow, Hartwell and his colleagues were able to isolate many temperature-sensitive mutants (Fig. 1A). By simply observing cells at the high temperature, these researchers identified mutants that were blocked at different stages of the cell cycle (Fig. 1B).

The identification of the CDC genes defined a new problem: how do they work in concert to guide cell proliferation, perhaps the most basic of life’s processes? Combining molecular biology and genetics in yeast, and with contributions from other model organisms (e.g. sea urchins; see below), the roles of the different CDC gene products were discovered. These advances came with the realization that the genes that control the cell cycle are largely conserved from yeast to human (e.g. Hartwell et al., 1970, 1973; Nurse, 1975). An exceptionally good example of this evolutionary conservation is the gene encoding cyclin-dependent kinase 1 (CDK1); the human CDK1 gene was first identified by its ability to substitute for the yeast version and allow yeast that were mutant for CDC28 (CDK1 homolog) to continue dividing (Lee and Nurse, 1987). The regulation of CDK1 by inputs from many of the CDC genes governs the progress of the cell cycle. Moreover, the impact of yeast studies extended in many directions, raising questions such as: how is cell proliferation regulated during development; how do disruptions in this regulation derail proliferation control in cancer; and how can drugs modulating the function of CDC genes be used to treat cancer (Table 1)?

In another example, a simple observation in budding yeast ignited research in the field of protein secretion, which is of central importance to cellular function (Novick et al., 1980; Novick and Schekman, 1979). The isolation of the first secretory (SEC) mutants, in the laboratory of Randy Schekman, was made possible because of the realization that protein secretion is required to build a cell wall outside the cell membrane (Novick and Schekman, 1979). Subsequently, secretion-defective yeast mutants were recognized to be denser than their wild-type counterparts. This trait was used to isolate mutants in additional secretory pathway components using gradient sedimentation (Novick et al., 1980). Again, the genes identified provided the foundation for the mechanistic dissection of protein secretion. The resulting understanding of how an intricate system of membranous vesicles traffics proteins from the inside to the outside of cells also led to our understanding of other processes, such as how nerves signal to one another through the release of neurotransmitters. Yeast studies thus provided an entrée into a new area of cell biology (Table 1).
Table 1. Studies on non-vertebrate eukaryotic model organisms that were recognized by Nobel prizes in Physiology or Medicine in the last 25 years*

| Model organism          | Awardees                              | Year of award; citation from nobelprize.org | Representative references |
|-------------------------|---------------------------------------|---------------------------------------------|---------------------------|
| Yeast, sea urchin, clam | “Leland H. Hartwell, *Tim Hunt and *Paul M. Nurse” | 2001; “for their discoveries of key regulators of the cell cycle” | Evans et al., 1983; Hartwell et al., 1970, 1973; Hunt et al., 1992; Lee and Nurse, 1987; Nurse, 1975; Standart et al., 1990; Standart and Hunt, 1990 |
| Yeast                   | “James E. Rothman, *Randy W. Schekman and **Thomas C. Südhof” | 2013; “for their discoveries of machinery regulating vesicle traffic, a major transport system in our cells” | ***Balch et al., 1984; ***Block et al., 1988; ***McMahon et al., 1995; Novick et al., 1980; Novick and Schekman, 1979 |
|                        | Yoshinori Ohsumi                      | 2016; “for his discoveries of mechanisms for autophagy” | ***Mizushima et al., 1998; ***Takeshige et al., 1992 |
| D. melanogaster         | Edward B. Lewis, *Christiane Nüsslein-Volhard and *Eric F. Wieschaus | 1995; “for their discoveries concerning the genetic control of early embryonic development” | ***Lewis, 1978; Nusslein-Volhard and Wieschaus, 1980 |
|                        | “Bruce A. Beutler and *Jules A. Hoffmann” | 2011; “for their discoveries concerning the activation of innate immunity” | Lemaitre et al., 1996; Poltorak et al., 1998 |
|                        | Jeffrey C. Hall, Michael Rosbash and Michael W. Young | 2017; “for their discoveries of molecular mechanisms controlling the circadian rhythm” | ***Bargiello et al., 1984; ***Hardin et al., 1990; ***Konopka and Benzer, 1971; ***Zehring et al., 1984 |
| C. elegans              | *Sydney Brenner, “H. Robert Horvitz and *John E. Sulston” | 2002; “for their discoveries concerning genetic regulation of organ development and programmed cell death” | Brenner, 1974; Ellis and Horvitz, 1986; Sulston and Horvitz, 1977; Yuan et al., 1993 |
|                        | *Andrew Z. Fire and *Craig C. Mello | 2006; “for their discovery of RNA interference – gene silencing by double-stranded RNA” | Fire et al., 1998 |
| Tetrahymena, yeast      | Elizabeth H. Blackburn, Carol W. Greider and Jack W. Szostak | 2008; “for the discovery of how chromosomes are protected by telomeres and the enzyme telomerase” | ***Greider and Blackburn, 1985; ***Szostak and Blackburn, 1982 |

*Awardees whose work in the model organism is discussed in this article, listed in the order in which their work is discussed. **These awardees shared the prize for work in experimental models other than non-vertebrate eukaryotic organisms; their representative works are cited (*** ) but not discussed in this article. Source: https://www.nobelprize.org/.

