Chapter

One Health and the Positive Effects of Alaskan Blueberries

Vincent F. Lembo and Cheryl A. Frye

Abstract

One Health is a principle that takes into account the interactions of humans, animals, the surrounding environment, and how they affect each other. In order to examine this concept in an experimental paradigm, the effects and benefits of wild Alaskan blueberries were compared to those from the continental United States (Lower-48 states) in human and animal studies. Blueberries have been hailed as a superfood for years now due to their high antioxidant levels and the positive effects they have on cardiovascular health and overall health and well-being. We hypothesize that although they are both beneficial, wild Alaskan blueberries have a greater positive effect on health and well-being than those from the lower 48. First, teachers and staff at the Anne Wien Elementary School in Fairbanks Alaska were provided with Alaskan and Lower-48 blueberries and asked to log the effects each coded sample had on their mental and physical health compared to a 5-day control period without blueberries. There was a significant stepwise positive effect of respondents reporting higher self-ratings of well-being overall. Alaskan blueberries significantly improved self-ratings of well-being compared to those from Lower-48 blueberries, albeit those blueberries did improve well-being compared to no blueberries. This experiment was replicated at a control site contemporaneously. The following year it was also replicated by participating teachers and staff at William S. Hackett Middle School in Albany, New York, as well as a control site. Further, lab rats, whose diets were supplemented with with Alaskan blueberries, performed better in exploratory and cognitive tests than did rats whose diets were supplemented with Lower-48 blueberries (who, similar to the previous trials, performed better than rats whose diets had not been supplemented at all). These findings suggest that blueberries have an overall positive effect on self-rated wellness in people and cognitive performance in lab rats and that Alaskan blueberries have a particularly greater and more beneficial effect. Whether this is due to greater antioxidant effects associated with higher altitude or fewer endocrine-disrupting contaminants in Alaska compared to the Lower-48 States is unknown and subject to ongoing investigation.

Keywords: antioxidants, cholesterol, triglycerides, low density liposaccharides, high blood pressure, hypercholesterolemia, cognitive decline, neurosteroids

1. Introduction

Consumption of fruits and vegetables is often correlated with brain and heart health and cancer prevention. One way that fruits and vegetables have beneficial effects on health is through their antioxidant actions. Not all fruits and vegetables are the same in terms of their antioxidant actions. When scientists tested 143

different plants for their antioxidant power: blueberries scored 4669 > cherries were 3747 and prune juice came in at 2036; avocado was at 1922; grapefruit had a power score of 1640, spinach was 1513 and raw broccoli was 1510 and pineapple was a low 385. These results on an ORAC scale compare items among food groups that show relative antioxidant activity. ORAC is a measure of water-soluble antioxidant levels and does not distinguish among those antioxidants that have benefits to humans and those that do not. It is simply an overall estimate of antioxidant activity in a particular experiment. Due to the limited utility of this index, the USDA's Nutrient Data Laboratory (NDL) removed the ORAC database from the NDL website [1].

Wild berries contain an impressive array of bioactive phytochemical compounds, which collectively present a range of biological activities targeting key mechanisms involved in healthy tissue development and aging [2, 3]. The human health-relevant bioactive properties of wild berries can be primarily attributed to their considerable diversity of polyphenolic constituents, typically exhibiting antioxidant, anti-inflammatory, and antimicrobial capabilities [4, 5]. The Nurses' Health Study evaluated 16,000 participants regarding cognitive decline and consumption of berries over a 20-year period. Greater intakes of flavonoids particularly from strawberries and blueberries were associated with slower cognitive decline in older adults [6]. This again supports the beneficial effects of blueberries.

Wild blueberries (*Vaccinium angustifolium*) are rich sources of polyphenols (e.g. flavonols, phenolic acids, anthocyanins) and decrease the risk of cardiovascular and degenerative diseases [7]. Memory function and mood in older adults with memory decline was improved by regular consumption of wild blueberries [8].

Various wild berry species endemic to Alaska and the circumpolar North that exhibit unique medicinal properties have long been appreciated by indigenous Arctic communities. Berry picking is a cherished tradition among all Alaskans that provides important physical and recreational activities for young and old alike. It has been an integral part of subsistence activities for thousands of years. Alaska is rich in wild edible berries that provide essential nutrients, especially vitamin C, and antioxidants for northern climates.

Interspecific differences, as well as environmental factors, such as geographic location and climatic variation, substantially influence the accumulation of berry phytochemicals and thus likely alter their profile of health-related bioactivities [9–11].

These experiments were designed to test the hypotheses that (1) wild blueberries would have beneficial short-term effects effect on healthy individuals (2) blueberries from Alaska would be more effective than those from the lower 48 (3) these effects would be seen across groups and species.

2. Methods

The protocol for Experiment 1 was approved by the Principal of Anne Wien Elementary School. All participants were volunteers and signed an informed consent indicating that they would comply with the guidelines of the experiment. At any time, volunteers could drop out as non-participants and their data would not be included.

2.1 Comparing effects of wild Alaskan blueberries to those from the lower 48 for effects on stress, mood, and affective function in people in Fairbanks, Alaska

The Native Alaskan blueberries used in this experiment were hand-picked by the authors, and their associates, the summer before the experiment began, and frozen

at −20 degrees Fahrenheit. These were low bush blueberries of large size, which are known to have high antioxidant value. The store-bought blueberries were purchased fresh from the local grocery store (Fred Meyers, Fairbanks, Alaska) around the same time. Half cup portions were prepackaged and labeled (A or B) and then placed in a refrigerator in the faculty lounge in the experimental locations.

