Cell and Tissue Research (2019) 377:383–396
https://doi.org/10.1007/s00441-019-03036-4

REVIEW

The *C. elegans* intestine: organogenesis, digestion, and physiology

Ivan Dimov[1] · Morris F. Maduro[2]

Received: 27 December 2018 / Accepted: 12 April 2019 / Published online: 7 May 2019

Abstract

The comparatively simple *Caenorhabditis elegans* intestine fulfills many of the complex functions of the mammalian digestive tract, liver, and fat tissues, while also having roles in pathogen defense, immunity, and longevity. In this review, we describe the structure of the *C. elegans* gut and how it develops from the embryonic precursor E. We examine what is currently known about how the animal's microbial diet is moved through the intestinal lumen, and how its enzymatic functions contribute to physiology and metabolism. The underlying gene regulatory networks behind both development and physiology are also described. Finally, we consider recent studies that examine metabolism and digestion and describe emerging areas for future work.

Keywords Digestion · Nematode · Invertebrate · Intestine · Metabolism · *C. elegans*

Introduction

The endodermal germ layer includes the digestive tract, through which breakdown of food and absorption of nutrients and minerals occurs. Whereas the digestive tract of higher animals is subdivided into many regions, that of the nematode, *Caenorhabditis elegans*, comprises only a simple pharynx (esophagus), intestine, and hindgut (rectum). The intestine is a simple tube consisting of 20 cells that runs some 80% of the length of the animal. In addition to serving as the primary site of digestion and nutrient absorption, the intestine also fulfills the functions of the liver, while also having central roles in pathogen infection, immunity, longevity, and detoxification of metals (Block et al. 2015; Ezcurra et al. 2018; Peres et al. 2018; Pukkila-Worley and Ausubel 2012; Troemel et al. 2008). In hermaphrodites, the intestine is also the site of production of yolk lipoprotein, vitellogenin, that is secreted and taken up by developing oocytes (Grant and Hirsh 1999; Kimble and Sharrock 1983). The *C. elegans* intestine has also served as a model for the study of various human diseases, including fat storage disorders, diabetes, and metabolic syndrome (Campbell and Fares 2010; Hermann et al. 2012; Silverman et al. 2009; Zhang et al. 2011b; Zhu et al. 2016).

Unlike insect and vertebrate models, *C. elegans* lacks the capacity to replace somatic cells. The intestine must therefore serve the animal for its life span. By contrast, in *Drosophila*, the juvenile intestine is replaced by new cells during the pupal stage, while adults maintain a population of stem cells that can regenerate different intestinal cell types (Buchon and Osman 2015; Miguel-Aliaga et al. 2018). In vertebrates, intestinal crypts contain stem cells that regularly generate and shed cells from the villi lining the intestinal epithelium (Gehart and Clevers 2018; Spence et al. 2011). The lack of cellular replacement in *C. elegans* means that the gut is more prone to permanent cell damage or loss due to injury or infection, but it also means that more subtle effects resulting from experimental manipulation, or aging, are easier to discover because they persist over time.

C. elegans animals are found around the world, often on rotting fruit, where they likely consume nutrient-rich substrates and bacteria (Kiontke and Sudhaus 2006; Kiontke et al. 2011). In the laboratory, *C. elegans* are typically grown on *Escherichia coli*, a diet of ~55% protein (Brenner 1974; Yilmaz and Walhout 2016). Other bacteria used in laboratory studies include *E. coli* modified to express double-stranded RNA (dsRNA) for feeding-based RNA interference (Kamath et al. 2001), species that provide a richer food source (Laurent et al. 2001; Macneil and Walhout 2013) or pathogenic bacteria (Jiang and Wang 2018; Khan et al. 2018). The choice of food and *C. elegans* strain can have drastic effects on metabolism, gene expression, and growth phenotypes (Celen et al. 2018;

✉ Morris F. Maduro
mmaduro@ucr.edu

[1] Graduate Program in Cell, Molecular and Developmental Biology, University of California, Riverside, CA 92521, USA

[2] Department of Molecular, Cell and Systems Biology, University of California, Riverside, CA 92521, USA

Macneil and Walhout 2013; Reinke et al. 2010; Xiao et al. 2015; Zhao et al. 2018). Unlike mammals, but similar to *Drosophila*, *C. elegans* cannot synthesize cholesterol and it must be added to culture media (Hieb and Rothstein 1968; Rauthan and Pilon 2011; Stiernagle 2006; Vinci et al. 2008).

As a small metazoan model system, *C. elegans* is amenable to rapid characterization of genes by forward mutation, and reverse genetics, i.e., with CRISPR/Cas9 and RNA interference (Ahringer 2006; Jorgensen and Mango 2002; Waaijers and Boxem 2014), and its transparency enables live cell imaging (Hutter 2012). The intestine is particularly amenable to RNA interference by ingestion of dsRNA (Whangbo et al. 2017). Historically, genetic approaches identified small numbers of genes important for gut function by mutant phenotype. More recently, systems biology and "omics" work have begun to characterize the entire metabolic enzyme network and determine how the animal responds to changes in its diet. Several reviews cover various aspects of the *C. elegans* intestine in detail (Altun and Hall 2009; Kormish et al. 2010; Maduro 2017; McGhee 2007); here, we will describe *C. elegans* intestine structure and development and highlight key features of the underlying molecular genetics. We will then give an overview of recent work on intestinal cell biology as it relates to digestion, metabolism, physiology, and aging and give examples of emerging areas.

Intestine structure

The *C. elegans* digestive tract is essentially a tube (Fig. 1). At the anterior end, the self-contained pharynx, analogous to the esophagus, comprises 58 cells with its own epithelium, musculature and neurons (Mango 2009; Sulston et al. 1983). Pumping of the pharynx muscles, and peristalsis through its lumen, directs food to the intestine (Song and Avery 2013). At the posterior end of the pharynx is the grinder, a cuticular structure that mechanically breaks up food particles.

The intestine is posterior to the grinder, separated from it by the pharyngeal-intestinal valve. The intestine consists of 20 cells arranged as "rings" (Fig. 1b). At the anterior, four cells form the ring int1. The remaining 16 cells are arranged in pairs, with each pair forming rings int2 through int9. The outside surface of the intestine is surrounded by a basement membrane that faces the pseudocoelomic space (Leung et al. 1999). The intestinal basement membrane contains type IV collagen and most of the protein components found in basement membranes that surround other *C. elegans* tissues (Kramer 2005).

On the apical side of gut cells is the lumen, elliptical in cross-section, and which is lined with membranous microvilli that form the brush border (Fig. 2). The microvillar projections are supported by the terminal web, comprised primarily of an intracellular network of actin. Within the lumenal space and just outside of the microvilli, is the glycocalyx, a region rich in glycoproteins that provides a physical barrier from pathogens, serves as the interface between digestive enzymes and macromolecules, and likely allows filtering of lumen contents from entering gut cells (McGhee 2007). Besides stem cells alluded to earlier, the *C. elegans* gut lacks additional cell types such as phagocytes and gland cells that are found in other invertebrates (Karasov and Douglas 2013).

At its posterior end, the intestine is connected to the hindgut through the intestinal-rectal valve. The hindgut provides passage of waste to the environment through the anus, regulated by three muscle sets, including two muscles that wrap around the ventral posterior of the intestine (White et al. 1986). These ventral posterior muscles, and the attachment of the intestine to the pharynx and hindgut, are the only rigid connections that the intestine has to the body (Altun and Hall 2009).

Parasitic nematodes have variations of this basic structure, likely reflecting adaptations to unique environments. For example, whereas the *C. elegans* gut is cellularized, some plant and animal parasitic nematodes have a syncytial intestine (Byers and Anderson 1973; Colley 1970). Within gut cells, the terminal web is absent in plant parasitic nematodes, and may only appear in the adult stage of animal parasitic nematodes (Bruce 1966; Byers and Anderson 1973; Colley 1970). Finally, whereas the *C. elegans* intestine connects to a small number of muscles, that of the pinworm *Aspiculuris tetraptera* is surrounded by a muscle fiber network (Lee and Anya 1968).

Intestine development

The *C. elegans* gut originates as a single cell called E (for endoderm) during early embryogenesis (Sulston et al. 1983). A series of synchronous mitoses results in 16 E descendants, four of which undergo an additional division to result in 20 cells. The gut is generally invariant in cell number but occasionally there are 21 or 22 cells instead of 20 (Asan et al. 2016). Embryogenesis takes about 13 h at 25 °C (Sulston et al. 1983). Internalization of the gut occurs when the two E daughters migrate from the ventral part of the embryo into the interior (Leung et al. 1999). The gut primordium undergoes a set of carefully orchestrated cell movements and polarization to result in longitudinally arranged pairs of cells (with four at the anterior). The movements of these cells have been described in detail (Asan et al. 2016; Leung et al. 1999; Rasmussen et al. 2013). In the latter half of embryogenesis, the lumen of the intestine begins to form (Fig. 2) (Leung et al. 1999; Maduro 2017). The lumen forms first in discontinuous regions starting at the 16E stage, when gut cells begin to polarize their contents apicobasally (Leung et al. 1999). Formation of the lumen is coincident with formation of adherens junctions around the lumen and between int rings