These genetic screens in yeasts (Fig. 1A) have changed the face of modern science and stimulated subsequent genetic analyses that allowed researchers to place genes in functional pathways even when insight into the biochemical properties of the gene products was lacking (e.g. Garvik et al., 1995). The result is deep understanding of cell cycle regulation and membrane biology underpins many branches of modern biology and has guided medical research and biotechnology. For example, our understanding of protein secretion from yeast allowed us to manufacture recombinant human insulin for biotechnology. For example, our understanding of protein secretion from yeast allowed us to manufacture recombinant human insulin for biotechnology.

**Drosophila**

How does one choose examples to adequately illustrate the impact of a model organism that is responsible for the confirmation of Mendelism (Box 1), the discovery of the first mutation and the demonstration that genetic traits are carried on chromosomes? The fruit fly *Drosophila melanogaster* has taught us many fundamental biological mechanisms, thanks to powerful genetic tools, which include phenotypic tractability, special ‘balancer’ chromosomes that allow long-term maintenance of lethal mutations, high fecundity and short life cycle, and the dedication of early fly geneticists, who learned to recognize every bristle on the fly or to detect the subtlest of deviations in wing shape or eye color. Many regulatory pathways studied in modern biology were either discovered in *Drosophila* or organized into regulatory circuits as a result of studies in *Drosophila*. For instance, although studies in mammals identified the Ras oncogenes (and others), we owe our understanding of their function to genetic studies of eye development in *Drosophila*, which revealed that Ras transmits signals from receptor tyrosine kinases (Simon et al., 1991; similar insights from worms are discussed below). Indeed, many signaling pathways of central interest to both normal biology and disease research bear the names of the genes discovered in *Drosophila* by scientists such as Christiane Nüsslein-Volhard, Eric F. Wieschaus and colleagues, including * hedgehog* (Nüsslein-Volhard and Wieschaus, 1980), *Notch* (Dexter, 1914; *Fig. 2A,B*) and *Toll* (Table 1). Toll was identified as a gene that functions in the establishment of dorsal/ventral polarity in the *Drosophila* embryo (Anderson et al., 1985a,b). The elegant genetic dissection of Toll function helped outline an elaborated pathway, which extended from extracellular signals to the activation of key conserved transcription factors, including NF-κB (called Dorsal in flies) and its relatives (Roth et al., 1989; Sen and Baltimore, 1986; Steward, 1989). Subsequently, it was shown that the same signaling system triggers an innate immune response in *Drosophila* through the work of Jules A. Hoffmann and his colleagues, and that this signaling system is the core component of innate immunity in mammals (Lemaitre et al., 1996; Poltorak et al., 1998). This pivotal discovery promoted a dramatic shift in the study of immunology from an emphasis on adaptive immunity to the more conserved innate system; the ratio of PubMed search results for ‘innate immunity’ vs ‘adaptive immunity’ was 1285:2860 in 1980 and 4423:2658 in 2017. Moreover, we now appreciate that NF-κB family transcription factors regulate almost all aspects of cell biology, from proliferation to inflammation and cell death (Zhang et al., 2017).

