

## *Caenorhabditis elegans* As Animal Models

The use of animal models to further understand diseases has become more prevalent over the years. Mice and rats are still the most common models of interest due to their genetic similarity to humans, ease of manipulation, and cost effectiveness along with a breadth of pharmacological literature (Holden-Dye et al., 2007). However, with the emergence of new diseases and their increasing complexity, it has become important to further study diseases in different models that offer new insights into the disease. A great example of such a model is *Caenorhabditis elegans* (roundworm), a transparent nematode that is widely used to study diseases such as Parkinson's and Alzheimer's, polycystic kidney disease, colon cancer, obesity, and sarcopenia in geriatrics.

*C. elegans* are an ideal organism to study diseases because they have a short life-span (3 weeks), rapidly proliferate with a life cycle of 3 days, are small in size which allows easy handling, and transparent for *in-vivo* visualization of individual neurons. Such *in-vivo* interactions have been able to provide results and data for protein-protein interactions, an unequivocally valuable piece of data for pharmacokinetics. They are also relatively easy to maintain and cost effective because they feed on bacteria and its various mutants, both of which make it ideal for genetic studies. The worm has 20,000 genes, 60-80% of which are similar to humans and of those 40% are analogous to human disease-related genes (Hasshoff et al., 2007).

Roundworms have provided great insight in to renal diseases such as polycystic kidney disease (PKD) which is most commonly an inherited autosomal dominant disease that causes cysts on the kidneys and is a major cause of end-stage renal disease. An underlying cause of PKD is a glycoprotein called polycystin-2, which has been identified in *C. elegans*. In humans,

polycystin-2 functions to maintain intracellular calcium homeostasis and renal tube development, whereas in *C. elegans* the glycoprotein controls male mating behavior. Despite the dissimilarity between the phenotypical functions of polycystin their “fundamental molecular and genetic interaction has been conserved” (Lipton, 2005) which allows for the study of PKD in *C. elegans*. Specifically, when LOV-1 is mutated, the round worm cannot find the female vulva. This gene is similar in molecular and genetic interactions to PKD-2 (gene that codes for polycystin-2) both of which are expressed in human cilia, implying the importance of cilia in treating PKD. Moreover, there is precedent for utilizing the *C. elegans*’ H-shaped secretory cells, which maintains homeostasis and osmolarity, for studying tubulogenesis. This study relates directly to PKD, as lesions caused by the disease lead to abnormally dilated renal tubules.

Similarly, neural cell death has been studied by using *C. elegans* as the model organisms. Neurodegenerative diseases such as Parkinson’s and Alzheimer’s are illnesses frequently diagnosed in elderly populations. Parkinson’s was found to be caused by cell death of dopamine producing cells (Martinez-Finley et al., 2011). Cell death can be triggered by genetic and environmental factors (e.g. exposure to toxic metals via chelation). Although a related disease, the cause for Alzheimer’s onset is still unknown however it is hypothesized that cholinergic neurons play a role. Nonetheless, a similar phenotype of extensive cell death is seen in both disorders.

*C. elegans* are an ideal model to study the effects of metals on neurons because of their translucent characteristic which allows for fluorescent labeling and observation of neuronal death instead of manipulating the organism surgically. Also, dopamine is responsible for movement, defecation, egg-laying and food sensation in *C. elegans* implying an assay that may be developed to analyze the effects of neurodegeneration. By utilizing *C. elegans*, Martinez-Finley et al. were

able to prove magnesium causes a “dose-dependent degeneration in dopaminergic neurons and the presence of a dopamine reuptake transporter (DAT1) is necessary for this neurodegeneration” (2011, p.4).