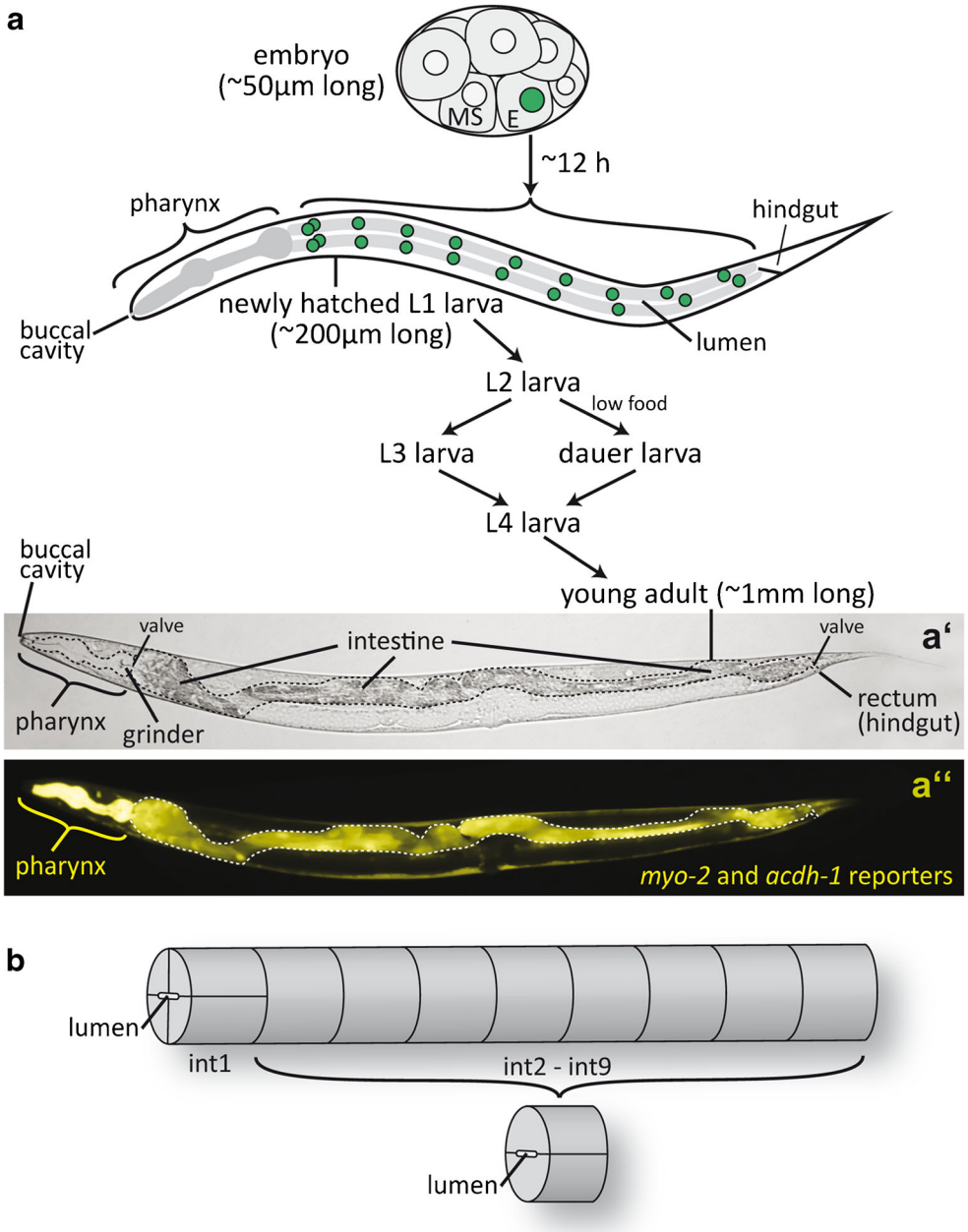

Fig. 1 Origin and basic structure of the *C. elegans* intestine. **a** The gut originates as the E blastomere in the early embryo (Sulston et al. 1983). After approximately 12 h at 25 °C, in the newly hatched first-stage juvenile (L1 larva), the intestine is found between the anterior pharynx and the posterior hindgut. Further development takes approximately 45 h to progress through the L2, L3, and L4 larval stages to adulthood. The alternative dauer larva stage occurs during conditions of low food availability and/or high population density. In the adult, the intestine (outlined by dashed black lines) is visible by the granular contents of the gut cytoplasm in a light microscope image. In the fluorescence image directly below, expression of *myo-2*::GFP and *acdh-1*::GFP transgenes mark pharynx muscle and gut nuclei/cytoplasm, respectively (MacNeil et al. 2013; Okkema et al. 1993). The gut is outlined as in the light micrograph. **b** The basic structure of the intestine as a set of rings, consisting of four cells in the anterior-most int1 ring and two cells in int2 through int9. The lumen is located apically at the interface between cells in the rings (Asan et al. 2016)

(Asan et al. 2016). The intestine is ready to function upon hatching. In the first larval stage, 14 additional nuclear divisions occur without cytokinesis, and a doubling of nuclear content accompanies each larval molt, resulting in 34 total nuclei of content 32C, in the same 20 cells that were present at hatching (Sulston et al. 1983).

The reproducible number of cells and nuclei in the *C. elegans* intestine suggests there has been strong selection for its maintenance. However, animals can accommodate slightly fewer numbers of intestinal E descendants or an excess with no apparent consequences to organogenesis and intestine function, at least in the laboratory (Choi et al. 2017; Clucas et al. 2002; Kostic and Roy 2002; Lee et al. 2016). For example, in mutant backgrounds that result in greater than 30 gut cells rather than the typical 20, a functional intestine still forms (Choi et al. 2017). Therefore, morphogenesis of the intestine does not require a fixed number of cells, and the nearly invariant number of 20 cells may represent a trade-off between resource allocation and maintaining enough cells to support integrity and flexibility of the lumen (Asan et al. 2016).

Genetics of gut development

Gut formation begins with specification of the E blastomere as the gut progenitor. An embryo in which E is not specified

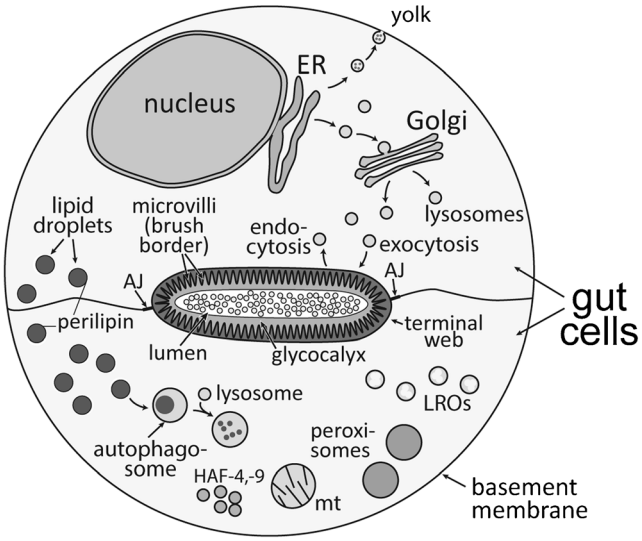

Fig. 2 The active cytoplasm of *C. elegans* enterocytes. A pair of gut cells, part of the same int ring, are diagrammed shown as a transverse section. Cellular contents are shown as examples of the diverse structures and processes that are found in gut cells as cited in the text. Sizes of structures are only approximate. Abbreviations: AJ, adherens junction; ER, endoplasmic reticulum; LRO, lysosome-related organelle (gut granule)

develops into an arrested larva, somewhat shorter than the wild type, and missing all intestinal tissue (Owraghi et al. 2010). As expected, the resulting larvae are inviable, presumably because they have no means by which to take in nutrients.

Specification of E occurs through the activation of a gene regulatory network of maternal and zygotic transcription factors that are expressed in a temporal sequence (Fig. 3). The earliest-acting factors that specify E are the paralogous *end-1* and *end-3* genes, which encode structurally similar GATA type transcription factors that are partially redundant (Maduro et al. 2005; Zhu et al. 1997). Upstream of the *end* genes, specification of E begins when the four-cell stage blastomere P_2 contacts the mother of the E cell, called EMS (Goldstein 1992). After EMS divides, the side of EMS in contact with P_2, its posterior side in normal embryos, becomes E (Goldstein 1993). The gene network that specifies gut was deduced through a combination of genetic and in vivo and in vitro DNA binding studies (Maduro 2017). Induction requires overlapping Wnt/MAPK/Src signaling that produces an endoderm-promoting state of the Wnt nuclear effector TCF/POP-1 in E (Rocheleau et al. 1997; Shetty et al. 2005; Shin et al. 1999; Thorpe et al. 2000). In this state, POP-1 interacts with the divergent β-catenin SYS-1 to become a weak transcriptional activator of the gene *end-1*. In parallel with the Wnt/β-catenin asymmetry pathway, maternally provided SKN-1, a bZIP/homeodomain-like transcription factor, activates expression of the *med-1* and *med-2* genes (Maduro et al. 2001). These encode divergent GATA-type transcription factors that together contribute to activation of *end-1* and *end-3* (Broitman-Maduro et al. 2005). The Sp1-like factor SPTF-3 also provides input into the *end* genes (Sullivan-Brown et al. 2016).

The result of these many inputs is to cause timely and robust activation of the *elt-2* GATA factor gene, which is both necessary and sufficient to specify and maintain intestinal cell identity (Fukushige et al. 1998; McGhee et al. 2009). The paralogous *elt-7* gene is also activated in the early E lineage and is expressed in parallel with *elt-2*, and the two factors have overlapping functions (Sullivan-Brown et al. 2016). When *elt-2* is absent, gut is still specified in the embryo, but animals arrest as larvae with abnormal intestines (Fukushige et al. 1998). Consistent with overlapping function of *elt-2* and *elt-7*, this phenotype is exacerbated when *elt-7* is also absent, even though loss of *elt-7* alone has no apparent phenotype (Sommermann et al. 2010). Hence, gut specification in *C. elegans* represents the collaborative input of multiple redundant factors working together to assure robust gut development (Choi et al. 2017).

Intestinal specification by an ELT-2-like factor appears to be widely conserved among nematodes. The parasitic nematode *Haemonchus contortus*, also known as the barber's pole worm, has an intestinal ELT-2-like protein that can cause development of intestinal tissue when its coding region is overexpressed in *C. elegans* (Couthier et al. 2004). The genome of the parasite *Ascaris suum* also encodes a putative ELT-2-like factor (Rosa et al. 2014). Putative ELT-2 orthologs are also found among *Pristionchus*, *Brugia*, and *Onchocera* species as listed in WormBase (http://www.wormbase.org). Direct evidence that an ELT-2-like factor regulates intestinal fate in these other species has not yet been obtained.

Upstream of ELT-2, the MED and END genes have orthologs only in more closely related *Caenorhabditis* species (Coroian et al. 2005; Gillis et al. 2007; Maduro et al. 2005). These are absent in more distant *Caenorhabditis* species, suggesting that some other mechanism must initiate gut specification (Maduro 2017). It is possible that activation of *elt-2* in E could specify gut directly, as overexpressed ELT-2 can achieve gut specification in *C. elegans* in the absence of *end-1* and *end-3* (Wiesenfahrt et al. 2015). Even among *Caenorhabditis* species that have MED and END orthologs, however, the upstream factors in the gut specification network may regulate gut specification differently. For example, in *C. elegans*, loss of *pop-1* function results in an "extra endoderm" phenotype in which another cell, MS, makes gut as well as E (Lin et al. 1995). In *C. briggsae*, however, loss of the *pop-1* ortholog results in the loss of endoderm specification (Lin et al. 2009). These differences illustrate the broader phenomenon of developmental system drift, common to such regulatory networks (Haag et al. 2018; True and Haag 2001).