2.1.1 Procedures for examining effects of blueberries

Experiment 1: In the first experiment, teachers and staff members at Anne Wien Elementary School (https://www.k12northstar.org/annewien) were provided with two samples of blueberries: hand-picked Alaskan blueberries, labeled A, and store-bought blueberries, labeled B, and surveys (see **Figure 1**) for them to answer after consuming the blueberries for 5 days or no blueberries control. This experiment was three weeks long. Participants consumed ½ a cup of one type of blueberries every day for one work week, repeated that process the next week with the other type of blueberries, and completed another survey after a third control week where no blueberries were consumed. Participants were allowed to use any sample any given week, so long as they only used that sample (or no sample) for the full work week and completed the other two sample weeks in the remaining time.

Custom surveys were created to enable participants to fully document the effects, positive, negative, or lack thereof, of the blueberries on their mental and physical health and well-being. Datasheets were turned in to the research supervisor at each site each week. There were 7 participants from Anne Wien Elementary (Fairbanks, AK) and 6 from an alternate site in Fairbanks (https://www.uafarc.com/overview), as well as 7 more from Hackett Middle School (Albany, NY) and 5 more from an alternate site in Albany later on for replication in the lower 48 States. (Experiment 2).

The protocol for Experiment 2 was approved by the principal of Hackett Middle School, all participants were volunteers and signed an informed consent indicating that they would comply with the guidelines of the experiment. At any time, volunteers could drop out as non-participants and their data would not be included.

Experiment 2: Comparing Effects of Wild Alaskan Blueberries to those from the Lower 48 for effects on Stress, Mood, and Affective Function in People in Albany, New York.

Experiment 2 was identical in procedure to experiment 1, the difference was the population that was tested. Experiment 2 took place at Hackett Middle School (https://www.albanyschools.org/schools/hackett/index) in Albany, New York about a year after experiment 1.

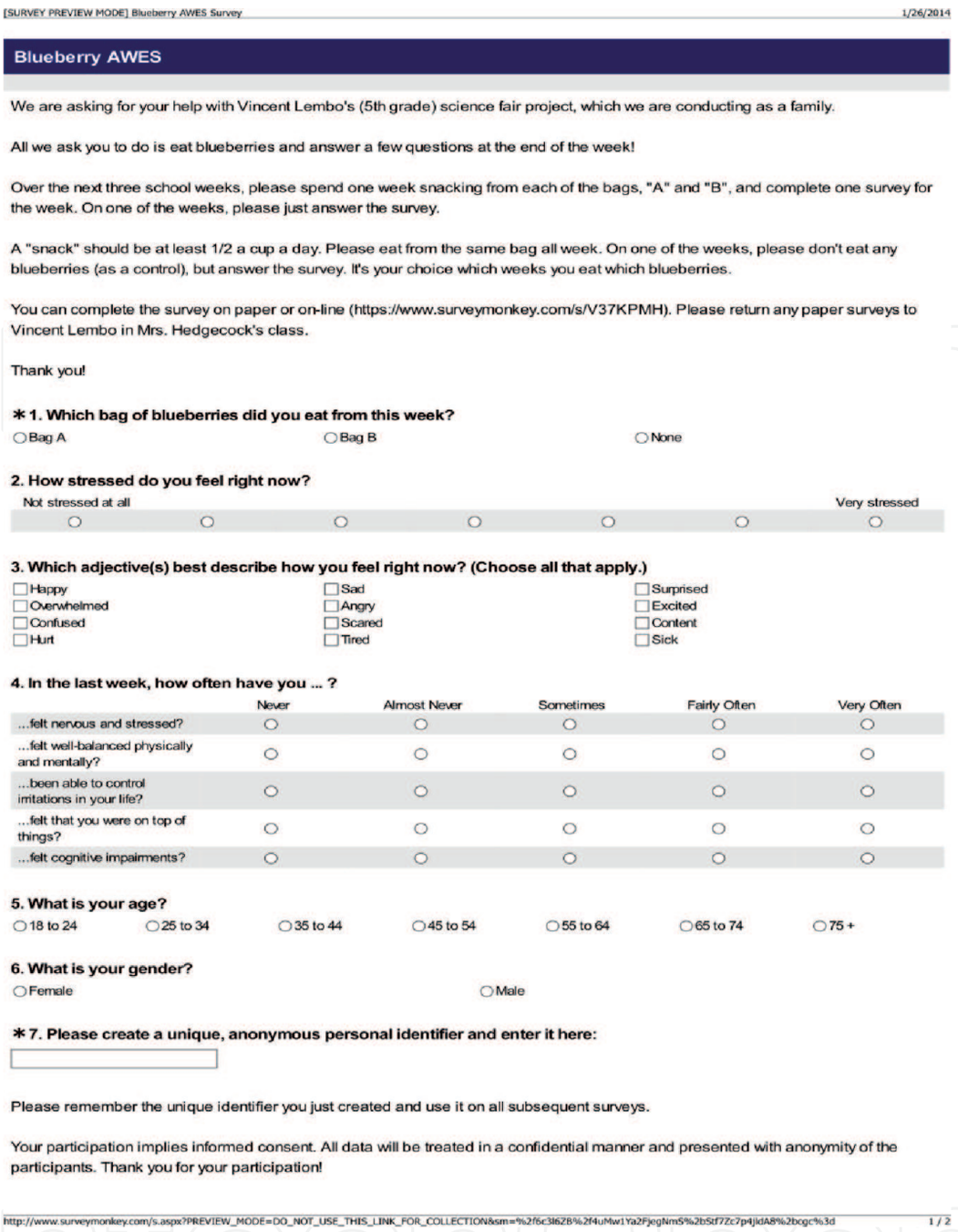
Experiment 3: Comparing Effects of Wild Alaskan Blueberries to those from the Lower 48 for effects on Stress, Mood, and Affective Function in Long-Evans Rats.