Unusual as it may seem, bristles on the leg of a fly helped us understand how the activity of genes is epigenetically regulated (Box 1). The regions that looked different within the chromosomes of moss and *Drosophila* were recognized in the early 1900s; densely stained heterochromatin and less densely stained euchromatin (Passarge, 1979). Continuing investigations showed that genes in heterochromatin were transcriptionally repressed and that this repression could be passed on to the next generation of cells (Brown and Nur, 1964). Surprisingly, the discovery of the
underlying mechanisms of epigenetic inheritance involved studies of flies’ bristles. The accurate formation of a set of leg bristles on male Drosophila, called the ‘sex comb’ for their role in mating (Fig. 2C,D), depends on the mechanisms that form heterochromatin. Mutations that result in the formation of additional sex combs identified a number of genes, with names like Polycomb and extra sex combs (Hannah-Alava, 1958; Reute and Spierer, 1992). Subsequent analyses demonstrated that these genes encode proteins that work together to regulate the addition or removal of chemical modifications to and from histones, the proteins that bind DNA and determine how chromatin (Box 1) is packaged (Piunti and Shilatifard, 2016). Another assay in Drosophila allowed the isolation of additional chromatin-modulating genes. The red color of the fly eye requires the expression of the gene white, which encodes a pigment transporter (O’Hare et al., 1984; Sullivan et al., 1974). Translocation of the white gene near heterochromatin (for example by a chromosome rearrangement) can produce flies with eyes that show ‘variegation’, whereby white patches appear next to normally pigmented red patches (Fig. 2E,F; e.g. Qi et al., 2006). This process is known as position effect variegation (Box 1), and it happens because of changes in chromatin state, so that the white gene locus is in the heterochromatin state and thus transcriptionally inactive in some cells but not others, whereas the DNA sequence of the gene remains unchanged (Ebert et al., 2004). This phenotype illustrates how epigenetic regulation determines gene expression profiles, not on the basis of DNA sequence changes, but on the basis of chromatin state. Founder cells with an epigenetically inactive white gene produce daughter populations that form a white patch in the eye, whereas neighboring cells with a transcriptionally active white gene are red. Mutations that suppress the formation of these white patches are called suppressors of variegation, or Su(var) mutations, and their discovery identified additional chromatin-modulating genes and gene regulatory networks (Ebert et al., 2004; Reute and Spierer, 1992; Sinclair et al., 1983). For example, Su(var)3-9 encodes a histone methyltransferase and Su(var)2-5 encodes a protein, now called heterochromatin protein 1 (HP1), that binds to histones methylated by Su(var)3-9 (Aagaard et al., 1999; Ebert et al., 2004; Eissenberg and Elgin, 2014; Eissenberg et al., 1990; James and Elgin, 1986; Rea et al., 2000; Tschiersch et al., 1994). Polycomb, Su(var)3-9 and HP1 are just three of many genes with crucial roles in maintaining cell identity through epigenetic regulation of gene expression that were discovered in Drosophila. Their initial identification in Drosophila subsequently led to the characterization of their highly conserved counterparts in humans, where they also regulate transcription and influence many aspects of cell identity, physiology and pathogenesis (Allis and Jenuwein, 2016; Lanzuolo and Orlando, 2012; Piunti and Shilatifard, 2016; Schuettengruber et al., 2007; Timms et al., 2016).
Caenorhabditis elegans

Studies of *C. elegans* development, like those in *Drosophila*, led to breakthroughs in our understanding of cellular signaling. Early researchers of *C. elegans* took advantage of the transparent bodies of these small nematodes and used simple polarization or differential interference contrast (DIC) optics (Box 1) to track the fate of cells as an embryo developed (Brenner, 1974; Sulston and Horvitz, 1977). What they learned was the remarkable conservation of cell fate decisions from worm to worm. In fact, cell behavior is so rigid in this organism that Sydney Brenner, John Sulston, Bob Horvitz and colleagues were able to construct an accurate lineage map of all post-embryonic cell divisions and cell deaths (Fig. 3; Sulston and Horvitz, 1977). This was a gold mine for geneticists looking for mutants that deviated from the wild-type pattern of cell division and cell death. *C. elegans* are hermaphrodites and can be mated to themselves (much like Mendel’s garden peas) to produce progeny, typically in large numbers and in the expected Mendelian ratios (Box 1) (Brenner, 1974). These and the other benefits of *C. elegans* have led to many fundamental insights into biology, including the identification of the regulators of Ras signaling that arose from the first cloning of the *Ras* gene (Beitel et al., 1990; Han and Sternberg, 1990; Sternberg and Han, 1998), the discovery of RNA interference (RNAi; Fire et al., 1998; Table 1) and of the first microRNA (Lee et al., 1993; Wightman et al., 1993). The discovery of RNAi by Craig Mello, Andrew Fire and colleagues came on the heel of prior recognition that experimental addition of exogenous RNA could interfere with the expression of the corresponding endogenous genes in plants and worms (Sen and Blau, 2006). Some of the results, such as the finding that both sense and anti-sense strands had the same repressive effect, however, could not be explained by the prevailing model that exogenous RNA interfered with gene function simply by hybridization with the endogenous RNA. Instead, *C. elegans* studies showed that double-stranded RNA acted catalytically to suppress gene expression, thereby spearheading a field that now has wide applications in basic research, biomedicine and pest control, among others (Perrimon et al., 2010).