The use of GATA transcription factors in specification and development of endodermal tissues is evolutionarily shared among protostomes and deuterostomes. For example, in

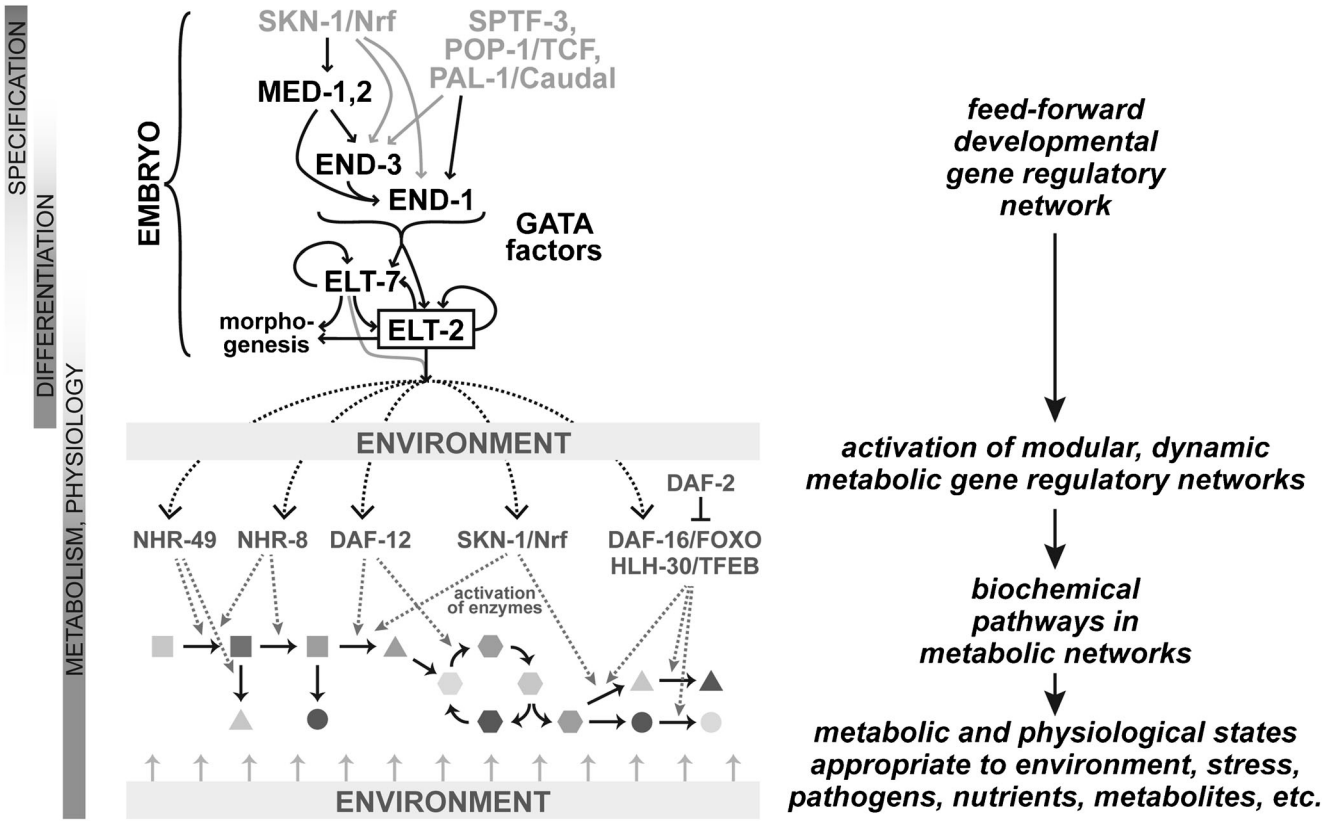

Fig. 3 Integration of a developmental gene regulatory network with metabolic networks that modulate physiological responses to the environment. Environmental influences on gene expression, enzyme activity, and metabolic flux include temperature, oxygen levels, metabolites, ascarosides, and pathogens. Example regulators are shown activating shared and distinct target genes encoding enzymes that function at different steps in biochemical pathways. This figure was drawn by combining and modifying similar figures in other reviews (Maduro 2017; Watson and Walhout 2014)

Drosophila, gut specification involves the GATA factors Serpent and dGATAe (Murakami et al. 2005; Okumura et al. 2005), while among vertebrates, the GATA4/5/6 factors are important for development of endoderm and other tissues (Lentjes et al. 2016; Zorn and Wells 2009).

Assembly of the gut primordium results from the orientation of cell divisions that occur from one round of mitosis to the next within the E lineage, enforced by cell-cell signaling using the Van Gogh/VANG-1 pathway that keeps the primordial cells in a stereotypical alignment (Asan et al. 2016; Leung et al. 1999). Between the stages at which there are 8 E descendants and 20 descendants, specific pairs of cells undergo migrations and reorientations within the primordium, guided by signaling of the Ephrin, Notch, and Netrin pathways (Asan et al. 2016).

Formation of the gut lumen occurs by the apical targeting of components that support the terminal web, but the full details of how the microvilli become assembled are not yet understood. The Ezrin/Radixin/Moesin membrane-cytoskeleton linker protein, ERM-1, and the actin ACT-5 are two essential components that support the cytoskeleton supporting lumenal microvilli (Gobel et al. 2004; MacQueen et al. 2005). Apicobasal polarity of developing gut cells, and proper endosomal trafficking, are required for proper lumen formation. Loss of the Clathrin *chc-1* results in mislocalization of these proteins and a disruption in lumen structure (Zhang et al. 2012). Loss of some enzymes in lipid biosynthesis, such as the sphingolipid biosynthetic enzyme LET-767, also disrupts apical localization of lumen components and hence lumen structure (Zhang et al. 2011a).

Digestion: physiology and metabolism

Digestion begins with mechanical and enzymatic breakage of the cell walls and membranes of microbes in the pharynx grinder (McGhee 2007). Partially broken down food that enters the intestine lumen gets exposed to enzymes that break down membranes and their lipid constituents, such as lysozymes, saponins, and lectins, enabling access to the cellular contents and also serving as a defense against microbes (Gravato-Nobre et al. 2016; Kandasamy et al. 2012; Mallo et al. 2002; Tarr 2012). Experimental details about the subcellular compartmentalization of digestion in *C. elegans* are somewhat lacking, perhaps owing to a primary focus of most work on establishing cause-and-effect genetic relationships,

coupled with optical challenges in determining protein localization due to the high autofluorescence of the gut. However, as fundamental digestive mechanisms are conserved, it is likely that in the *C. elegans* gut, macromolecules are partially hydrolyzed in the lumen prior to endocytosis (i.e., extracellular digestion), after which further breakdown occurs when endocytic vesicles internalize lumen contents and fuse with acidified lysosomes inside gut cells (intracellular digestion) (Karasov and Douglas 2013; McGhee 2007).

As would be expected, many digestive enzymes are expressed in the *C. elegans* intestine, the most abundant of which are proteases (McGhee et al. 2007). Examples of proteases include astacins (NAS), metalloproteases that are found within the digestive tract (Mohrlen et al. 2003); the aspartic protease ASP-1, found in both the lumen and intracellular lysosomes (Tcherepanova et al. 2000); and aminopeptidase P which likely functions in intracellular hydrolysis (Laurent et al. 2001). Enzymes for other macromolecules include an amylase-like enzyme, *C50B6.7*, that is orthologous to human amylases, and which is expressed in the intestine (McGhee et al. 2007; Mulder et al. 2003), and the deoxyribonuclease (DNase) NUC-1 (Hevelone and Hartman 1988). For information on digestion and metabolism of other macromolecules, the reader is referred to comprehensive reviews on lipid and carbohydrate metabolism and intermediary metabolism (Braeckman et al. 2009; Lemieux and Ashrafi 2015; Watts and Ristow 2017).

Among nematodes, core metabolic pathways are generally conserved, with missing or additional pathways apparent particularly in parasitic species, as predicted by genome sequence comparisons (Martin et al. 2012; Wylie et al. 2008; Yin et al. 2008). Recent genomics approaches have been used to infer the structure of the metabolome by identifying orthologous genes that encode enzymes and integrating information from metabolic pathway databases and published literature (Gebauer et al. 2016; Witting et al. 2018; Yilmaz and Walhout 2016). These types of studies, confirm that both intracellular and extracellular digestion occur in *C. elegans*. In one recent work, over 600 metabolic enzymes in almost 2000 reactions were identified and inferred to occur in the cytosol, mitochondria, or extracellularly based on prior work (Yilmaz and Walhout 2016). The resulting network, called iCEL1273, also incorporates transport proteins, supported in part by published literature. The resulting model can be used to integrate phenotype and genotype data to predict the biochemical flux of nutrients into *C. elegans* biomass and waste products (Yilmaz and Walhout 2016).

One class of biomolecules deserves special mention. Unlike DNA, dsRNA escapes digestion into monomers in *C. elegans*. The gut can internalize dsRNA through the lumen, resulting in systemic gene silencing by RNA interference (Timmons and Fire 1998). dsRNA is specifically imported into intestinal cells from the lumen by the transporter SID-2 (Winston et al. 2007). This is likely to be an adaptive form of immunity against RNA viruses, as *C. elegans* strains with defects in the RNAi pathway show increased susceptibility to virus replication (Ashe et al. 2013; Gammon et al. 2017; Lu et al. 2005). Susceptibility to RNAi by dsRNA ingestion varies across species related to *C. elegans* (Nuez and Felix 2012).

Dynamic acidification in the lumen during digestion

C. elegans is essentially a filter feeder (Seymour et al. 1983). Food transits the digestive tract in as little as a few minutes, suggesting it is adapted for the animal's short life cycle and simple diet (Ghafouri and McGhee 2007). Enzymatic breakdown of macromolecules in animal digestive tracts is generally associated with a low-pH compartment. Recent work has elucidated the details of acidification of the intestinal lumen in *C. elegans*. The intestinal lumen is weakly acidic on average, around pH 4.4 (Allman et al. 2009). A region of higher acidity, 1–2 pH units lower, starts in the posterior third of the gut lumen, gets translocated to the very anterior over several seconds, and remains for several seconds more before raising in pH (Bender et al. 2013). During this time, the posterior lumen reacidifies slowly over 30 s, and the cycle repeats every 45 s (Allman et al. 2009; Bender et al. 2013). This wave of protons is tied to the defecation motor program (DMP), a set of regular body muscle contractions that propels food through the intestine posteriorly and out the anus (Thomas 1990). Movement of the acidic region in the opposite direction results from the dynamic activity of proton pumps along the lumen (Bender et al. 2013). The DMP itself is regulated by the propagation of a wave of intestinal calcium ions generated in gut cells (Nehrke et al. 2008; Teramoto and Iwasaki 2006). Consistent with a role for the defecation cycle in nutrient uptake, the DMP is required for the internalization of fatty acids from the lumen into gut cells (Sheng et al. 2015). Hence, *C. elegans* digestion relies on coordinated and rhythmical physiological changes along the digestive tract that produce cyclical compartments of high acidity.