The University of Alaska Fairbanks Institutional Animal Care and Use Committee approved the animal care and experimental procedures (IACUC assurance number 497513).

2.2 Rats

2.2.1 Husbandry

Long-Evans rats were housed in polypropylene cages with wood shavings in a temperature (22 ± 1°C) and humidity (60–80%) controlled room of the Biological Research and Diagnostics (BiRD) Facility vivarium on a reversed 12:12 h dark-light cycle. Pups were weaned at 19–21 days of age and housed with same-sex littermates until the end of all experiments. Food (Purina Mills, Rat Diet, St. Louis, MO) and water were available *ad libitum*. Rats were also provided ½ cup blueberries from



[SURVEY PREVIEW MODE] Blueberry AWES Survey 1/26/2014

Blueberry AWES

We are asking for your help with Vincent Lembo's (5th grade) science fair project, which we are conducting as a family.

All we ask you to do is eat blueberries and answer a few questions at the end of the week!

Over the next three school weeks, please spend one week snacking from each of the bags, "A" and "B", and complete one survey for the week. On one of the weeks, please just answer the survey.

A "snack" should be at least 1/2 a cup a day. Please eat from the same bag all week. On one of the weeks, please don't eat any blueberries (as a control), but answer the survey. It's your choice which weeks you eat which blueberries.

You can complete the survey on paper or on-line (https://www.surveymonkey.com/s/V37KPMH). Please return any paper surveys to Vincent Lembo in Mrs. Hedgecock's class.

Thank you!

***1. Which bag of blueberries did you eat from this week?**

○ Bag A ○ Bag B ○ None

2. How stressed do you feel right now?

Not stressed at all ○ ○ ○ ○ ○ ○ ○ Very stressed

3. Which adjective(s) best describe how you feel right now? (Choose all that apply.)

☐ Happy ☐ Sad ☐ Surprised
☐ Overwhelmed ☐ Angry ☐ Excited
☐ Confused ☐ Scared ☐ Content
☐ Hurt ☐ Tired ☐ Sick

4. In the last week, how often have you ... ?

	Never	Almost Never	Sometimes	Fairly Often	Very Often
...felt nervous and stressed?	○	○	○	○	○
...felt well-balanced physically and mentally?	○	○	○	○	○
...been able to control irritations in your life?	○	○	○	○	○
...felt that you were on top of things?	○	○	○	○	○
...felt cognitive impairments?	○	○	○	○	○

5. What is your age?

○ 18 to 24 ○ 25 to 34 ○ 35 to 44 ○ 45 to 54 ○ 55 to 64 ○ 65 to 74 ○ 75 +

6. What is your gender?

○ Female ○ Male

***7. Please create a unique, anonymous personal identifier and enter it here:**

Please remember the unique identifier you just created and use it on all subsequent surveys.

Your participation implies informed consent. All data will be treated in a confidential manner and presented with anonymity of the participants. Thank you for your participation!

http://www.surveymonkey.com/s.aspx?PREVIEW_MODE=DO_NOT_USE_THIS_LINK_FOR_COLLECTION&sm=%2f6c3l62B%2f4uMw1Ya2FjegNm5%2bStf7Zc7p4jldA8%2bcgc%3d 1 / 2

Figure 1.
This figure depicts the survey used by the participants of each trial to record the effects (or lack of effects) that each sample of blueberries, as well as no blueberries, has had on them. This survey was completed at the end of each trial week by each participant and is completely anonymous. Several measures were provided for the user to describe and show how they felt after each trial week including a list of pronouns that the participant could choose best reflects them that week and survey questions that would help identify if blueberries were having a positive or negative effect on the participant. Demographic information was also collected for each participant.

Alaska or the lower 48 or none (as a control) each day for 4–5 days leading up to their anticipated 5th day of proestrus when they were tested.

2.2.2 Estrous cycle

Female rats in proestrus were used for the study. The proestrus stage was determined daily by both visual and vaginal cytology methods. Vaginal cytology was performed on those females that showed visual signs of proestrus. The females for which proestrus was confirmed through cytology were subjected to behavioral testing the same day. This procedure was conducted daily until the desired sample

sizes were achieved for a specific test. When the females cycled to their next proestrus, which was typically on the 4th or the 5th day, they were subjected to the next behavioral test in the schedule and this cycle was continued until all behavioral tests were completed. Typically, the no blueberry control week was the interval washout period between tests 1 and 3, which were randomized to blueberry condition.

2.2.3 Experimental design

All female rats were at least 60 days old at the start of the experiment. All behavioral tests were done in the dark phase of the light–dark cycle. For females, behavioral tests were conducted every 4–5 days depending on their cycle stage. All females underwent behavioral tests once every 5–10 days: open field (on day 5 or 10) and novel object recognition (on day 15 or 20). A minimum gap of 5 days for females between the two behavioral tests minimized the behaviors interfering with each other. An observer blind to the conditions of experimental animals and the hypothesized outcome of the study collected all data.

2.2.4 Open field

Behavior in the open field is used as a measure of exploration, anxiety, and locomotor behavior [12, 13]. The open field (76 cm x 57 cm x 35 cm) has a 48-square grid floor (6 x 8 squares, 9.5 cm/side): there is an overhead light illuminating the central squares (all but the 24 perimeter squares were considered central). Behavior was recorded by the ANYMaze video-tracking program (Stoelting Co., IL, USA). The apparatus was cleaned before and after each test. Per previous methods, rats were placed in the open field and the path of their exploration was recorded for five minutes. The number of squares entered by rats in the center or periphery of the grid was calculated and these data were added together to yield the total number of squares entered. Prior reports indicate that total square entries in this task are robustly modulated by the hormonal status of female rats and by steroid-sensitive manipulations [14–16]. Because the current study utilized a sample of female rats that were all matched on the phase of estrous cycle, motor differences were expected to be minimized. Thus, central square entries were utilized as an index of anti-anxiety, and total square entries as an index of thigmotaxis, and motor behavior. Additionally, engaging in this task provides habituation for object recognition, which occurs in the same box on the following days.