A key disease-relevant field that *C. elegans* genetics research pioneered is programmed cell death. Apoptosis, a form of programmed cell death, was first recognized in mammalian cells (Kerr et al., 1972) and it was known that enzymes called caspases were important for executing this cellular process. But it was not until Bob Horvitz and colleagues took a genetic approach to analyzing programmed cell death during worm development that our understanding of apoptosis truly advanced. These researchers exploited the invariant sequence of cell division and cell death during *C. elegans* development to screen for mutants in which too many or too few cells died (e.g. Ellis and Horvitz, 1986). Further studies led to the isolation of the gene that encodes the key caspase, *ced-3* (for *cell death mutant number 3*) (Yuan et al., 1993). *C. elegans* studies thus identified the genes that match mammalian caspase genes. Using genetic analysis in *C. elegans*, the enzymes that trigger and execute apoptosis were placed in an ordered pathway and their activators and repressors were identified (Metzstein et al., 1998). These efforts laid the foundation for a deeper understanding of apoptosis in mammalian systems (Table 1).

Sea urchins

Classical forward genetics is not the only tool in the biologist’s toolkit. Research into the embryonic development of marine invertebrates, such as the sea urchin, provides an excellent

![Fig. 3. *C. elegans* cell-lineage map and multi-vulva mutants.](image)

(A) Experiment and data that led to the discovery of RNAi, summarized from Fire et al., 1998. (B) Cell-lineage map of *C. elegans*. Reproduced under a Creative Commons license from Kimble and Seidel, 2013. (C,D) Wild-type and multi-vulva (Muv) mutant worms. * indicates the vulva; arrows point to ectopic vulvae. Figures modified and reproduced under a Creative Commons license from de la Cova and Greenwald, 2012. Specification of the vulval fate occurs through Ras/MAPK signaling (Kornfeld, 1997; Sundaram and Han, 1996). Mutants in which this pathway is misregulated can show the Muv phenotype (e.g. Gu et al., 1998), which led to the discovery of regulators of Ras/MAPK signaling.

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### Table 1

| Gene tested | Phenotype          |
|-------------|--------------------|
| *unc-22*    | Twitching larvae   |
| *unc-54*    | Paralyzed larvae   |
| *hlh-1*     | Lumpy-dumpy larvae |

| Injected RNA | % affected |
|--------------|------------|
| Sense or antisense RNA | 0-2       |
| Sense-antisense RNA hybrid | 80-100    |

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SPECIAL ARTICLE

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example of how combining biochemistry and cell biology results in a powerful discovery strategy. The practical advantages of using sea urchins attracted many researchers: low cost and relative ease of maintenance (Sluder, 2016); availability of a large quantity of sperm and eggs to enable biochemical studies; rapid, nearly synchronous cell cycles (40-90 min depending upon the temperature) that allow one to study multiple divisions in a short time; and optical transparency of the eggs of some species, useful for imaging. Sea urchin zygotuses are remarkably robust and tolerate, for example, mechanical fragmentation or microinjection (Box 1) (Heasman, 2002). The publicly available genome sequence of some species provides access to genes of interest (Cameron et al., 2015; Sea Urchin Genome Sequencing Consortium et al., 2006) and genome editing in zygotes with CRISPR/Cas9 has been reported (Lin and Su, 2016).

Sea urchin gametes and zygotuses have been studied since the late 1800s. Theodore Boveri used polyspermy (Box 1) to force a fertilized egg to divide its contents unequally into daughter cells. He analyzed the resulting abnormal zygotuses and, with crisp deductive logic, Boveri considered and then discarded each cellular component as the bearer of genetic information until he identified the structures that we now call chromosomes (Boveri, 1974). Boveri’s chromosome theory of inheritance was beautifully complemented by Mendel’s work as re-discovered by Sutton (Sutton, 1903) and was amply supported by Morgan’s experimental data from Drosophila (Morgan, 1911). Since then, many fundamental insights into cell biology have been gained using sea urchin gametes and zygotes. These include the identification of bindin, a protein that facilitates egg-sperm interactions for fertilization (Glab & Vacquier, 1977; Vacquier and Moy, 1977); an understanding of how complex changes to the cytoskeleton and cell membranes are orchestrated to form a cleavage furrow (Box 1) (Pollard, 2004); the identification of gene regulatory networks that control early zygotic development (Davidson et al., 2002); understanding how centrosomes (Box 1) are assembled and duplicated (reviewed in Sluder, 2014); and the discovery of the cyclin proteins that drive the cell cycle in sea urchin zygotes and also in clam embryo extracts (Evans et al., 1983; Hunt et al., 1992; Standart et al., 1990; Standart and Hunt, 1990). We, the authors, still remember the excitement at the recognition that the cyclins that drive cleavage in sea urchin zygotes are homologous to the CDC proteins that drive the cell cycle in yeast (Booher and Beach, 1988; Hagan et al., 1988). These and many other findings from sea urchin models paved the way for subsequent discoveries in mammalian systems.