Regulation of metabolism by transcription factor and signaling networks

The *C. elegans* intestine achieves regulation of catabolic and anabolic processes through multiple pathways that integrate response to nutrients and environmental conditions at multiple levels (Fig. 3). Transcription factors and regulatory pathways, many of which share conservation with similar pathways in humans, have been identified that modulate changes in biochemical flux and intestinal physiology (Watson and Walhout

2014). The SKN-1/Nrf factor, important for the early initial specification of the E fate in embryos, is active in the adult gut as a major regulator of the response to oxidative stress (An and Blackwell 2003). The nuclear hormone receptor (NHR) family of factors is highly amplified in *C. elegans* and many of these are known to regulate metabolic networks in the intestine (Arda et al. 2010; Watson and Walhout 2014). These include NHR-49, a central regulator of the *C. elegans* stress response, and the breakdown of fats by beta-oxidation (Goh et al. 2018; Hu et al. 2018; Van Gilst et al. 2005); NHR-8, which regulates cholesterol and bile acid metabolism (Magner et al. 2013); and the NHR DAF-12, which regulates enzymes in catabolism including those in the citric acid cycle (Hochbaum et al. 2011; McCormick et al. 2011; Watson and Walhout 2014). The mediator subunit MDT-15 and the sterol regulatory element-binding protein SBP-1 work with NHR-49 in regulation of fat accumulation, and MDT-15 itself has additional roles in response to oxidative stress (Goh et al. 2014; Moreno-Arriola et al. 2016; Taubert et al. 2006).

The environment also modulates metabolic functions during development. Young *C. elegans* larvae, exposed to conditions of limiting food or high population density, enter an alternative third larval stage known as the dauer larva (Fig. 1). In this stage, metabolism and physiology are altered, and development paused, until food becomes available (Androwski et al. 2017). A central component of both dauer formation and metabolism is Insulin/IGF-1 signaling, or IIS (Murphy and Hu 2013). Mutation of the gene encoding the insulin receptor DAF-2 results in constitutive dauer formation at higher temperatures (Kimura et al. 1997). In adults, mutation of *daf-2* extends lifespan and increases stress resistance, connecting metabolic changes occurring in dauer larvae and in adults (Kenyon et al. 1993). DAF-2 functions through a conserved phosphatidylinositol-3-kinase/protein kinase B/Target of Rapamycin (PI3K/Akt/TOR) pathway (Chen et al. 2013; Paradis and Ruvkun 1998). The life extension mediated by loss of *daf-2* depends primarily on the activity of the conserved FOXO transcription factor DAF-16 (Lin et al. 2001). DAF-16/FOXO and the helix-loop-helix factor HLH-30/TFEB both translocate to intestinal nuclei to regulate genes that promote longevity and stress resistance (Lin et al. 2018). Insulin-like peptides (ILPs), of which 40 are encoded in the *C. elegans* genome, are part of a dynamic signaling system involving DAF-16/FOXO that maintains metabolic homeostasis depending on nutrient availability (Kaplan et al. 2018). ILPs mediate communication between neurons and the intestine, explaining how nutritional signals in the animal affect metabolism in the gut (Hung et al. 2014).

Intestinal endosomes and storage granules

Different types of granules or membrane-bound storage organelles have been identified in *C. elegans* enterocytes as shown diagrammatically in Fig. 2. The most visible of these are gut granules, which contain birefringent material that is visible under polarized light (Hermann et al. 2005). Gut granules are membrane-bound, lysosome-related organelles (LROs) enriched in various substances including zinc (Coburn and Gems 2013; Hermann et al. 2012; Roh et al. 2012). The granules also fluoresce blue under UV light due to the presence of glycosylated anthranilic acid (Coburn et al. 2013). Gut cells also contain conventional acidified lysosomes that are enriched in chloride and calcium ions (Chakraborty et al. 2017; Narayanaswamy et al. 2018). Lipid droplets are a major site of fat storage that can be detected by different methods, including staining with Oil Red O and dark field microscopy (Fouad et al. 2017; Lapierre et al. 2013; O'Rourke et al. 2009). As in other animals including humans, lipid droplets are surrounded by perilipin, a lipid storage regulatory protein that recognizes the phospholipid monolayer surrounding the droplet and regulates lipid metabolism (Beller et al. 2010; Chughtai et al. 2015; Copic et al. 2018; Hashani et al. 2018). Gut cells also contain peroxisomes, which serve important functions in fatty acid metabolism (Artyukhin et al. 2018; Yokota et al. 2002).

Because of the role of endosomes in digestion, the *C. elegans* gut is also a model for the study of endosomal trafficking, which relies on conserved endosomal proteins such as the GTPases RAB-5 and RAB-10 (Chen et al. 2006; Liu et al. 2018; Sato et al. 2014). Maintenance of the lumen over time also relies on proper apical targeting of endosomes, which relies on the conserved adapter proteins Clathrin and AP1 (Zhang et al. 2012). Enterocytes in hermaphrodites contain vesicles carrying yolk lipoprotein from the endoplasmic reticulum to the basement membrane (Grant and Hirsh 1999). Finally, a novel endosome type of unknown function has been recently identified that is distinct from LROs, lipid droplets, and vesicles carrying yolk and is associated with the ABC transporters HAF-4 and HAF-9 (Tanji et al. 2016).

Dietary restriction, lipid storage, autophagy, and life span

In *C. elegans*, a complex relationship exists among diet, mobilization or storage of lipids, autophagy, and life span (Lapierre et al. 2011; Lemieux and Ashrafi 2015; Seah et al. 2016). Dietary (caloric) restriction (DR) can be achieved in *C. elegans* by diluting *E. coli* cultures, growing worms in liquid culture, or using an *eat-2* mutant which has decreased pharyngeal pumping (Cypser et al. 2013; Gelino et al. 2016; Lenaerts et al. 2008; Palgunow et al. 2012). DR during the larval stages slows development and extends post-reproductive lifespan (Palgunow et al. 2012). The intestine and epidermis (hypodermis) are major storage sites for lipid droplets, and animals undergoing DR show an apparent

increase in their number (Palgunow et al. 2012; Zhang et al. 2010). Loss of germline stem cells also increases life span, and this increased longevity correlates with increased lipids in the intestine (Hsin and Kenyon 1999; Wang et al. 2008). Consistent with this, DR induces fatty acid synthesis gene expression in a manner dependent on DAF-16/FOXO (Amrit et al. 2016), and in response, fat catabolism genes are activated by NHR-49 and SKN-1 (Ratnappan et al. 2014; Steinbaugh et al. 2015). Autophagy, which delivers lipid droplets to lysosomes for lipophagy in the intestine, is a necessary component of this fat breakdown (Lapierre et al. 2011). Autophagy also mediates the life-span extension seen in animals undergoing dietary restriction (Gelino et al. 2016; Jia and Levine 2007).

Recent studies point to a causative role of yolk lipoprotein production in the regulation of autophagy and remodeling of lipids, which is proposed to promote long lifespan. In animals undergoing dietary restriction, longevity results from decreased expression of vitellogenin genes, and activation of DAF-16/FOXO (Seah et al. 2016). In germline-less animals, the retention of yolk causes activation of SKN-1, which in turn promotes remodeling of lipids to maintain lipid homeostasis (Steinbaugh et al. 2015). As animals age, the intestine undergoes progressive detrimental changes (McGee et al. 2011). Recently, these aging-related pathologies have been proposed to result from "run-on" vitellogenesis that consumes intestine cell contents, from within, by autophagy (Ezcurra et al. 2018). In this model, the conversion of intestinal biomass into yolk into late adulthood results in the reduction of volume of intestinal cells and widening of the lumen (both of which are seen in senescing adults) and extracellular buildup of yolk pools. Interestingly, autophagy has a role in promoting senescence by converting gut cell biomass into yolk; hence, the role of intestinal autophagy in life span depends on the role it plays in promoting lipoprotein synthesis or lipid breakdown (Ezcurra et al. 2018; Gelino et al. 2016). The complex interplay among different mechanisms for metabolic homeostasis continues to be an active area of study.

Future directions

The *C. elegans* gut continues to be an important system for study. The field now has access to systems-level descriptions of how the intestine functions in time and space at a finely detailed level, incorporating environmental signals, biochemical pathways, and gene regulation (Fig. 3). Some emerging areas, which we did not have time to discuss in detail above, include the following:

Transgenerational inheritance Transgenerational epigenetic inheritance is observed by changes in gene expression when prior generations are exposed to dietary extremes such as starvation or high-glucose diets (Rechavi et al. 2014; Tauffenberger and Parker 2014). These necessarily involve transmission of information from the intestine to the germline, and understanding this process may shed light on mechanisms that may mediate human epigenetic inheritance influenced by diet (Kadayifci et al. 2018).

Metabolic network models As alluded to above, recent studies have systematically identified all metabolic enzymes encoded in the *C. elegans* genome to generate metabolic network models (Gebauer et al. 2016; Witting et al. 2018; Yilmaz and Walhout 2016). Coupled with metabolomics analyses, these models are a computational framework for predicting changes in metabolic flux that can be validated or tested experimentally, for example, by whole-animal metabolomics studies (Witting et al. 2018). Future studies that could integrate high-resolution protein localization studies may add new insights into subcellular compartmentalization of digestive functions in this species.

The microbiome Recent work has begun to analyze the microbiota found in the *C. elegans* natural environment, with a view toward analyzing host-microbe interactions (Dirksen et al. 2016; Gerbaba et al. 2017; Tan and Shapira 2011; Zhang et al. 2017a). Future studies could reveal many new insights into interplay among environmental metabolites, metabolic flux, cellular physiology, and immunity.

Ascarosides as signaling molecules Ascarosides are a nematode-specific group of diverse glycolipids based on the sugar ascarylose, and they are known to modulate behavioral, developmental and environmental responses (von Reuss 2018). These are synthesized by enzymes that also function in fat breakdown in the intestine (Panda et al. 2017). The relationship of *C. elegans* metabolism to ascaroside synthesis and the effects of some 100 known ascarosides on physiology are areas of active study (von Reuss and Schroeder 2015; Zhang et al. 2017b; Zhou et al. 2018).

The glycocalyx Although the glycocalyx is the main interface between the lumen and the apical surface of intestine cells, the molecular biology of its structure and function is not well understood (McGhee 2007). Development of new methods to study the glycocalyx may be able to reinvigorate interest in the digestive processes that occur within the lumen, particularly now that the dynamics of lumen acidification are understood (Bender et al. 2013).