2.2.5 Object recognition

The object recognition task is a working memory task that primarily relies on cortical functioning and, to a lesser extent, hippocampal functioning [17–19]. This task was used as modified from previously published methods [20–22]. During training, rats are placed in a white open field (76 cm x 57 cm x 35 cm) in a brightly-lit testing room. Rats are allowed 3 min to explore the open field, which contains two identical objects in adjacent corners. These objects were colored spherical shapes (plastic toys in the shape of oranges). The time spent investigating the two identical objects (plastic toys) within a 5 cm distance in the open field arena was recorded for 3 min with the ANYMaze video tracking program (Stoelting.co, Chicago, IL). Rats were then taken out of the arena and returned to their holding cages for 4 hrs. After 4 hrs., one of the objects was replaced with a novel object of different shape and size, and animals were then reintroduced into the arena and allowed to explore the objects for 3 min. Time spent exploring the familiar and novel objects were recorded. The preference of one object over another was assessed through the Recognition

Percentage, which is the time spent on the novel object relative to the time spent on both novel and familiar objects: [RI = TN/(TN + TF) x 100] where TN is time spent on the novel object and TF is time spent on the familiar object). A greater percentage of time spent exploring the novel object as a function of the total amount of time spent exploring both objects during testing (duration spent with novel object/(duration spent with novel object + duration spent with familiar object) Å x 100) is considered an index of enhanced cognitive performance in these tasks.

2.2.6 Statistical analyses: one-way repeated measure analyses of variance was utilized to examine the effects of the condition (Blueberries) on self ratings

Post hoc analyses to determine group differences consisted of repeated t-tests with Bonferroni-corrections. Results are only reported for effects where the overall ANOVAs were significant, at the alpha level of 0.05, which was the case for each experiment. Overall effects and specific group differences are described below and in the tables and figures and their legends.

3. Results

Experiment 1a: Self-reports of health and wellness of middle-aged elementary school teachers in Fairbanks, Alaska were significantly greater following 5 days of consumption of Alaskan blueberries compared to blueberries from the Lower-48, which still had a beneficial effect compared to no blueberry consumption.

Self-reports of beneficial effects on stress, anxiety, and cognition are reported among women participants in Fairbanks, Alaska (demographics in **Table 1** left side, far) after 5 days of consumption of Alaskan blueberries > Lower-48 blueberries > no blueberries. Repeated measures analysis of variance revealed that there was an overall interaction between the type of blueberries and effects on self-ratings. *Post hoc* analyses revealed consumption of Alaskan berries resulted in higher ratings overall than did berries from the Lower-48 or no consumption of berries. This effect was seen for ratings of calmness, feelings of being well-balanced, and ability to maintain control and stay on top of things. Notably, self-reports of cognitive benefits of blueberries from Alaska, as well as the Lower-48, were not different but were significantly higher than the consumption of no berries among the participants at Anne Wien Elementary School. See **Figure 2**.

Experiment 1b: Demographics are described in **Table 1** (left center). Self-reports of health and wellness of young adult lab workers in Fairbanks, Alaska were significantly greater following 5 days of consumption of Alaskan blueberries compared to blueberries from the Lower-48, which still had a beneficial effect compared to no blueberry consumption.

To confirm and extend these findings, the experiment was replicated at an alternative site in Fairbanks (see **Table 1**, left middle, for demographics). There were even greater statistically significant interactions between types of blueberries and effects on self-reports at this alternative site than at Anne Wien Elementary School. Ratings for weeks following consumption of Alaskan blueberries > Lower-48 blueberries >no blueberries for self-reports of feelings of calmness, being well-balanced, in control, on top of things, cognitive benefits. Thus, women in Alaska who consumed blueberries for 5 days experienced beneficial effects as per their self-ratings. The magnitude of these effects was greater at the alternative site. See **Figure 3**.

	Fairbanks, AK		Albany, NY	
Location	**Anne-Wien Eiementary School**	**Alternate site**	**Hackett Middle School Albany, NY**	**Alternate site**
n = of participants	7	8	6	5
Age (avg. years)	42.3	33.6	40.0	48.7
Education (avg. years)	15.3	14.7	16.0	15.4
Ethnicity	5 Caucasian 2 Native Alaskan	3 Caucasian 2 Native Alaskan 2 Biracial 1 African American	2 Caucasian 2 African American 2 Latina	3 Caucasian 1 African American 1 Native Alaskan 1 Native American

Table 1.
The mean average demographics obtained from the Anne-Wien elementary school and the alternative site in Fairbanks (Byrd animal facility staff at U Alaska Fairbanks) on the left side. On the right side are the demographics for the Hackett middle School in Albany and the alternative site in Albany, NY (Comprehensive Neuropsychological Services).