Concluding remarks

As useful as model organisms are, they are not human, and have their limitations. Three-dimensional (3D) cell-culture model systems, such as organoids derived from human stem cells, have been much-hailed because they are the only human-cell-based model system to recapitulate the cellular complexity of tissues (Huch et al., 2017). Challenges exist, such as high cost and the phenotypic variability in traits, including organoid size, shape, cellular composition and 3D architecture, even when the organoids are produced under identical conditions (Huch et al., 2017), but these may be overcome as the technology matures. Lab-grown organoids should be useful for toxicity testing and for the generation of tissues for biobanks, but experts doubt that they will replace animal models as discovery tools (Bredenoord et al., 2017). We would go further to argue that organoids will not and should not replace non-vertebrate model organisms as discovery tools. We, the authors, speculate that the best use of organoids may be to transfer knowledge acquired in model organisms to humans. Given this position, what do we think are future discoveries that could be expected from curiosity-driven research in model organisms?

One obvious answer concerns the inner workings deep within the cell. For example, all cells, regardless of origin, need to make proteins, but also must get rid of proteins that are no longer needed. Genetic studies in budding yeast, complemented by biochemical and structural analyses, are providing us with exquisite details on how the protein machines responsible for destroying other proteins are assembled (e.g. Li et al., 2017; Sokolova et al., 2015). Likewise, we anticipate that studies in yeast and other model organisms may provide a deeper understanding of how proteins are synthesized, how they are modified with different chemical moieties, and how they are transported across and out of the cell. For such dissection of fundamental cellular processes, stem cells or organoids are neither required nor cost effective. Additionally, as with examples described in this article, we expect fundamental insights gleaned from model organisms to lead to better understanding of biology in all organisms, including humans.

Moving beyond cell biology to whole-animal biology, two examples illustrate the kind of new knowledge that can be gained from studying the biology of model organisms. An interesting question that has interested biologists for many years is how organisms, and the organs within them, attain their characteristic size. The Drosophila Hippo pathway plays as a growth rheostat that fine tunes the balance of proliferation and cell death during development, thereby helping organs attain their correct size and shape. It does so by regulating the transcription factor Yorkie (Yki; YAP in human), the targets of which include genes that regulate cell proliferation and apoptosis (Hariharan, 2015; Irvine and Harvey, 2015). The latest twist in the Hippo signaling tale involves Drosophila insulin-like peptide 8 (Dilp8), a peptide hormone secreted by injured tissues (Colombani et al., 2012; Garelli et al., 2012). Secreted Dilp8 accumulates in the circulatory system of the fly, from where it finds its way to the brain and stimulates neurons that subsequently repress the production of the molting hormone, thereby postponing the next stage in fly development. This signaling throughout the animal allows injured organs more time to grow and catch up in size relative to their undamaged counterparts. Strikingly, dilp8 mutant flies lose coordinated organ growth, such that wings are mismatched in size (Garelli et al., 2012). dilp8 was recently found to be a transcriptional target of the Hippo pathway (Boone et al., 2016). The current thinking is that Hippo not only fine tunes growth within a tissue, but also acts via Dilp8 to delay development until organs at distant sites in a body are matched in size. When the pathway is functional, wing growth on opposite sides of the body is coordinated. Subsequently, the Hippo pathway has emerged as an important regulator of the growth needed to repair injured tissues in a variety of organisms, including vertebrates (Hong et al., 2016; Patel et al., 2017; Zhang and Del Re, 2017). Given these roles, it is not surprising that the Hippo pathway – first discovered in Drosophila – is also increasingly implicated in growth control in human cells and in the development of many human cancers (Hong et al., 2016; Patel et al., 2017). This example highlights how a strategy of ‘discovery first’ can be of tremendous benefit to biomedicine.

* C. elegans provides another example of a model in which new insights about biology remain to be made. C. elegans hermaphrodites can produce sperm or egg, but not both simultaneously. A recently published study found that fatty acids (FAs) in the gut can alter the germline of hermaphrodites to stimulate production of either oocytes or sperm (Tang and Han, 2017). FAs exert their effect by altering the level of myristic acid
(a form of FA) in the germline, which in turn affects the level of myristoylation, a process in which myristic acid is covalently attached to proteins. One protein controlled by myristoylation turns out to be the MAP kinase MPK-1, which determines germ-cell fate (Tang and Han, 2017). This biological phenomenon is conserved in Caenorhabditis remanei, a worm species that lives as male or female and not as a hermaphrodite, where the knocking down of the myristic acid processing enzyme ACS-4 can masculinize genetically female worms into producing sperm.