Caenorhabditis inopinata*, a sister species of *C. elegans The newly described species *Caenorhabditis inopinata*, genetically a close relative of *C. elegans*, spends most of its life cycle in the interior of the fruit of the fig *Ficus septica* and disperses through its pollinating wasp *Ceratosolen* (Woodruff and

Phillips 2018). Because of these differences in life histories between *C. elegans* and *C. inopinata*, a study of the intestine and digestion between the two is likely to reveal key evolutionary adaptations of the latter species to a very different environment than that of *C. elegans*. The genome of *C. inopinata* has been sequenced, and transgenic and RNAi methods similar to those of *C. elegans* have been developed, which should enable functional genetic studies between the two species (Kanzaki et al. 2018).

Conclusion

The *C. elegans* intestine continues to be a valuable model in which to study organ structure and function, from small-scale studies investigating small numbers of genes, to systems biology approaches integrating metabolomics, genetics, behavior, and environment. With so many dimensions of *C. elegans* biology now being actively studied, future studies are likely to continue to reveal new insights about intestine development and function that are relevant across animals.

Acknowledgments We apologize to the many colleagues whose work we could not cite for space reasons. The light and fluorescence microscopy images in Fig. 1 were provided by Gina Broitman-Maduro.

Funding Work done in the Maduro lab that was cited here was funded by NSF Grant IOS No. 1258054.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

Informed consent Not applicable.

References

Ahringer J 2006. Reverse genetics (April 6, 2006). In: T.C.e.R. Community (ed.) WormBook

Allman E, Johnson D, Nehrke K (2009) Loss of the apical V-ATPase a-subunit VHA-6 prevents acidification of the intestinal lumen during a rhythmic behavior in C. elegans. Am J Phys Cell Phys 297: C1071–C1081

Altun ZF, Hall DH 2009. Alimentary system, intestine. WormAtlas

Amrit FR, Steenkiste EM, Ratnappan R, Chen SW, McClendon TB, Kostka D, Yanowitz J, Olsen CP, Ghazi A (2016) DAF-16 and TCER-1 facilitate adaptation to germline loss by restoring lipid homeostasis and repressing reproductive physiology in C. elegans. PLoS Genet 12:e1005788

An JH, Blackwell TK (2003) SKN-1 links C. elegans mesendodermal specification to a conserved oxidative stress response. Genes Dev 17:1882–1893

Androwski RJ, Flatt KM, Schroeder NE, 2017. Phenotypic plasticity and remodeling in the stress-induced *Caenorhabditis elegans* dauer Wiley Interdiscip Rev Dev Biol 6

Arda HE, Taubert S, MacNeil LT, Conine CC, Tsuda B, Van Gilst M, Sequerra R, Doucette-Stamm L, Yamamoto KR, Walhout AJ (2010) Functional modularity of nuclear hormone receptors in a Caenorhabditis elegans metabolic gene regulatory network. Mol Syst Biol 6:367

Artyukhin AB, Zhang YK, Akagi AE, Panda O, Sternberg PW, Schroeder FC (2018) Metabolomic "dark matter" dependent on peroxisomal beta-oxidation in Caenorhabditis elegans. J Am Chem Soc 140: 2841–2852

Asan A, Raiders SA, Priess JR (2016) Morphogenesis of the C elegans intestine involves axon guidance genes. PLoS Genet 12:e1005950

Ashe A, Belicard T, Le Pen J, Sarkies P, Frezal L, Lehrbach NJ, Felix MA, Miska EA (2013) A deletion polymorphism in the Caenorhabditis elegans RIG-I homolog disables viral RNA dicing and antiviral immunity. Elife 2:e00994

Beller M, Bulankina AV, Hsiao HH, Urlaub H, Jackle H, Kuhnlein RP (2010) PERILIPIN-dependent control of lipid droplet structure and fat storage in Drosophila. Cell Metab 12:521–532

Bender A, Woydziak ZR, Fu L, Branden M, Zhou Z, Ackley BD, Peterson BR (2013) Novel acid-activated fluorophores reveal a dynamic wave of protons in the intestine of Caenorhabditis elegans. ACS Chem Biol 8:636–642

Block DH, Twumasi-Boateng K, Kang HS, Carlisle JA, Hanganu A, Lai TY, Shapira M (2015) The developmental intestinal regulator ELT-2 controls p38-dependent immune responses in Adult C. elegans. PLoS Genet 11:e1005265

Braeckman BP, Houthoofd K, Vanfleteren JR, 2009. Intermediary metabolism. WormBook, 1–24

Brenner S (1974) The genetics of Caenorhabditis elegans. Genetics 77: 71–94

Broitman-Maduro G, Maduro MF, Rothman JH (2005) The noncanonical binding site of the MED-1 GATA factor defines differentially regulated target genes in the C. elegans mesendoderm. Dev Cell 8:427–433

Bruce RG (1966) The fine structure of the intestine and hind gut of the larva of Trichinella spiralis. Parasitology 56:359–365

Buchon N, Osman D (2015) All for one and one for all: regionalization of the Drosophila intestine. Insect Biochem Mol Biol 67:2–8

Byers JR, Anderson RV (1973) Morphology and ultrastructure of the intestine in a plant-parasitic nematode, Tylenchorhynchus dubius. J Nematol 5:28–37

Campbell EM, Fares H (2010) Roles of CUP-5, the Caenorhabditis elegans orthologue of human TRPML1, in lysosome and gut granule biogenesis. BMC Cell Biol 11:40

Celen I, Doh JH, Sabanayagam CR (2018) Effects of liquid cultivation on gene expression and phenotype of C elegans. BMC Genomics 19: 562

Chakraborty K, Leung K, Krishnan Y (2017) High lumenal chloride in the lysosome is critical for lysosome function. Elife 6:e28862

Chen CC, Schweinsberg PJ, Vashist S, Mareiniss DP, Lambie EJ, Grant BD (2006) RAB-10 is required for endocytic recycling in the Caenorhabditis elegans intestine. Mol Biol Cell 17:1286–1297

Chen D, Li PW, Goldstein BA, Cai W, Thomas EL, Chen F, Hubbard AE, Melov S, Kapahi P (2013) Germline signaling mediates the synergistically prolonged longevity produced by double mutations in daf-2 and rsks-1 in C. elegans. Cell Rep 5:1600–1610

Choi H, Broitman-Maduro G, Maduro MF (2017) Partially compromised specification causes stochastic effects on gut development in C. elegans. Dev Biol 427:49–60

Chughtai AA, Kassak F, Kostrouchova M, Novotny JP, Krause MW, Saudek V, Kostrouch Z (2015) Perilipin-related protein regulates lipid metabolism in C elegans. PeerJ 3:e1213

Clucas C, Cabello J, Bussing I, Schnabel R, Johnstone IL (2002) Oncogenic potential of a C. elegans cdc25 gene is demonstrated by a gain-of-function allele. EMBO J 21:665–674

Coburn C, Allman E, Mahanti P, Benedetto A, Cabreiro F, Pincus Z, Matthijssens F, Araiz C, Mandel A, Vlachos M, Edwards SA, Fischer G, Davidson A, Pryor RE, Stevens A, Slack FJ, Tavernarakis N, Braeckman BP, Schroeder FC, Nehrke K, Gems D (2013) Anthranilate fluorescence marks a calcium-propagated necrotic wave that promotes organismal death in C elegans. PLoS Biol 11:e1001613

Coburn C, Gems D (2013) The mysterious case of the C. elegans gut granule: death fluorescence, anthranilic acid and the kynurenine pathway. Front Genet 4:151

Colley FC (1970) Strongyloides myopotomi: fine structure of the body wall and alimentary tract of the adult and third-stage larva. Exp Parasitol 28:420–434

Copic A, Antoine-Bally S, Gimenez-Andres M, La Torre Garay C, Antonny B, Manni MM, Pagnotta S, Guihot J, Jackson CL (2018) A giant amphipathic helix from a perilipin that is adapted for coating lipid droplets. Nat Commun 9:1332

Coroian C, Broitman-Maduro G, Maduro MF (2005) Med-type GATA factors and the evolution of mesendoderm specification in nematodes. Dev Biol 289:444–455

Couthier A, Smith J, McGarr P, Craig B, Gilleard JS (2004) Ectopic expression of a Haemonchus contortus GATA transcription factor in Caenorhabditis elegans reveals conserved function in spite of extensive sequence divergence. Mol Biochem Parasitol 133:241–253

Cypser JR, Kitzenberg D, Park SK (2013) Dietary restriction in C. elegans: recent advances. Exp Gerontol 48:1014–1017

Dirksen P, Marsh SA, Braker I, Heitland N, Wagner S, Nakad R, Mader S, Petersen C, Kowallik V, Rosenstiel P, Felix MA, Schulenburg H (2016) The native microbiome of the nematode Caenorhabditis elegans: gateway to a new host-microbiome model. BMC Biol 14:38

Ezcurra M, Benedetto A, Sornda T, Gilliat AF, Au C, Zhang Q, van Schelt S, Petrache AL, Wang H, Guardia Y, Bar-Nun S, Tyler E, Wakelam MJ, Gems D (2018) C elegans eats its own intestine to make yolk leading to multiple senescent pathologies. Curr Biol 28:3352

Fouad AD, Pu SH, Teng S, Mark JR, Fu M, Zhang K, Huang J, Raizen DM, Fang-Yen C (2017) Quantitative assessment of fat levels in Caenorhabditis elegans using dark field microscopy. G3 (Bethesda) 7:1811–1818

Fukushige T, Hawkins MG, McGhee JD (1998) The GATA-factor elt-2 is essential for formation of the Caenorhabditis elegans intestine. Dev Biol 198:286–302

Gammon DB, Ishidate T, Li L, Gu W, Silverman N, Mello CC (2017) The antiviral RNA interference response provides resistance to lethal arbovirus infection and vertical transmission in Caenorhabditis elegans. Curr Biol 27:795–806

Gebauer J, Gentsch C, Mansfeld J, Schmeisser K, Waschina S, Brandes S, Klimmasch L, Zamboni N, Zarse K, Schuster S, Ristow M, Schauble S, Kaleta C (2016) A genome-scale database and reconstruction of Caenorhabditis elegans metabolism. Cell Syst 2:312–322

Gehart H, Clevers H (2018) Tales from the crypt: new insights into intestinal stem cells. Nat Rev Gastroenterol Hepatol 16:19–34

Gelino S, Chang JT, Kumsta C, She X, Davis A, Nguyen C, Panowski S, Hansen M (2016) Intestinal autophagy improves healthspan and longevity in C elegans during dietary restriction. PLoS Genet 12:e1006135

