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Probiotics Effect on Healthspan in the Model Organism Caenorhabditis elegans

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Abstract

In America, there are high rates of obesity and obesity related diseases partially due to a high glucose diet. Possessing a propensity towards reversing a glucose rich diet, probiotics, live microorganisms such as yogurt, were used to combat this. To experiment with humans let alone mice would be tedious, so, the microscopic nematode, *Caenorhabditis elegans* was used as a model. Because *C. elegans* possesses homology with humans, they can be used to study probiotics with application in humans. Sponsored by the Creative Inquiry program, this project analyzed the relationship between different probiotics and their effect on lifespan.

With *Escherichia coli* OP50 as the control and varying *Lactobacillus* strains as the experimental groups, lifespan assays were conducted. Our results show that certain probiotics increase the lifespan and reverse the consequences of a high glucose diet. Conversely, certain probiotic species, such as *Lactobacillus plantarum*, can decrease and promote negative health consequences in *C. elegans*. From these results, the understanding of the human microbiome and better treatments for obesity and obesity related diseases can be made.

Introduction

The Western diet consists of foods rich in glucose. In turn, high glucose consumption leads to gut microbiota alterations, which causes increased levels in Firmicutes and decreased Bacteroidetes. Additionally, there exists an increased paracellular permeability. These changes are linked with the obesity phenotype. One way to possibly improve one's diet is consuming products containing probiotics. Probiotics are microorganisms that promote good health and are ubiquitously present in our daily lives. The probiotics used in this experiment were the *Lactobacillus fermentum, Lactobacillus plantarum, Lactobacillus delbrueckii, Bacillus subtilis,* and *Bacillus coagulans*. These probiotics are found in the gastrointestinal tract, dairy products, and spoiled milk respectively.

C. elegans were used because of their short lifespans and simple maintenance. The lifespan of *C. elegans* maintained on *E. coli* OP50 was approximately 7-19 days.[2] These probiotics can be supplemented into a diet and may be a cost-effective way to decrease the effects of obesity. We hypothesized that certain *Lactobacillus* and *Bacillus* strains will reverse the negative effects of a high glucose diet using *C. elegans* as a model.

Aims

1. Determine the effects certain probiotic strains have on C. elegans lifespan.
2. Determine if the same probiotic strains can reverse the health consequences of a high glucose diet in *C. elegans*.

Methods

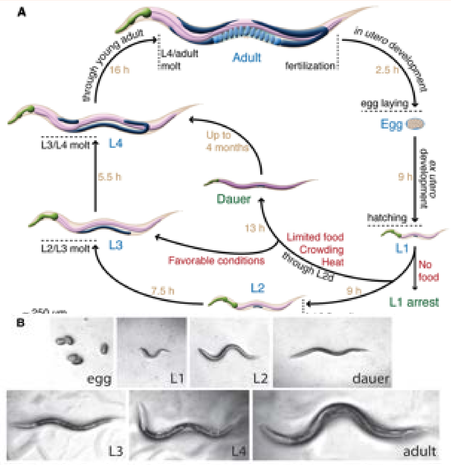

Figure 1. *C. elegans* life cycle and developmental stages. (Z. F. Altun and D. H. Hall (2012) Handbook of C. elegans Anatomy. In WormAtlas. http://www.wormatlas.org/hermaphrodite/hermaphroditehomepage.htm)

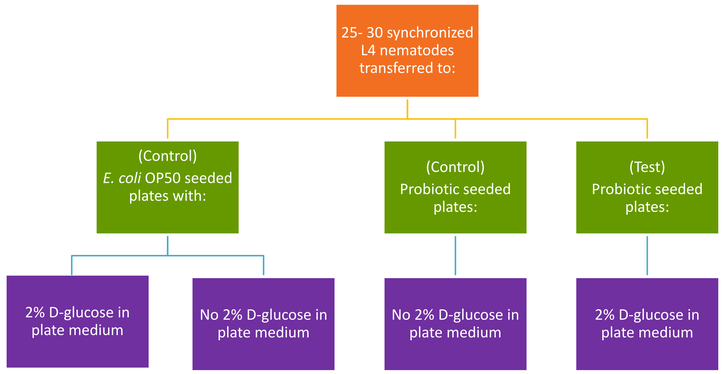

Figure 2. Diagram of *C. elegans* lifespan assay experiment using various probiotics.