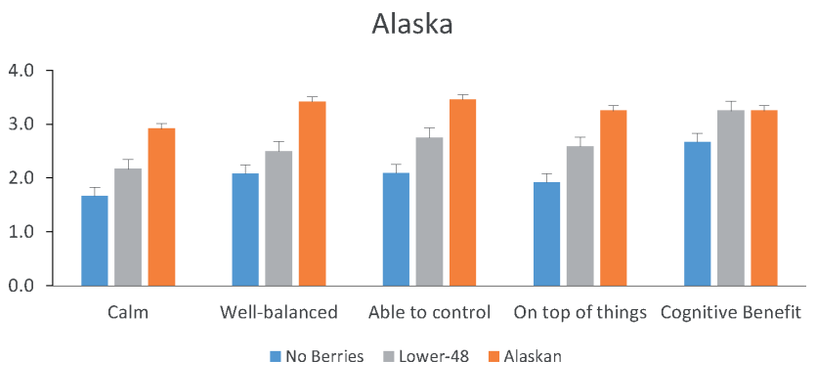


Figure 2.
This figure represents the mean + standard error of the mean of self-ratings in several categories by each participant in Fairbanks, Alaska at Anne-Wien Elementary School. This figure shows a significant overall effect of treatment (blueberries) to increase ratings of calm, a sense of well-balanced and ability to control surroundings and stay on top of things, except for self-ratings of cognitive benefit compared to consumption of no blueberries. Furthermore, consumption of wild Alaskan blueberries (orange) increased the participant's rating in each of the aforementioned categories compared to Lower-48 berries (gray) or no berries (blue).

Experiment 2a: Self-reports of health and wellness of middle-aged middle school teachers in Albany, New York were significantly greater following 5 days of consumption of Alaskan blueberries compared to blueberries from the Lower-48, which still had a beneficial effect compared to no blueberry consumption.

Self-reports of beneficial effects on stress, anxiety, and cognition are reported among women participants in Albany, New York after 5 days of consumption of Alaskan blueberries > Lower-48 blueberries > no blueberries. One-way, repeated analysis of variance revealed that there were overall interactions between the type of blueberries and effects on self-ratings. Some of the same patterns were observed among the participants at Hackett Middle School (Albany, NY) that were previously seen at Anne Wien Elementary (Fairbanks, AK). In particular, self-reports of feeling well-balanced, as well as in control and on top of things were higher in participants when they consumed Alaskan blueberries for a week compared to when they consumed Lower-48 blueberries or no blueberries. Demographics are reported in **Table 1** center-right. See **Figure 3** for results.

Experiment 2b: Self-reports of health and wellness of middle-aged office workers in Albany, New York were significantly greater following 5 days of consumption of Alaskan blueberries, compared to blueberries from the Lower-48.

There were even greater statistically significant interactions between types of blueberries and effects on self-reports at this alternative site than at Hackett Middle School in Albany, NY. Ratings for weeks following consumption of Alaskan blueberries > Lower-48 blueberries ≥ no blueberries for self-ratings of feelings of calmness, well-balanced. See demographics in **Table 1** and ratings in **Table 2**.

Experiment 3: Female Long-Evans rats whose diets were supplemented by Alaskan blueberries > Lower-48 blueberries > no supplement (control) were significantly more active, less anxious, and performed better in a novel object recognition task.

There were 8 female Long-Evans rats in each of the 3 trial groups. Those who had 5 days of access to Alaskan blueberries made more total entries (245 entries) into the brightly lit open field than did those with access to blueberries from the Lower-48 (223 entries), as well as those with no blueberry supplementation (196 entries) in a

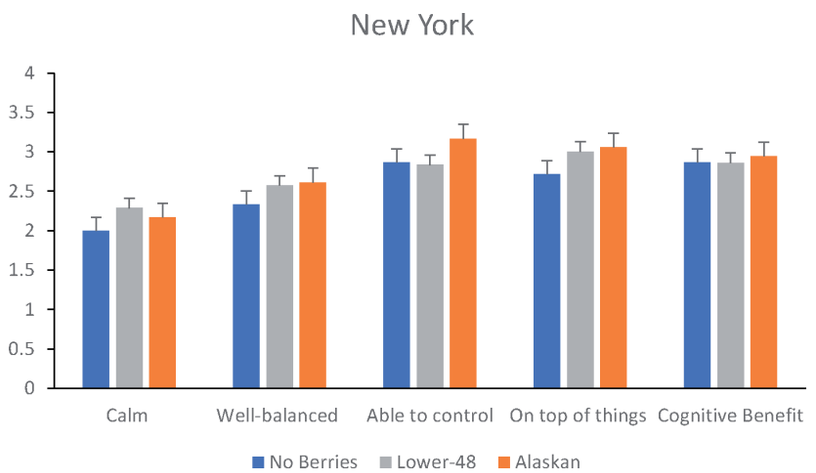


Figure 3.
This figure represents the mean + standard error of the mean of self-ratings in several categories by each participant in Albany, New York at Hackett Middle School. This figure shows a significant overall effect of treatment (blueberries) to increase ratings wild Alaskan blueberries (orange) to overall increase participant's rating in each of the aforementioned categories compared to Lower-48 berries (gray) or no berries (blue).

Self-ratings-alternative sites						
	Fairbanks-bird animal facility UAlaska			**Albany-comprehensive neuropsychological services**		
Blueberry Source	**None**	**Lower 48**	**Alaskan**	**none**	**Lower 48**	**Alaskan**
Self-Ratings						
Calmness	2.1	3.3	4.2	1.5	2.3	3.6
Well Balanced	1.6	2.5	3.6	1.8	2.9	4.3
Able to Control	1.5	2.3	3.8	1.3	2.4	2.7
On Top of Things	2.5	3.5	4.5	1.9	2.7	3.5
Cognitive Benefit	2.9	4.2	5.4	2.4	3.2	3.9

Table 2.
The mean average self-ratings obtained from the Fairbanks (n = 8) and Albany sites alternative site (n = 5) after 5 days of no consumption blueberries, lower 48 or Alaskan blueberries.