Gerbaba TK, Green-Harrison L, Buret AG (2017) Modeling host-microbiome interactions in Caenorhabditis elegans. J Nematol 49:348–356

Ghafouri S, McGhee JD (2007) Bacterial residence time in the intestine of Caenorhabditis elegans. Nematology 9:87–91

Gillis WJ, Bowerman B, Schneider SQ (2007) Ectoderm- and endomesoderm-specific GATA transcription factors in the marine annelid Platynereis dumerilli. Evol Dev 9:39–50

Gobel V, Barrett PL, Hall DH, Fleming JT (2004) Lumen morphogenesis in C. elegans requires the membrane-cytoskeleton linker erm-1. Dev Cell 6:865–873

Goh GY, Martelli KL, Parhar KS, Kwong AW, Wong MA, Mah A, Hou NS, Taubert S (2014) The conserved mediator subunit MDT-15 is required for oxidative stress responses in Caenorhabditis elegans. Aging Cell 13:70–79

Goh GYS, Winter JJ, Bhanshali F, Doering KRS, Lai R, Lee K, Veal EA, Taubert S (2018) NHR-49/HNF4 integrates regulation of fatty acid metabolism with a protective transcriptional response to oxidative stress and fasting. Aging Cell 17:e12743

Goldstein B (1992) Induction of gut in Caenorhabditis elegans embryos. Nature 357:255–257

Goldstein B (1993) Establishment of gut fate in the E lineage of C. elegans: the roles of lineage-dependent mechanisms and cell interactions. Development 118:1267–1277

Grant B, Hirsh D (1999) Receptor-mediated endocytosis in the Caenorhabditis elegans oocyte. Mol Biol Cell 10:4311–4326

Gravato-Nobre MJ, Vaz F, Filipe S, Chalmers R, Hodgkin J (2016) The invertebrate lysozyme effector ILYS-3 is systemically activated in response to danger signals and confers antimicrobial protection in C. elegans. PLoS Pathog 12:e1005826

Haag ES, Fitch DHA, Delattre M (2018) From "the worm" to "the worms" and back again: the evolutionary developmental biology of nematodes. Genetics 210:397–433

Hashani M, Witzel HR, Pawella LM, Lehmann-Koch J, Schumacher J, Mechtersheimer G, Schnolzer M, Schirmacher P, Roth W, Straub BK (2018) Widespread expression of perilipin 5 in normal human tissues and in diseases is restricted to distinct lipid droplet subpopulations. Cell Tissue Res 374:121–136

Hermann GJ, Scavarda E, Weis AM, Saxton DS, Thomas LL, Salesky R, Somhegyi H, Curtin TP, Barrett A, Foster OK, Vine A, Erlich K, Kwan E, Rabbitts BM, Warren K (2012) C. elegans BLOC-1 functions in trafficking to lysosome-related gut granules. PLoS One 7:e43043

Hermann GJ, Schroeder LK, Hieb CA, Kershner AM, Rabbitts BM, Fonarev P, Grant BD, Priess JR (2005) Genetic analysis of lysosomal trafficking in Caenorhabditis elegans. Mol Biol Cell 16:3273–3288

Hevelone J, Hartman PS (1988) An endonuclease from Caenorhabditis elegans: partial purification and characterization. Biochem Genet 26:447–461

Hieb WF, Rothstein M (1968) Sterol requirement for reproduction of a free-living nematode. Science 160:778–780

Hochbaum D, Zhang Y, Stuckenholz C, Labhart P, Alexiadis V, Martin R, Knolker HJ, Fisher AL (2011) DAF-12 regulates a connected network of genes to ensure robust developmental decisions. PLoS Genet 7:e1002179

Hsin H, Kenyon C (1999) Signals from the reproductive system regulate the lifespan of C. elegans. Nature 399:362–366

Hu Q, D'Amora DR, MacNeil LT, Walhout AJM, Kubiseski TJ (2018) The Caenorhabditis elegans oxidative stress response requires the NHR-49 transcription factor. G3 (Bethesda) 8:3857–3863

Hung WL, Wang Y, Chitturi J, Zhen M (2014) A Caenorhabditis elegans developmental decision requires insulin signaling-mediated neuron-intestine communication. Development 141:1767–1779

Hutter H (2012) Fluorescent protein methods: strategies and applications. Methods Cell Biol 107:67–92

Jia K, Levine B (2007) Autophagy is required for dietary restriction-mediated life span extension in C. elegans. Autophagy 3:597–599

Jiang H, Wang D (2018) The microbial zoo in the C. elegans intestine: bacteria, fungi and viruses. Viruses 10:E85

Jorgensen EM, Mango SE (2002) The art and design of genetic screens: caenorhabditis elegans. Nat Rev Genet 3:356–369

Kadayifci FZ, Zheng S, Pan YX (2018) Molecular mechanisms underlying the link between diet and DNA methylation. Int J Mol Sci 19: 4055

Kamath RS, Martinez-Campos M, Zipperlen P, Fraser AG, Ahringer J (2001) Effectiveness of specific RNA-mediated interference through ingested double-stranded RNA in Caenorhabditis elegans. Genome Biol 2:RESEARCH0002

Kandasamy S, Khan W, Evans F, Critchley AT, Prithiviraj B (2012) Tasco(R): a product of Ascophyllum nodosum enhances immune response of Caenorhabditis elegans against Pseudomonas aeruginosa infection. Mar Drugs 10:84–105

Kanzaki N, Tsai IJ, Tanaka R, Hunt VL, Liu D, Tsuyama K, Maeda Y, Namai S, Kumagai R, Tracey A, Holroyd N, Doyle SR, Woodruff GC, Murase K, Kitazume H, Chai C, Akagi A, Panda O, Ke HM, Schroeder FC, Wang J, Berriman M, Sternberg PW, Sugimoto A, Kikuchi T (2018) Biology and genome of a newly discovered sibling species of Caenorhabditis elegans. Nat Commun 9:3216

Kaplan REW, Maxwell CS, Codd NK, Baugh LR (2018) Pervasive positive and negative feedback regulation of insulin-like signaling in Caenorhabditis elegans. Genetics

Karasov WH, Douglas AE (2013) Comparative digestive physiology. Compr Physiol 3:741–783

Kenyon C, Chang J, Gensch E, Rudner A, Tabtiang R (1993) A C. elegans mutant that lives twice as long as wild type. Nature 366: 461–464

Khan F, Jain S, Oloketuyi SF (2018) Bacteria and bacterial products: foe and friends to Caenorhabditis elegans. Microbiol Res 215:102–113

Kimble J, Sharrock WJ (1983) Tissue-specific synthesis of yolk proteins in Caenorhabditis elegans. Dev Biol 96:189–196

Kimura KD, Tissenbaum HA, Liu Y, Ruvkun G (1997) Daf-2, an insulin receptor-like gene that regulates longevity and diapause in Caenorhabditis elegans. Science 277:942–946

Kiontke K, Sudhaus W 2006. Ecology of Caenorhabditis species. WormBook, 1–14

Kiontke KC, Felix MA, Ailion M, Rockman MV, Braendle C, Penigault JB, Fitch DH (2011) A phylogeny and molecular barcodes for Caenorhabditis, with numerous new species from rotting fruits. BMC Evol Biol 11:339

Kormish JD, Gaudet J, McGhee JD (2010) Development of the C. elegans digestive tract. Curr Opin Genet Dev 20:346–354

Kostic I, Roy R (2002) Organ-specific cell division abnormalities caused by mutation in a general cell cycle regulator in C. elegans. Development 129:2155–2165

Kramer JM 2005. Basement membranes. WormBook, 1–15

Lapierre LR, Gelino S, Melendez A, Hansen M (2011) Autophagy and lipid metabolism coordinately modulate life span in germline-less C. elegans. Curr Biol 21:1507–1514

Lapierre LR, Silvestrini MJ, Nunez L, Ames K, Wong S, Le TT, Hansen M, Melendez A (2013) Autophagy genes are required for normal lipid levels in C. elegans. Autophagy 9:278–286

Laurent V, Brooks DR, Coates D, Isaac RE (2001) Functional expression and characterization of the cytoplasmic aminopeptidase P of Caenorhabditis elegans. Eur J Biochem 268:5430–5438

Lee D, Anya A (1968) Studies on the movement, the cytology and the associated micro-organisms of the intestine of Aspiculuris tetraptera (Nematoda). J Zool 156:9–14

Lee YU, Son M, Kim J, Shim YH, Kawasaki I (2016) CDC-25.2, a C. elegans ortholog of cdc25, is essential for the progression of intestinal divisions. Cell Cycle 15:654–666

Lemieux GA, Ashrafi K (2015) Insights and challenges in using C. elegans for investigation of fat metabolism. Crit Rev Biochem Mol Biol 50:69–84

Lenaerts I, Walker GA, Van Hoorebeke L, Gems D, Vanfleteren JR (2008) Dietary restriction of Caenorhabditis elegans by axenic culture reflects nutritional requirement for constituents provided by metabolically active microbes. J Gerontol A Biol Sci Med Sci 63: 242–252

Lentjes MH, Niessen HE, Akiyama Y, de Bruine AP, Melotte V, van Engeland M (2016) The emerging role of GATA transcription factors in development and disease. Expert Rev Mol Med 18:e3

Leung B, Hermann GJ, Priess JR (1999) Organogenesis of the Caenorhabditis elegans intestine. Dev Biol 216:114–134

Lin K, Hsin H, Libina N, Kenyon C (2001) Regulation of the Caenorhabditis elegans longevity protein DAF-16 by insulin/IGF-1 and germline signaling. Nat Genet 28:139–145

Lin KT, Broitman-Maduro G, Hung WW, Cervantes S, Maduro MF (2009) Knockdown of SKN-1 and the Wnt effector TCF/POP-1 reveals differences in endomesoderm specification in C. briggsae as compared with C. elegans. Dev Biol 325:296–306

Lin R, Thompson S, Priess JR (1995) Pop-1 encodes an HMG box protein required for the specification of a mesoderm precursor in early C. elegans embryos. Cell 83:599–609

Lin XX, Sen I, Janssens GE, Zhou X, Fonslow BR, Edgar D, Stroustrup N, Swoboda P, Yates JR 3rd, Ruvkun G, Riedel CG (2018) DAF-16/FOXO and HLH-30/TFEB function as combinatorial transcription factors to promote stress resistance and longevity. Nat Commun 9: 4400

Liu H, Wang S, Hang W, Gao J, Zhang W, Cheng Z, Yang C, He J, Zhou J, Chen J, Shi A (2018) LET-413/Erbin acts as a RAB-5 effector to promote RAB-10 activation during endocytic recycling. J Cell Biol 217:299–314