Results

Diet	Mean±SE (Days)	N	P-value	% Decrease of Lifespan
E. coli OP50	13.442±0.225	95	-	-
OP50+2% glucose	11.333±0.207	69	<0.05	15.70%
B. subtilis	13.477±0.202	86	-	-
Bs+2% glucose	12.759±0.138	87	<0.05	5.30%
B. coagulans	17.261±0.221	92	-	-
Bc+2% glucose	15.301±0.142	73	<0.05	11.39%
L. fermentum	16.663±0.262	92	-	-
Lf+2% glucose	13.986±0.240	73	<0.05	16.10%
L. plantarum	15.085±0.273	82	-	-
Lp+2% glucose	5.342±0.130	73	<0.05	64.60%
L. delbrueckii	15.48±0.256	75	-	-
Ld+2% glucose	14.135±0.318	74	<0.05	8.70%

Table 1. Lifespan of nematodes on high glucose diet and probiotics at 25°C Lifespan of nematodes was shortened with all strains when supplemented with glucose. Lifespan restoration was observed in *Lactobacillus delbrueckii*, *Bacillus subtilis*, and *Bacillus coagulans*. Nematodes that were fed *Lactobacillus plantarum* exhibited an extreme decrease in lifespan when supplemented with 2% D-glucose. Data shown are representative of one round of lifespan assays.

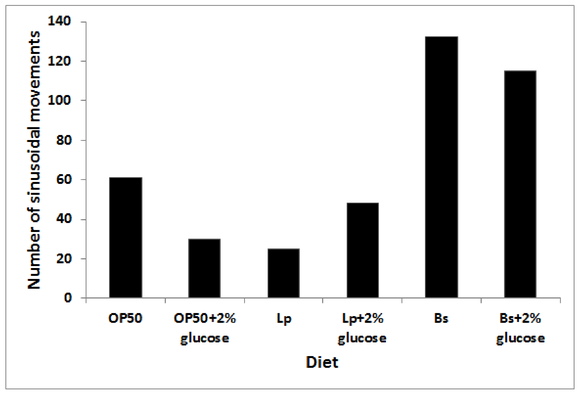

Figure 3. Preliminary body bend assay data L4 nematodes were placed on different diets--OP50, OP50 with 2% glucose, *L. plantarum*, *L. plantarum* with 2% glucose, *B. subtilis*, and *B. subtilis* with 2% glucose. After three minute intervals for 10 nematodes, sinusoidal motion movements were recorded for each of the diets.

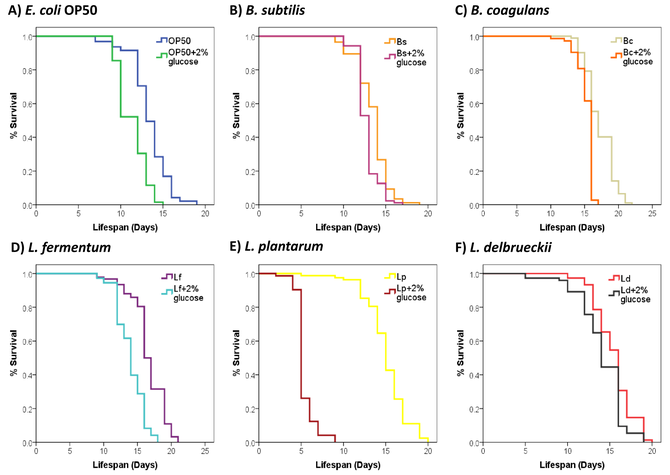

Figure 4. Lifespan curves of nematodes fed on each probiotic with or without 2% glucose supplemented into diet at 25°C.

Probiotics Used

Escherichia coli OP50 (control) - uracil auxotroph and *C. elegans* maintenance food source
Bacillus subtilis - generally found in soil
Bacillus coagulans - found in spoiled milk and Kombucha tea
Lactobacillus fermentum - found in sourdough breads
Lactobacillus plantarum - found in fermented plant products such as pickes and kimchi
Lactobacillus delbrueckii - generally found in dairy products

Conclusion

1. Consistent with previous research, the probiotic strains tested extended nematodes' lifespan.
2. Consistent with previous research, 2% D-glucose supplementation decreased nematodes' lifespan.
3. Probiotics alleviated the effects of a high glucose diet in *C. elegans* in a strain dependent manner.
4. *L. plantarum* enhanced the detrimental effects of a high glucose diet in *C. elegans*, resulting in a drastic reduction of lifespan.
5. Body bend assay data suggests that *L. planatarum* enhanced the detrimental effects of a high glucose diet.

Future Directions & Significance

Future directions include determination of long-term effects of probiotic supplementation, investigation of probiotic targeted pathways, experimentation with higher level organisms, and more body bends and brood size assays with the same probiotics.

From the lifespan data, with the exception of *L. planatarum*, the probiotics tend to increase the lifespan of the nematodes. As a possibility, there is a chance of creating both cost-effective treatments for obesity and obesity related diseases and also better understanding of the metabolic pathways of obesity and obesity-related diseases.

Acknowledgments

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