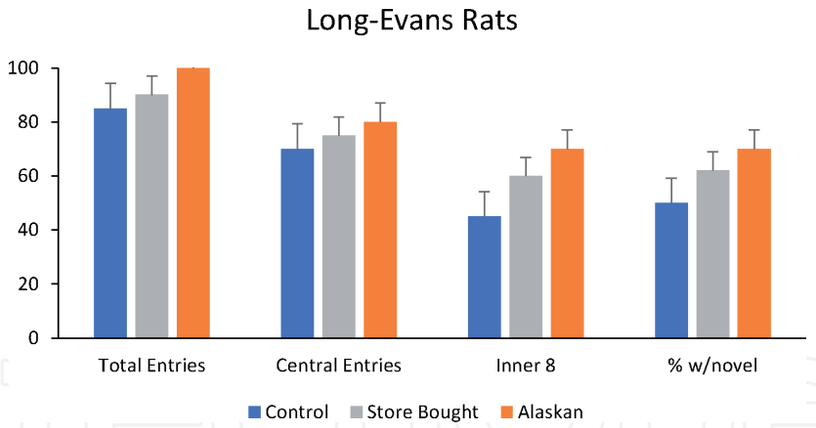


Figure 4.
This figure represents the scores in several tasks by Long-Evans laboratory rats at the BiRD Animal Research and Development facility at UAF as their diets were supplemented with Lower-48 or Alaskan blueberries, or no berries at all similar to human participants in the first trials. This figure shows that blueberries increase the number of total entries as well as central entries, Inner8 entries, and time spent with a novel object compared to consumption of no blueberries. Furthermore, consumption of wild Alaskan blueberries (orange) increased the rat's scores in each of the aforementioned categories compared to Lower-48 berries (gray) or no berries (blue).

5-minute period. The number of central entries (all but peripheral squares) during this 5-minute task was also greater for rats with access to Alaskan blueberries (120 entries) than rats whose diets were supplemented with blueberries from the Lower-48 (113 entries) as well as rats whose diets were not supplemented (93 entries). The number of inner 8 entries (central squares of open field) during this 5-minute task was greater for rats with access to Alaskan blueberries (25 entries) compared to rats whose diets were supplemented with blueberries from the Lower-48 (19 entries), as well as rats whose diets were not supplemented (13 entries). When rats were tested in the novel object recognition task, those consuming Alaskan berries spent 70% of their time than the novel object, whereas rats consuming Lower-48 berries spent only 61% of their time with the object. Rats whose diets were not supplemented spent the least amount of time, 51% (no greater than chance levels), with the novel object during testing (See **Figure 4**).

4. Discussion

The results of these experiments were consistent with the proposed hypotheses. First, blueberry consumption had beneficial short-term effects on healthy individuals. Blueberries had positive effects on the Alaskan participants' health and well-being after 5 days of consumption. This was also observed in the Albany cohorts. Participants consuming blueberries rated their levels of calm, well-balanced, ability to control themselves, and stay on top of things, and cognitive benefit, higher than when they consumed no berries. Second, this effect was seen more obviously when participants consumed Alaskan blueberries. They rated themselves even higher in the aforementioned categories than when they consumed Lower-48 blueberries. This effect was not identical; however, in New York, data from participants show an effect of blueberries but not a statistically significant difference between Alaskan blueberries and berries from the Lower-48. Third, blueberries had similar patterns of effects in rats. Notably, in more global tasks, such as total entries and central entries in an open field, there was an effect of blueberries, irrespective of their source, to improve performance among rats. However, in more challenging tasks, such as the Inner 8 entries and the percent of time with novel object, consumption of Alaskan blueberries had a greater effect than did those from the lower-48. In summary, short term consumption of blueberries can have

beneficial effects among some individuals. Further, the consumption of blueberries from Alaska had a more amplified effect than did berries from the Lower-48. Among Long Evans laboratory rats, Lower-48 or Alaskan blueberries increased motor activity and central entries (all non-peripheral squares) compared to control rats that did not consume any blueberries. However, rats consuming Alaskan blueberries made more Inner-8 entries, indicating that they were significantly bolder than rats that consumed Lower-48 blueberries or none at all. They also showed greater recollection and memory skills in the object recognition task than did those that consumed Lower-48 blueberries or none.

As mentioned in previous studies, blueberries as well as other fruits and vegetables have been linked to improving memory function as well as mood in older adults [23–25]. Our findings confirm and extend previous findings that blueberries can be beneficial to staving off cardiovascular disease, cognitive decline, and other age-related deleterious effects when consumed as part of a lifestyle. Here we show that short-term consumption of blueberries, 5 days of consumption, has perceived cognitive benefit among women age 30–55. Our participants' self-ratings of their perceived cognitive benefit from short term consumption of blueberries, as well as high ratings in other categories, such as their ability to remain calm and well-balanced, show that short term consumption of blueberries has the ability to improve mood and mental well-being among women age 30–55. Further, in our third experiment, healthy adult rats' performances benefited from blueberry consumption to have enhanced motor function, exploratory behavior, anti-anxiety effects, and learning. These latter findings are clearly objective, as they are not self-ratings and they demonstrate cross-species performance effects. Together, these findings confirm and extend the previous literature that older individuals can benefit from blueberry consumption as part of their lifestyle to indicate that short-term consumption of blueberries improves self-ratings of neurotypical women age 30–55 as well as objective effects on performance in rats. Another explanation may be that people in Alaska have greater access and exposure to foods with higher antioxidant levels than do people from the Lower 48 [26].