*C. elegans* are being used to study a wide range of diseases including cancer and specifically colon cancer. Tumor growth in colon cancer is stimulated when cancer cells acquire a set of somatic mutations. Certain mutations can cause chromosome instability in tumors, which can be used to target cancer cells. Other mutations allow the cells to evade checkpoints and grow in numerous amounts. Mutations in chromosome stability genes (CIN) occur early in tumor development therefore they are good targets for the inhibition of metastasis (McLellan et al., 2009). *C. elegans* were useful particularly in studying colon cancer because it is easier to conduct gene knockdown experiments with the use of RNA interference in *C. elegans* than performing analogous knockdown experiments in tissue cultures. First, yeasts (*S. cerevisiae*) were used to predict gene pairs that could be tested in proliferating somatic tissue, then the isolated genes were used with somatic vulval cells of *C. elegans* to test gene interactions that could possibly cure colon cancer. The formation of the vulva is mediated by a pathway that is similar to humans which further justifies the use of roundworms to study colon cancer. This research suggests a set of gene interactions that were conserved between *S. cerevisiae* and *C. elegans* that could be the target of potential drugs to halt the growth of cancer cells (McLellan et al., 2009).

Although *C. elegans* lack adipocytes and other genes that are hypothesized to cause obesity in humans, they have similar fat regulatory pathways, which warrants their use in obesity studies. Obesity is more prevalent than ever in the United States and it is caused by more than just a poor diet. Biologically, it occurs when the intake of fat and expenditure of fats as energy is

imbalanced. Energy intake and expenditure is regulated by the central nervous system, specifically by the hypothalamus. Leptin, a hormone secreted by the hypothalamus suppresses food intake and promotes energy utilization. Whereas insulin, released from the pancreas controls energy balance by regulating storage of glucose. These neurohormones amongst others play a key role in obesity and have shown to coordinate behavioral, physiological, and metabolic responses. All of the factors mentioned above are interconnected and influence energy balance (Ashrafi, 2007).

Targeted gene deletions, mutagenesis screens and RNA interference screens in *C. elegans* have been used to identify 300 genes that when inactivated cause fat reduction, and approximately 100 gene inactivations that cause fat accumulation (Ashrafi, 2007). The goal is to decrease obesity by manipulating the genes and other factors such as food sensation, neuroendocrine signaling, uptake, transport, storage and utilization of fats.

The ability to easily observe age-related anatomical changes in *C. elegans* due to their transparency has made them useful for studying adiposity and sarcopenia (generative loss of skeletal muscle mass and strength) in geriatrics. As we age, the accumulation of fat and loss of muscle mass and strength can limit our independence and predispose us to various diseases. Studying such changes in *C. elegans* could prevent many diseases associated with old age such as osteoporosis, and improve the lives of elders. Roundworms have comparable lipid and muscle homeostasis genetic pathways, and also lose muscle as they age, therefore being a good representation of aging in humans (Wolkow, 2010). Studies conducted by Wolkow revealed that neuroendocrine and environmental signals regulate fat metabolism and dysfunction in such pathways during aging can cause adiposity and sarcopenia. Specifically, the insulin/IGF-I pathway regulates expression of heat shock proteins and antioxidant enzymes thereby promoting

longevity and stress-resistance if the pathway is uninterrupted (Wolkow, 2010). Observation of tissue decline during *C. elegans* lifespan revealed tissue damage that occurred by oxidative damage and proteotoxicity (Wolkow, 2010). Now that specific pathways that cause muscle loss and fat accumulation have been identified with the use of *C. elegans* as animal models, the next step is to find therapeutic strategies by targeting “modulating muscle muscarinic or autophagic targets” (Wolkow, 2010).

One can also insert fluorescently labeled isotopes and watch their movement due to the transparent nature of *C. elegans*. Although roundworms appear very different from us they are good in representing all of the aforementioned diseases because of their similarity to our genes and metabolic pathways, the underlying key to understanding many diseases.

PKD, Parkinson’s, Alzheimer’s, colon cancer, and obesity are just some of the diseases that *C. elegans* are being used to study. The species has been helpful in creating anthelmintic drugs, understanding behavioral avoidance of pathogens, unraveling the mechanism of action of volatile anesthetics, and various topics in gerontology. *C. elegans* have helped us discover target genes for colon cancer drugs, understand the effects of metals in neurodegenerative diseases, and genetic factors that affect obesity and degenerative loss of skeletal muscle mass and strength as we age.

## References

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