What do these examples tell us about the future the world might hold? They illustrate the complex interconnections between the germline and the soma through nutrients and signaling in worms, and a complex system of growth control and tissue repair that involves multiple organs and many secreted hormones in flies. They exemplify how model organisms can be used to understand how multiple organ systems interact inside an intact body, and to identify molecules that mediate these interactions. Such a deep understanding of biology and its complexities, we argue, will provide a basis for future breakthroughs in medicine. It will be a long time, if ever, before we can model complex interactions in organisms or cultured cells. Until then, it is curiosity-driven research in model organisms that will continue to satiate our appetite for understanding the wonderful and mysterious natural world that we live in.

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References
Aagaard, L., Laible, G., Slenko, P., Schmid, M., Dorn, R., Schotta, G., Kuhlthutig, S., Wolf, A., Leberserger, A., Singh, P. B. et al. (1999). Functional mammalian homologues of the Drosophila PEV-modifier Su(var)3-9 encode centromere-associated proteins which complex with the heterochromatin component M31. EMBO J. 18, 1923-1938.

Ali-Murthy, Z., Lott, S. E., Eisen, M. B. and Kornberg, T. B. (2013). An essential role for the Hedgehog signalling pathway coordinates organ growth and limits developmental variability in Schizosaccharomyces pombe: possible interaction of the gene product with cdc13+ gene and a cytological study of its role in fission yeast mitosis. EMBO J. 22, 2321-2327.

Boveri, T. (1894). On multipolar mitosis as a means of analysis of the cell nucleus. In Foundations of Experimental Embryology (ed. B. H. Willier and J. M. Oppenheimer), pp. 74-97. New York: Hafner Press.

Brunst-Mascher, I., Civelekoglu-Scholey, G. and Scholey, J. M. (2014). Analysis of mitotic protein dynamics and function in Drosophila embryos by live cell imaging and quantitative modeling. Methods Mol. Biol. 1136, 3-30.

Cameron, R. A., Kudtarkar, P., Gordon, S. M., Worley, K. C. and Gibbs, R. A. (2015). Do echinoderm genomes measure up? Mol. Genom 22, 1-9.

Cassio, D. J., Biehs, B. and Kornberg, T. B. (2011). A novel interaction between hedgehog and Notch promotes proliferation at the anterior-posterior organizer of the Drosophila wing. Genetics 187, 485-499.

Colombani, J., Andersen, D. S. and Leopold, P. (2012). Secreted peptide Dlp8 coordinates Drosophila tissue growth with developmental timing. Science 336, 582-585.

Davidson, E. H., Rast, J. P., Oliveri, P., Ransick, A., Celestani, C., Yuh, C. H., Minokawa, T., Amore, G., Hinman, V., Arenas-Mena, C. et al. (2002). A genomic regulatory network for development. Science 295, 1669-1678.

de la Covia, C. and Greenwald, I. (2012). SEL-10/Fbw7-dependent negative feedback regulation of LIN-45/Braf signaling in C. elegans via a conserved phosphodephosphogen. Genes Dev. 26, 2524-2535.

Dexter, J. S. (1914). The analysis of a case of continuous variation in Drosophila by its study of linkage relations. Am. Nat. 49, 712-758.

David, J. and Tully, T. (1998). Gene discovery in Drosophila: new insights for learning and memory. Annu. Rev. Neurosci. 21, 407-444.

Ebert, A., Schotta, G., Lein, S., Kubicek, S., Krauss, V., Jenuwein, T. and Reuter, G. (2004). Su(var) genes regulate the balance between euchromatin and heterochromatin in Drosophila. Dev. 18, 2973-2983.

Eisenberg, J. C. and Elgin, S. C. R. (2014). A structural chromosomal protein regulating transcription. Trends Genet. 30, 103-110.

Eisenberg, J. C., James, T. C., Foster-Hartnett, D. M., Hartnett, T., Ngan, V. and Elgin, S. C. (1990). Mutation in a heterochromatin-specific chromosomal protein is associated with suppression of position-effect variegation in Drosophila melanogaster. Proc. Natl Acad. Sci. USA 87, 9923-9927.

Ellis, H. M. and Horvitz, H. R. (1986). Genetic control of programmed cell death in the nematode C. elegans. Cell 44, 817-829.

Evans, T., Rosenthal, E. T., Youngblom, J., Distel, D. and Hunt, T. (1983). Cyclin: a protein specified by maternal mRNA in sea urchin eggs that is destroyed at each cleavage division. Cell 33, 389-396.

Fang, F. C. and Casadevall, A. (2010). Lost in translation--basic science in the era of translational research. Infect. Immun. 78, 563-566.

Fatehullah, A., Tan, S. H. and Barker, N. (2016). Organisms as an in vitro model of human development and disease. Nat. Cell Biol. 18, 249-254.

Fire, A., Xu, S. Q., Montgomery, M. K., Kostas, S. A., Driver, S. E. and Mello, C. C. (1998). Potent and specific genetic interference by double-stranded RNA in Caenorhabditis elegans. Nature 391, 806-811.

