Effects of Alaska low bush cranberry and Alaska bog blueberry on Parkinson's-like α-synuclein aggregation in a transgenic model of *C . elegans*.

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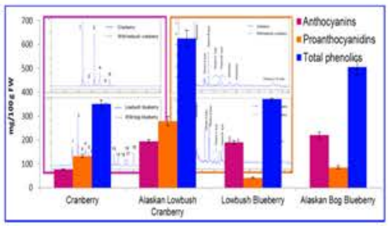
Introduction

•Parkinson's Disease is the second most prevalent neurodegenerative disorder affecting more than 10 million worldwide[1]

•Pathologically, Parkinson's is expressed by the aggregation of α-synuclein proteins and a loss of dopaminergic neurons in the midbrain, resulting in impairment of cognitive and motor functions[2]

•Alaskan botanicals like Bog Blueberry, Low Bush Cranberry and Crowberry have been shown to contain higher amounts of polyphenols relative to other related species[3]

• Preliminary data shows that certain levels of Cranberry and Blueberry extract separately improve lifespan, motility, and protein aggregation in *C. elegans*



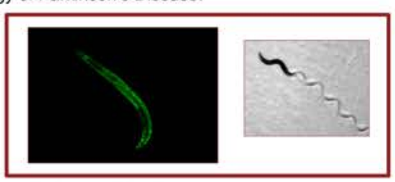
Alaskan berries contains more polyphenols compared to other botanicals (Grace et al., 2013).

Methods & Materials

Model used

Caenorhabditis elegans

• *C. elegans model* OW13 (P(unc-54)::alpha-synuclein::YFP+unc-119) with human α-synuclein expressed in body wall muscle. These worms have impaired motor function and express the molecular pathology of Parkinson's Disease.



Confocal images of OW13 *C. elegans* showing α-synuclein aggregation in head and tail of worm.

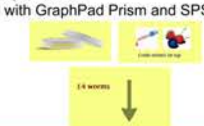
Alaskan Low Bush Cranberry & Bog Blueberry: Extraction and Characterization

Alaskan low bush cranberry (*Vaccinum vitis-idaea*)

• Crude extract was collected using 80% acetone/ Rotavap extraction.
• Anthocyanin and Flavonoids were quantified using pH-differential assay with cyanidin-3-glucoside equivalent (Song et al., 2013) and flavonoid content with $NaNO_2/Al/NaOH$ assay with a catechin standard curve (Herald et al., 2012).
• Anthocyanin content mg C3G L-1FW)- 201; Flavonoid content (mg CAE 100 g-1 FW) 813

Methods

• Age-matched population of worms were raised on live *OP-50-1* bacteria with treatment spread on top.
• Imaging performed on Day 7 using fluorescent microscope and densities were quantified using ImageJ.
• Lifespan assay was performed.
• Data was analyzed with GraphPad Prism and SPSS 22 software.



Results

Combination Study

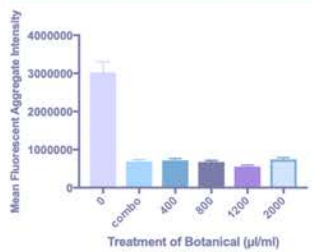
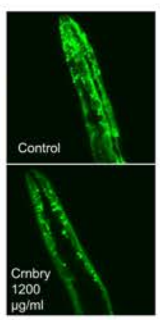


Fig. 1: Mean fluorescent intensity of α-synuclein aggregation in Day 7 worms of OW13 strain of C. elegans fed Alaskan Low Bush Cranberries (400, 800, 1200, and 2000 µg/ml) and one combination group (400 µg/ml Alaskan Bog Blueberry + 1200 µg/ml Alaskan Low Bush Cranberry) . The data represents the mean ± SEM
(n = 15)

Fig. 2: Mean Motility Assay based on an A,B,C grade on day 1 and day 12 OW13 strain of C. elegans fed Alaskan Low Bush Cranberries (400, 800, 1200, and 2000 µg/ml) and one combination group (400 µg/ml Alaskan Bog Blueberry + 1200 µg/ml Alaskan Low Bush Cranberry). The data represents the mean ± SEM (n=3) Control (0's) used from previous study, new control experiments underway soon.

Cranberry Study



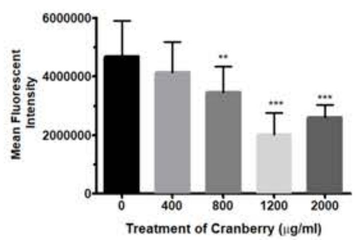


Fig 3: Mean fluorescent intensity of α-synuclein aggregation in Day 7 worms of OW13 strain of C. elegans fed Alaskan Low Bush Cranberries (0, 400, 800, 1200, and 2000 µg/ml). The data represent the mean ± SEM (n = 15) with significant differences between the control and treatments, $p<0.05$.

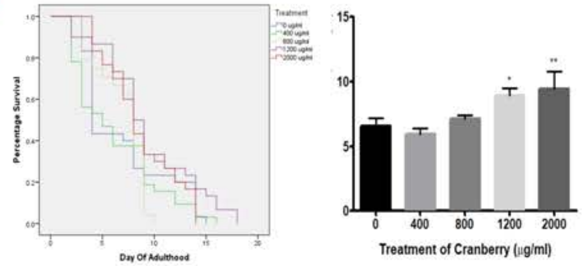


Fig 4: Representative survival curve (left) for low bush cranberry treatment groups (0, 400, 800, 1200 and 2000 ug/ml) and mean survival times for all treatment groups in all replicates (N = 25 per treatment group). Asterisks denote significant lifespan extension ($p < 0.05$; Kaplan–Meier log-rank test) from 3 independent trials. Bars represent mean ± standard error of the mean between all replicates of a treatment dose.

Discussion

• Results reveal that doses of 800,1200 and 2000 µg/ml of Cranberry reduce the α-synuclein aggregation in day 7 OW13 worms.

• Doses 1200 and 2000 µg/ml of Cranberry also improve lifespan in OW13 worms.

• Combination of 400 µg/ml of Blueberry and 1200 µg/ml of Cranberry reduce α-synuclein aggregation in day 7 OW13 worms.

• Combination and 400 µg/ml of Cranberry doses of increase the motility of day 12 OW13 worms.

Acknowledgements

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