Consumption of Alaskan blueberries seemed to have a greater effect among participants in Alaska compared to participants in New York. An important question to ask is, what underlies this effect? One explanation could be that experiment 1 was conducted in Fairbanks, Alaska at the end of the winter season. This time of year in Alaska is also known as breakup and is associated with higher rates of suicide. There is tension in communities throughout Alaska caused by long harsh winters, and fresh blueberries may have had a greater effect on the moods and mental well-being of consumers during this sensitive time. Experiment 2 was conducted almost a full year later in Albany, New York, where the winters are much less harsh and dark. There may be some confounding variables due to these time frames. For example, the wild Alaskan blueberries had to be frozen for a longer period of time between experiments, 8 months for Experiment 1, versus 20 months, which may have caused a depletion in the potency of the Alaskan berries.

Another interesting finding relating to group differences was that the Alaskan blueberries had a greater effect among the office workers controls compared to the teachers in both control cohorts in Fairbanks and Albany. A likely explanation for this is that the office workers had a much calmer work environment than did the schoolteachers. They had greater control over their scope of work and when they engaged in their work activities, compared to being on a rigorous schedule with a limited break time that was preset and absolute. Further, office workers were not responsible for or surrounded by 20–30 children each day. They had a much quieter work environment. As such, the blueberries may have had a greater effect on individuals who were in a position to feel greater effects of the blueberries because

they could engage in other behaviors to enhance their wellbeing (such as listening to music, while at work engaging in brief meditation activities, having time for self-care).

All experiments have limitations, one possible limitation of this experiment was that the pool of participants was small, only 6–8 participants in each experiment. However, the fact that significant results were observed and replicated across multiple domains indicates the power and consistency of the effects shown in all 5 experiments. This limitation was out of our control because all participants were volunteers, and some participant's data were excluded because they did not participate in all 3 trial weeks. Another limitation is that a large majority of the participants in both trials were women, and mostly fell into the 30–55-year-old age range. A more diverse population next time might verify the results and show that the consumption of blueberries, especially wild Alaskan berries, are beneficial to everyone, and not just these populations. The notion that beneficial effects were seen in male rats suggests that this is a highly conserved effect and could be replicated on other populations. Another factor considered in this study is that living in Alaska is much closer to the earth and people in Alaska are much more affected by their environment. Experiences such as having 20 hours of sunlight each day in the summer and 20 hours of darkness in the winter have significant effects on mental and cognitive health and well-being. Also, the food that they eat is more likely to directly affect them; many Alaskans do not eat a strictly market diet but consume fish, game, and berries.

Blueberries are known to have a number of phytochemicals that contain several bioactive compounds that cause different physiological effects. One phytochemical are flavanols that contain the bioactive compounds *Epicatechin-gallates, Procyanidins,* and *Catechin* [27–30]. The physiological and pharmacological effects of these compounds include activities of antioxidants that increase free-radical scavenging, decrease the hypothalamic inflammation of microglia overactivation in the brain, and improve cognition. Further, other phytochemical effects include activities at phenolic acids, including the bioactive compounds phenolic acid and ferulic acid, which can have antioxidant and anti-inflammatory properties, as well as improve cognition and stave off neurodegeneration [31]. Blueberries are considered a functional food, which is defined as one that has clinically proven health benefits. Bioactive compounds such as flavonoids can suppress the release of cytokines such as IL-1beta and TNF-alpha from activated microglia. Flavonoids also affect nitric oxide synthase, inhibit activation of NADPH-oxidase, and down-regulate pro-inflammatory transcription factors including NF-kappa B which plays a role in intestinal responses through caffeic acid. NF-kappa B influences intestinal inflammatory responses through flavonoids' actions in part involving caffeic acid which inhibits the expression of TNF-alpha, IL-beta, CO-2, INOS, and other inflammatory factors [32–34].

An important question is how do blueberries have beneficial effects through the mechanisms described above? Particularly relevant to this study are Vitamins D, A, E, and K (fat-soluble vitamins). Vitamin D is considered a (neuro) steroid that has effects on calcium, metabolism, and absorption of calcium and phosphorus from the intestine. Vitamin D exerts many other biological effects including processes involved in brain development and neuronal activity [35]. Deficiency in Vitamin D is considered a risk factor for neuropsychiatric disorders, including post partum depression, major depression and schizophrenia [36–39]. It causes alterations in brain structure and in dopamine and glutamate signaling, which are hallmarks of depression, anxiety, drug abuse and schizoaffective disorders. Functional foods rich in micronutrients that have the ability to stimulate PPAR, in addition to exert important anti-inflammatory actions may also induce significant mood- elevating

properties, although the underlying mechanisms are not fully understood. PPAR might work in synergism with stimulation of neurosteroid biosynthesis to exert their beneficial effects by decreasing inflammation and relieving mood symptoms.

In summary, these experiments revealed that five days of blueberry consumption has beneficial effects on middle-aged women in Fairbanks, Alaska, as well as Albany, New York. The initial study in Fairbanks, Alaska (during "breakup season"), revealed a greater sensitivity and responsiveness to Alaskan blueberries than those from the Lower-48. Albeit, the second experiment, conducted in Albany, New York, showed a beneficial effect of blueberry consumption but not a differential effect between wild Alaskan and store-bought berries from the Lower-48. This may be due to the decomposition of antioxidants, flavonoids, and other important health factors caused by freezer storage for a full year. Our third experiment, using an animal model, conducted over the summer between the two human experiments, revealed beneficial effects of both Alaskan and Lower-48 blueberries when given as a supplement to male Long Evans rats. The rats that consumed blueberries had improved motor skills and exploratory behavior, irrespective of the source of blueberries. However, in more challenging tasks, such as entries into the Inner 8 squares in an open field (an anti-anxiety measure) and performance on the object recognition task, Alaskan blueberries had a greater effect on performance than did berries from the Lower-48 or no berries. To address the mechanism underlying these effects, we will be extracting the bioactive compounds and creating an Alaskan elixir to reassess their relative efficacy of Alaskan blueberries versus those from the Lower-48 on people as well as animal subjects.