Forsburg, S. L. (2001). The art and design of genetic screens: yeast. Nat. Rev. Genet. 2, 659-668.

Garelli, A., Gontijo, A. M., Migueira, V., Caparros, E. and Dominguez, M. (2012). Imaginal discs secrete insulin-like peptide 8 to mediate plasticity of growth and maturation. Science 336, 579-582.

Garvik, B., Carson, M. and Hartwell, L. (1995). Single-stranded DNA arising at telomeres incdc13 mutants may constitute a specific signal for the RAD9 checkpoint. Mol. Cell. Biol. 15, 6128-6138.

Glabe, C. G. and Vacquier, V. D. (1977). Species specific agglutination of eggs by bindin isolated from sea urchin sperm. Nature 267, 836-838.

Gu, T., Orita, S. and Han, M. (1998). Caenorhabditis elegans SUR-5, a novel but conserved protein, negatively regulates LET-60 Ras activity during vulval induction. Mol. Cell. Biol. 18, 4556-4564.

Hagan, I., Hayles, J. and Nurse, P. (1988). Cloning and sequencing of the cyclin-related cdc13+ gene and a cytological study of its role in fission yeast mitosis. J. Cell Sci. 91, 587-596.

Han, M. and Sternberg, P. W. (1990). Let-60, a gene that specifies cell fates during C. elegans vulval induction, encodes a ras protein. Cell 63, 921-931.

Hand, E., Mole, B., Morello, L., Tollefson, J., Wadman, M. and Witze, A. (2013). A back seat for basic science. Nature 496, 277-279.

Hannah-Alava, A. (1958). Developmental genetics of the posterior legs in Drosophila melanogaster. Genetics 43, 879-905.

Hariharan, I. K. (2015). Organ size control: lessons from Drosophila. Dev. Cell 34, 255-265.

Hartwell, L. H., Culotti, J. and Reid, B. (1970). Genetic control of the cell-division cycle in yeast. I. Detection and regulation ofcdc mutants. Proc. Natl. Acad. Sci. USA 66, 352-359.

Hartwell, L. H., Mortimer, R. K., Culotti, J. and Culotti, M. (1973). Genetic control of the cell division cycle in yeast: V. Genetic analysis of cdc mutants. Genetics 74, 267-286.

Heasman, J. (2002). How moral oligos: making sense of antisense? Dev. Biol. 243, 209-214.

Hong, A. W., Meng, Z. and Guan, K.-L. (2016). The Hippo pathway in intestinal regeneration and disease. Nat. Rev. Gastroenterol. Hepatol. 13, 324-337.

Huch, M., Knoblich, J. A., Lutolf, M. P. and Martinez-Arias, A. (2017). The hope and the hype of organoid research. Development 144, 938-941.

Hunt, T., Luca, F. C. and Ruderman, J. V. (1992). The requirements for protein synthesis and degradation, and the control of death by cyclins A and B in the meiotic and mitotic cell cycles of the clam embryo. J. Cell Biol. 116, 707-724.
Irvine, K. D. and Harvey, K. F. (2015). Control of organ growth by patterning and hippo signaling in Drosophila. Cold Spring Harb. Perspect. Biol. 7.

James, T. C. and Elgin, S. C. (1986). Identification of a nonhistone chromosomal protein associated with heterochromatin in Drosophila melanogaster and its gene. Mol. Cell. Biol. 6, 3862-3872.

Kenyon, C. (2011). The long-lived mutants: discovery of the insulin/IGF-1 pathway for ageing. Philos. Trans. R. Soc. Lond. B Biol. Sci. 366, 9-16.

Kerr, J. F. R., Wyllie, A. H. and Currie, A. R. (1972). Apoptosis: a basic biological phenomenon with wide-ranging implications in tissue kinetics. Br. J. Cancer 26, 239-247.

Kimble, J. and Seidel, H. C. (2013). E. coli germline stem cells and their niche. In SterneBook [Internet]. Cambridge, MA: Harvard Stem Cell Institute.

Kornfeld, K. (1997). Viral development in Caenorhabditis elegans. Trends Genet. 13, 55-61.

Kravitz, E. A. and Fernandez, M. P. (2015). Aggression in Drosophila. Behav. Neurosci. 129, 549-563.

Lanzuoco, C. and Orlando, V. (2012). Memories from the polyclom group proteins. Annu. Rev. Genet. 46, 561-589.

Lee, M. G. and Nurse, P. (1987). Complementation used to clone a human homologue of the fission yeast cell cycle kinase cdc2. Nature 327, 31-35.

Levinson, A. and Shilatifard, A. (2014). The first long-lived mutants: discovery of the insulin/IGF-1 pathway for ageing.