Lu R, Maduro M, Li F, Li HW, Broitman-Maduro G, Li WX, Ding SW (2005) Animal virus replication and RNAi-mediated antiviral silencing in Caenorhabditis elegans. Nature 436:1040–1043

Macneil LT, Walhout AJ (2013) Food, pathogen, signal: the multifaceted nature of a bacterial diet. Worm 2:e26454

MacNeil LT, Watson E, Arda HE, Zhu LJ, Walhout AJ (2013) Diet-induced developmental acceleration independent of TOR and insulin in C. elegans. Cell 153:240–252

MacQueen AJ, Baggett JJ, Perumov N, Bauer RA, Januszewski T, Schriefer L, Waddle JA (2005) ACT-5 is an essential Caenorhabditis elegans actin required for intestinal microvilli formation. Mol Biol Cell 16:3247–3259

Maduro M, Hill RJ, Heid PJ, Newman-Smith ED, Zhu J, Priess J, Rothman J (2005) Genetic redundancy in endoderm specification within the genus Caenorhabditis. Dev Biol 284:509–522

Maduro MF (2017) Gut development in C. elegans. Semin Cell Dev Biol 66:3–11

Maduro MF, Meneghini MD, Bowerman B, Broitman-Maduro G, Rothman JH (2001) Restriction of mesendoderm to a single blastomere by the combined action of SKN-1 and a GSK-3beta homolog is mediated by MED-1 and -2 in C. elegans. Mol Cell 7:475–485

Magner DB, Wollam J, Shen Y, Hoppe C, Li D, Latza C, Rottiers V, Hutter H, Antebi A (2013) The NHR-8 nuclear receptor regulates cholesterol and bile acid homeostasis in C. elegans. Cell Metab 18: 212–224

Mallo GV, Kurz CL, Couillault C, Pujol N, Granjeaud S, Kohara Y, Ewbank JJ (2002) Inducible antibacterial defense system in C. elegans. Curr Biol 12:1209–1214

Mango SE (2009) The molecular basis of organ formation: insights from the C. elegans foregut. Annu Rev Cell Dev Biol 25:597–628

Martin J, Abubucker S, Heizer E, Taylor CM, Mitreva M (2012) Nematode.net update 2011: addition of data sets and tools featuring next-generation sequencing data. Nucleic Acids Res 40:D720–D728

McCormick M, Chen K, Ramaswamy P, Kenyon C (2011) New genes that extend Caenorhabditis elegans' lifespan in response to reproductive signals. Aging Cell 11:192–202

McGee MD, Weber D, Day N, Vitelli C, Crippen D, Herndon LA, Hall DH, Melov S (2011) Loss of intestinal nuclei and intestinal integrity in aging C. elegans. Aging Cell 10:699–710

McGhee JD 2007. The C. elegans intestine. WormBook, 1-36

McGhee JD, Fukushige T, Krause MW, Minnema SE, Goszczynski B, Gaudet J, Kohara Y, Bossinger O, Zhao Y, Khattra J, Hirst M, Jones SJ, Marra MA, Ruzanov P, Warner A, Zapf R, Moerman DG, Kalb JM (2009) ELT-2 is the predominant transcription factor controlling differentiation and function of the C. elegans intestine, from embryo to adult. Dev Biol 327:551–565

McGhee JD, Sleumer MC, Bilenky M, Wong K, McKay SJ, Goszczynski B, Tian H, Krich ND, Khattra J, Holt RA, Baillie DL, Kohara Y, Marra MA, Jones SJ, Moerman DG, Robertson AG (2007) The ELT-2 GATA-factor and the global regulation of transcription in the C. elegans intestine. Dev Biol 302:627–645

Miguel-Aliaga I, Jasper H, Lemaitre B (2018) Anatomy and physiology of the digestive tract of Drosophila melanogaster. Genetics 210:357–396

Mohrlen F, Hutter H, Zwilling R (2003) The astacin protein family in Caenorhabditis elegans. Eur J Biochem 270:4909–4920

Moreno-Arriola E, El Hafidi M, Ortega-Cuellar D, Carvajal K (2016) AMP-activated protein kinase regulates oxidative metabolism in Caenorhabditis elegans through the NHR-49 and MDT-15 transcriptional regulators. PLoS One 11:e0148089

Mulder NJ, Apweiler R, Attwood TK, Bairoch A, Barrell D, Bateman A, Binns D, Biswas M, Bradley P, Bork P, Bucher P, Copley RR, Courcelle E, Das U, Durbin R, Falquet L, Fleischmann W, Griffiths-Jones S, Haft D, Harte N, Hulo N, Kahn D, Kanapin A, Krestyaninova M, Lopez R, Letunic I, Lonsdale D, Silventoinen V, Orchard SE, Pagni M, Peyruc D, Ponting CP, Selengut JD, Servant F, Sigrist CJ, Vaughan R, Zdobnov EM (2003) The InterPro database, 2003 brings increased coverage and new features. Nucleic Acids Res 31:315–318

Murakami R, Okumura T, Uchiyama H (2005) GATA factors as key regulatory molecules in the development of Drosophila endoderm. Develop Growth Differ 47:581–589

Murphy CT, Hu PJ 2013. Insulin/insulin-like growth factor signaling in C. elegans. WormBook, 1-43

Narayanaswamy N, Chakraborty K, Saminathan A, Zeichner E, Leung K, Devany J, Krishnan Y (2018) A pH-correctable, DNA-based fluorescent reporter for organellar calcium. Nat Methods 16:95–102

Nehrke K, Denton J, Mowrey W (2008) Intestinal Ca2+ wave dynamics in freely moving C. elegans coordinate execution of a rhythmic motor program. Am J Phys Cell Phys 294:C333–C344

Nuez I, Felix MA (2012) Evolution of susceptibility to ingested double-stranded RNAs in Caenorhabditis nematodes. PLoS One 7:e29811

O'Rourke EJ, Soukas AA, Carr CE, Ruvkun G (2009) C. elegans major fats are stored in vesicles distinct from lysosome-related organelles. Cell Metab 10:430–435

Okkema PG, Harrison SW, Plunger V, Aryana A, Fire A (1993) Sequence requirements for myosin gene expression and regulation in Caenorhabditis elegans. Genetics 135:385–404

Okumura T, Matsumoto A, Tanimura T, Murakami R (2005) An endoderm-specific GATA factor gene, dGATAe, is required for the terminal differentiation of the Drosophila endoderm. Dev Biol 278: 576–586

Owraghi M, Broitman-Maduro G, Luu T, Roberson H, Maduro MF (2010) Roles of the Wnt effector POP-1/TCF in the C. elegans endomesoderm specification gene network. Dev Biol 340:209–221

Palgunow D, Klapper M, Doring F (2012) Dietary restriction during development enlarges intestinal and hypodermal lipid droplets in Caenorhabditis elegans. PLoS One 7:e46198

Panda O, Akagi AE, Artyukhin AB, Judkins JC, Le HH, Mahanti P, Cohen SM, Sternberg PW, Schroeder FC (2017) Biosynthesis of modular Ascarosides in C. elegans. Angew Chem Int Ed Eng 56: 4729–4733

Paradis S, Ruvkun G (1998) Caenorhabditis elegans Akt/PKB transduces insulin receptor-like signals from AGE-1 PI3 kinase to the DAF-16 transcription factor. Genes Dev 12:2488–2498

Peres TV, Arantes LP, Miah MR, Bornhorst J, Schwerdtle T, Bowman AB, Leal RB, Aschner M (2018) Role of Caenorhabditis elegans AKT-1/2 and SGK-1 in manganese toxicity. Neurotox Res 34:584–596

Pukkila-Worley R, Ausubel FM (2012) Immune defense mechanisms in the Caenorhabditis elegans intestinal epithelium. Curr Opin Immunol 24:3–9

Rasmussen JP, Feldman JL, Reddy SS, Priess JR (2013) Cell interactions and patterned intercalations shape and link epithelial tubes in C elegans. PLoS Genet 9:e1003772

Ratnappan R, Amrit FR, Chen SW, Gill H, Holden K, Ward J, Yamamoto KR, Olsen CP, Ghazi A (2014) Germline signals deploy NHR-49 to modulate fatty-acid beta-oxidation and desaturation in somatic tissues of C. elegans. PLoS Genet 10:e1004829

Rauthan M, Pilon M (2011) The mevalonate pathway in C elegans. Lipids Health Dis 10:243

Rechavi O, Houri-Ze'evi L, Anava S, Goh WSS, Kerk SY, Hannon GJ, Hobert O (2014) Starvation-induced transgenerational inheritance of small RNAs in C. elegans. Cell 158:277–287

Reinke SN, Hu X, Sykes BD, Lemire BD (2010) Caenorhabditis elegans diet significantly affects metabolic profile, mitochondrial DNA levels, lifespan and brood size. Mol Genet Metab 100:274–282

Rocheleau CE, Downs WD, Lin R, Wittmann C, Bei Y, Cha YH, Ali M, Priess JR, Mello CC (1997) Wnt signaling and an APC-related gene specify endoderm in early C. elegans embryos. Cell 90:707–716

Roh HC, Collier S, Guthrie J, Robertson JD, Kornfeld K (2012) Lysosome-related organelles in intestinal cells are a zinc storage site in C. elegans. Cell Metab 15:88–99

Rosa BA, Jasmer DP, Mitreva M (2014) Genome-wide tissue-specific gene expression, co-expression and regulation of co-expressed genes in adult nematode Ascaris suum. PLoS Negl Trop Dis 8: e2678

Sato K, Norris A, Sato M, Grant BD, 2014. C. elegans as a model for membrane traffic. WormBook, 1-47

Seah NE, de Magalhaes Filho CD, Petrashen AP, Henderson HR, Laguer J, Gonzalez J, Dillin A, Hansen M, Lapierre LR (2016) Autophagy-mediated longevity is modulated by lipoprotein biogenesis. Autophagy 12:261–272

Seymour MK, Wright KA, Doncaster CC (1983) The action of the anterior feeding apparatus of *Caenorhabditis elegans* (Nematoda: Rhabditida). J Zool 201:527–539

Sheng M, Hosseinzadeh A, Muralidharan SV, Gaur R, Selstam E, Tuck S (2015) Aberrant fat metabolism in Caenorhabditis elegans mutants with defects in the defecation motor program. PLoS One 10: e0124515

Shetty P, Lo MC, Robertson SM, Lin R (2005) C. elegans TCF protein, POP-1, converts from repressor to activator as a result of Wnt-induced lowering of nuclear levels. Dev Biol 285:584–592

Shin TH, Yasuda J, Rocheleau CE, Lin R, Soto M, Bei Y, Davis RJ, Mello CC (1999) MOM-4, a MAP kinase kinase kinase-related protein, activates WRM-1/LIT-1 kinase to transduce anterior/posterior polarity signals in C. elegans. Mol Cell 4:275–280

Silverman GA, Luke CJ, Bhatia SR, Long OS, Vetica AC, Perlmutter DH, Pak SC (2009) Modeling molecular and cellular aspects of human disease using the nematode Caenorhabditis elegans. Pediatr Res 65:10–18

Sommermann EM, Strohmaier KR, Maduro MF, Rothman JH (2010) Endoderm development in Caenorhabditis elegans: the synergistic action of ELT-2 and -7 mediates the specification → differentiation transition. Dev Biol 347:154–166

Song BM, Avery L (2013) The pharynx of the nematode C elegans: a model system for the study of motor control. Worm 2:e21833

Spence JR, Lauf R, Shroyer NF (2011) Vertebrate intestinal endoderm development. Dev Dyn 240:501–520

Steinbaugh MJ, Narasimhan SD, Robida-Stubbs S, Moronetti Mazzeo LE, Dreyfuss JM, Hourihan JM, Raghavan P, Operana TN, Esmaillie R, Blackwell TK, 2015. Lipid-mediated regulation of SKN-1/Nrf in response to germ cell absence. Elife 4

Stiernagle T, 2006. Maintenance of C. elegans. WormBook, 1–11

Sullivan-Brown JL, Tandon P, Bird KE, Dickinson DJ, Tintori SC, Heppert JK, Meserve JH, Trogden KP, Orlowski SK, Conlon FL, Goldstein B (2016) Identifying regulators of morphogenesis common to vertebrate neural tube closure and Caenorhabditis elegans gastrulation. Genetics 202:123–139

Sulston JE, Schierenberg E, White JG, Thomson JN (1983) The embryonic cell lineage of the nematode Caenorhabditis elegans. Dev Biol 100:64–119

Tan MW, Shapira M (2011) Genetic and molecular analysis of nematode-microbe interactions. Cell Microbiol 13:497–507

Tanji T, Nishikori K, Haga S, Kanno Y, Kobayashi Y, Takaya M, Gengyo-Ando K, Mitani S, Shiraishi H, Ohashi-Kobayashi A (2016) Characterization of HAF-4- and HAF-9-localizing organelles as distinct organelles in Caenorhabditis elegans intestinal cells. BMC Cell Biol 17:4

Tarr DE (2012) Distribution and characteristics of ABFs, cecropins, nemapores, and lysozymes in nematodes. Dev Comp Immunol 36: 502–520

Taubert S, Van Gilst MR, Hansen M, Yamamoto KR (2006) A mediator subunit, MDT-15, integrates regulation of fatty acid metabolism by NHR-49-dependent and -independent pathways in C. elegans. Genes Dev 20:1137–1149

Tauffenberger A, Parker JA (2014) Heritable transmission of stress resistance by high dietary glucose in Caenorhabditis elegans. PLoS Genet 10:e1004346

Tcherepanova I, Bhattacharyya L, Rubin CS, Freedman JH (2000) Aspartic proteases from the nematode Caenorhabditis elegans. Structural organization and developmental and cell-specific expression of asp-1. J Biol Chem 275:26359–26369

Teramoto T, Iwasaki K (2006) Intestinal calcium waves coordinate a behavioral motor program in C. elegans. Cell Calcium 40:319–327

Thomas JH (1990) Genetic analysis of defecation in Caenorhabditis elegans. Genetics 124:855–872

Thorpe CJ, Schlesinger A, Bowerman B (2000) Wnt signalling in Caenorhabditis elegans: regulating repressors and polarizing the cytoskeleton. Trends Cell Biol 10:10–17

Timmons L, Fire A (1998) Specific interference by ingested dsRNA. Nature 395:854

Troemel ER, Felix MA, Whiteman NK, Barriere A, Ausubel FM (2008) Microsporidia are natural intracellular parasites of the nematode Caenorhabditis elegans. PLoS Biol 6:2736–2752

True JR, Haag ES (2001) Developmental system drift and flexibility in evolutionary trajectories. Evol Dev 3:109–119

Van Gilst MR, Hadjivassiliou H, Jolly A, Yamamoto KR (2005) Nuclear hormone receptor NHR-49 controls fat consumption and fatty acid composition in C. elegans. PLoS Biol 3:e53

Vinci G, Xia X, Veitia RA (2008) Preservation of genes involved in sterol metabolism in cholesterol auxotrophs: facts and hypotheses. PLoS One 3:e2883

von Reuss SH (2018) Exploring modular glycolipids involved in nematode chemical communication. Chimia (Aarau) 72:297–303

von Reuss SH, Schroeder FC (2015) Combinatorial chemistry in nematodes: modular assembly of primary metabolism-derived building blocks. Nat Prod Rep 32:994–1006

Waaijers S, Boxem M (2014) Engineering the Caenorhabditis elegans genome with CRISPR/Cas9. Methods 68:381–388

Wang MC, O'Rourke EJ, Ruvkun G (2008) Fat metabolism links germline stem cells and longevity in C. elegans. Science 322:957–960

Watson E, Walhout AJ (2014) Caenorhabditis elegans metabolic gene regulatory networks govern the cellular economy. Trends Endocrinol Metab 25:502–508

Watts JL, Ristow M (2017) Lipid and carbohydrate metabolism in Caenorhabditis elegans. Genetics 207:413–446

Whangbo JS, Weisman AS, Chae J, Hunter CP (2017) SID-1 domains important for dsRNA import in Caenorhabditis elegans. G3 (Bethesda) 7:3887–3899

White JG, Southgate E, Thomson JN, Brenner S (1986) The structure of the nervous system of the nematode Caenorhabditis elegans. Philos Trans R Soc Lond Ser B Biol Sci 314:1–340

Wiesenfahrt T, Berg JY, Nishimura EO, Robinson AG, Goszczynski B, Lieb JD, McGhee JD (2015) The function and regulation of the GATA factor ELT-2 in the C. elegans endoderm. Development 143:483–491

Winston WM, Sutherlin M, Wright AJ, Feinberg EH, Hunter CP (2007) Caenorhabditis elegans SID-2 is required for environmental RNA interference. Proc Natl Acad Sci U S A 104:10565–10570

Witting M, Hastings J, Rodriguez N, Joshi CJ, Hattwell JPN, Ebert PR, van Weeghel M, Gao AW, Wakelam MJO, Houtkooper RH, Mains A, Le Novere N, Sadykoff S, Schroeder F, Lewis NE, Schirra HJ, Kaleta C, Casanueva O (2018) Modeling meets metabolomics—the WormJam consensus model as basis for metabolic studies in the model organism Caenorhabditis elegans. Front Mol Biosci 5:96

Woodruff GC, Phillips PC (2018) Field studies reveal a close relative of C elegans thrives in the fresh figs of Ficus septica and disperses on its Ceratosolen pollinating wasps. BMC Ecol 18:26

Wylie T, Martin J, Abubucker S, Yin Y, Messina D, Wang Z, McCarter JP, Mitreva M (2008) NemaPath: online exploration of KEGG-based metabolic pathways for nematodes. BMC Genomics 9:525

Xiao R, Chun L, Ronan EA, Friedman DI, Liu J, Xu XZ (2015) RNAi interrogation of dietary modulation of development, metabolism, behavior, and aging in C. elegans. Cell Rep 11:1123–1133

Yilmaz LS, Walhout AJ (2016) A Caenorhabditis elegans genome-scale metabolic network model. Cell Syst 2:297–311

Yin Y, Martin J, Abubucker S, Scott AL, McCarter JP, Wilson RK, Jasmer DP, Mitreva M (2008) Intestinal transcriptomes of nematodes: comparison of the parasites Ascaris suum and Haemonchus contortus with the free-living Caenorhabditis elegans. PLoS Negl Trop Dis 2: e269

Yokota S, Togo SH, Maebuchi M, Bun-Ya M, Haraguchi CM, Kamiryo T (2002) Peroxisomes of the nematode Caenorhabditis elegans: distribution and morphological characteristics. Histochem Cell Biol 118: 329–336

Zhang H, Abraham N, Khan LA, Hall DH, Fleming JT, Gobel V (2011a) Apicobasal domain identities of expanding tubular membranes depend on glycosphingolipid biosynthesis. Nat Cell Biol 13:1189–1201

Zhang H, Kim A, Abraham N, Khan LA, Hall DH, Fleming JT, Gobel V (2012) Clathrin and AP-1 regulate apical polarity and lumen formation during C. elegans tubulogenesis. Development 139:2071–2083

Zhang J, Bakheet R, Parhar RS, Huang CH, Hussain MM, Pan X, Siddiqui SS, Hashmi S (2011b) Regulation of fat storage and reproduction by Kruppel-like transcription factor KLF3 and fat-associated genes in Caenorhabditis elegans. J Mol Biol 411:537–553

Zhang J, Holdorf AD, Walhout AJ (2017a) C. elegans and its bacterial diet as a model for systems-level understanding of host-microbiota interactions. Curr Opin Biotechnol 46:74–80

Zhang SO, Box AC, Xu N, Le Men J, Yu J, Guo F, Trimble R, Mak HY (2010) Genetic and dietary regulation of lipid droplet expansion in Caenorhabditis elegans. Proc Natl Acad Sci U S A 107:4640–4645

Zhang YK, Sanchez-Ayala MA, Sternberg PW, Srinivasan J, Schroeder FC (2017b) Improved synthesis for modular Ascarosides uncovers biological activity. Org Lett 19:2837–2840

Zhao Y, Long L, Xu W, Campbell RF, Large EE, Greene JS, McGrath, P.T., 2018. Changes to social feeding behaviors are not sufficient for fitness gains of the Caenorhabditis elegans N2 reference strain. Elife 7

Zhou Y, Wang Y, Zhang X, Bhar S, Jones Lipinski RA, Han J, Feng L, Butcher RA (2018) Biosynthetic tailoring of existing ascaroside pheromones alters their biological function in C elegans. Elife 7: e33286

Zhu G, Yin F, Wang L, Wei W, Jiang L, Qin J (2016) Modeling type 2 diabetes-like hyperglycemia in C. elegans on a microdevice. Integr Biol (Camb) 8:30–38

Zhu J, Hill RJ, Heid PJ, Fukuyama M, Sugimoto A, Priess JR, Rothman JH (1997) End-1 encodes an apparent GATA factor that specifies the endoderm precursor in Caenorhabditis elegans embryos. Genes Dev 11:2883–2896

Zorn AM, Wells JM (2009) Vertebrate endoderm development and organ formation. Annu Rev Cell Dev Biol 25:221–251