Poltorak, A., He, X., Smirnova, I., Liu, M. Y., Van Huffel, C., Du, X., Birdwell, D., Qi, D., Jin, H., Lilja, T. and Mannervik, M. (2000). Regulation of chromatin structure by site-specific histone H3 methyltransferases. Nature 406, 593-599.

Reutel, G. and Spierer, P. (1992). Position effect variegation and chromatin proteins. BioEssays 14, 605-612.

Roth, S., Stein, D. and Nüsslein-Volhard, C. (1989). A gradient of nuclear localization of the dorsal protein determines dorsoventral pattern in the Drosophila embryo. Cell 59, 1189-1202.

Schuetzengruber, B., Chourrout, D., Vervoort, M., Leblanc, B. and Cavalli, G. (2010). Gene regulation by polycomb and trithorax proteins. Cell 142, 735-745.

Sea Urchin Genome Sequencing Consortium. Sodergren, E., Weigmann, K., Klapper, R., Strasser, T., Rickert, C., Technau, G., Ja, Z., Lenardo, M. J. and Baltimore, D. (2017). One to only two: a short history of the centrosome and its duplication. Cilia 5, 35.

Sokolova, V., Li, F., Polovin, G. and Park, S. (2015). Proteasome activation is mediated via a functional interaction of the RP1 C-terminal tail following chaperone-dependent assembly. Sci. Rep. 5, 14909.

Standart, N. and Hunt, T. (1990). Control of translation of masked mRNAs in clam oocytes. Enzyme 44, 106-119.

Standart, N., Dale, M., Steward, E. and Hunt, T. (1990). Maleal mRNA from clam oocytes can be specifically translated in vitro by antisen complementary to the 3′ untranslated region. Genes Dev. 4, 2157-2168.

Stemberg, P. W. and Han, M. (1998). Genetics of RAS signaling in C. elegans. Trends Genet. 14, 466-472.

Steward, R. (1989). Relocalization of the dorsal protein from the cytoplasm to the nucleus correlates with its function. Cell 59, 1179-1188.

Sullivan, D. T., Grillo, S. L. and Kitos, R. J. (1974). Subcellular localization of the first three enzymes of the oomochromic synthetic pathway in Drosophila melanogaster. J. Exp. Biol. 188, 225-233.

Sulston, J. E. and Horvitz, H. R. (1981). Post-embryonic cell lineages of the nematode Caenorhabditis elegans. Dev. Biol. 56, 110-156.

Sundaram, M. and Han, M. (1996). Control and integration of cell signaling pathways during C. elegans vulval development. BioEssays 18, 473-480.

Sutton, W. S. (1993). The chromosomes in heredity. Biol. Bull. 4, 231-250.

Tang, H. and Han, M. (2017). Fatty acids regulate germline sex determination through ACS4-dependent modification. Cell 169, 467-469.e13.

Timms, R. T., Tchasonovikarova, I. A. and Lehner, P. J. (2016). Position-effect variegation revisited: HUSHing up heterochromatin in human cells. BioEssays 38, 333-343.

Tischler, B., Hofmann, A., Krauss, V., Dorn, R., Korge, P. and Reuter, G. (1994). The protein encoded by the Drosophila position-effect variegation suppressor gene Su(var)3-9 combines domains of antagonistic regulators of homeotic gene complexes. EMBO J. 13, 3822-3831.

Vaccquier, V. and Mody, G. W. (1977). Isolation of bindin: the protein responsible for suspension of sperm to sea urchin eggs. Proc. Natl. Acad. Sci. USA 74, 2456-2460.

Weimann, K., Klapper, R., Strasser, T., Rickert, C., Technau, G., Jäckle, H., Janning, W. and Klämbt, C. (2003). FlyMove—a new way to look at development of Drosophila. Trends Genet. 19, 310-311.

Wightman, B., Ha, I. and Ruvkun, G. (1993). Posttranscriptional regulation of the heterochronic gene lin-14 by lin-4 mediates temporal pattern formation in C. elegans. Cell 75, 855-862.

Yuan, J., Shaham, S., Ledoux, S., Ellis, H. M. and Horvitz, H. R. (1993). The C. elegans cell death gene ced-3 encodes a protein similar to mammalian interleukin-1-converting enzyme. Cell 75, 641-652.

Zhang, Y. and Del Re, D. P. (2017). A growing role for the Hippo signaling pathway in the heart. J. Mol. Med. (Berl.) 95, 465-472.

Zhang, Q., Lenardo, M. J. and Baltimore, D. (2017). 30 years of NF-kappaB: a blossoming of relevance to human pathology. Cell 168, 37-57.
Summary: Although biologists are increasingly being tasked with problem solving rather than discovery, curiosity-driven research using model organisms has and will continue to pay dividends for improving the human condition.

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