5. Conclusion

In conclusion, it seems that blueberries can have short term effects as functional foods when consumed by individuals over a 5-day period. The precise mechanisms underlying these effects are unclear, however, differences between Fairbanks and Albany suggest that blueberries from a higher altitude may be more effective because they contain more flavonoids and more antioxidant effects. The beneficial effects of flavonoids may be mediated in part through actions of neurosteroids, such as Vitamin-D, allopregnanolone, 3α diol, which can inhibit deleterious effects of activation of PPAR and other physiological and pharmacological effects [34].

Acknowledgements

Research herein was supported in part by grants National Institute of Mental Health (MH06769801, RMH067698B) and the National Institute of General Medical Sciences of the National Institutes of Health (P20GM103395). The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health. Assistance in the studies described herein provided by members of the laboratory, past, and present, is greatly appreciated.

DOI: http://dx.doi.org/10.5772/intechopen.94738

IntechOpen

Author details

Vincent F. Lembo and Cheryl A. Frye*
Alaska INBRE Program, University of Alaska Fairbanks, Fairbanks, AK, USA

*Address all correspondence to: cherylafrye@gmail.com

References

[1] Oxygen radical absorbance capacity (ORAC) of selected foods, release 2. 2010. [cited 2012 Oct 10] US Department of Agriculture Agricultural Research Service. http://www.ars.usda.gov/Services/docs.htm?docid=15866.

[2] Paredes-Lopez O., Cervantes-Ceja M. L., Vigna-Perez M., Hernandez-Perez T. (2010). Berries: improving human health and healthy aging and promoting quality life—a review. Plant Foods Hum. Nutr. 65 (3), 299-308. 10.1007/s11130-010-0177-1

[3] Li Y., Zhang J. J., Xu D. P., Zhou T., Zhou Y., Li S., et al. (2016). Bioactivities and health benefits of wild fruits. Int. J. Mol. Sci. 17 (8), 1258. 10.3390/ijms17081258

[4] Seeram N. P. (2008). Berry fruits: compositional elements, biochemical activities, and the impact of their intake on human health, performance, and disease. J. Agric. Food Chem. 56 (3), 627-629. 10.1021/jf071988k

[5] Nile S. H., Park S. W. (2014). Edible berries: bioactive components and their effect on human health. Nutrition 30 (2), 134-144. 10.1016/j.nut.2013.04.007

[6] Devore EE, Kang JH, Breteler MM, Grodstein F. Dietary intakes of berries and flavonoids in relation to cognitive decline. Ann Neurol. 2012;72:135-43. doi: 10.1002/ana.23594.

[7] Michenaud-Rague A, Robinson S, Landsberger S. Trace elements in 11 fruits widely-consumed in the USA as determined by neutron activation analysis. J Radioanal Nucl Chem. 2012;291:237-40. doi: 10.1007/s10967-011-1266-9.

[8] Krikorian R, Schidler MD, Nash TA, Kalt W, Vinqvist-Tymchuk MR, Shukitt-Hale B, et al. Blueberry supplementation improves memory in older adults. J Agric Food Chem. 2010;58:3996-4000.

[9] Howard L. R., Clark J. R., Brownmiller C. (2003). Antioxidant capacity and phenolic content in blueberries as affected by genotype and growing season. J. Sci. Food Agric. 83 (12), 1238-1247. 10.1002/jsfa.1532

[10] Lila M. A. (2006). The nature-versus-nurture debate on bioactive phytochemicals: the genome versus terroir. J. Sci. Food Agric. 86 (15), 2510-2515. 10.1002/jsfa.2677

[11] Karppinen K., Zoratti L., Nguyenquynh N., Haggman H., Jaakola L. (2016). On the developmental and environmental regulation of secondary metabolism in Vaccinium spp. berries. Front. Plant Sci. 7, 655. 10.3389/fpls.2016.00655

[12] Blunt MJ, Blizard PJ. Recall and retrieval of anatomical knowledge. Br J Med Educ. 1975;9(4):252-263.

[13] Frye CA, Petralia SM, Rhodes ME. Estrous cycle and sex differences in performance on anxiety tasks coincide with increases in hippocampal progesterone and 3alpha,5alpha-THP. Pharmacol Biochem Behav. 2000;67(3):587-596. doi:10.1016/s0091-3057(00)00392-0

[14] Birke LI, Archer J. Open-field behaviour of oestrous and dioestrous rats: evidence against an 'emotionality' interpretation. Anim Behav. 1975;23(3):509-512. doi:10.1016/0003-3472(75)90127-x

[15] Frye CA, Paris JJ, Rhodes ME. Increasing 3alpha,5alpha-THP following inhibition of neurosteroid biosynthesis in the ventral tegmental area reinstates anti-anxiety, social, and sexual

behavior of naturally receptive rats. Reproduction. 2009;137(1):119-128. doi:10.1530/REP-08-0250

[16] Walf AA, Frye CA. The use of the elevated plus maze as an assay of anxiety-related behavior in rodents. Nat Protoc. 2007;2(2):322-328. doi:10.1038/nprot.2007.44

[17] Ennaceur A, Neave N, Aggleton JP. Spontaneous object recognition and object location memory in rats: the effects of lesions in the cingulate cortices, the medial prefrontal cortex, the cingulum bundle and the fornix. Exp Brain Res. 1997;113(3):509-519. doi:10.1007/pl00005603

[18] Broadbent JM, Thomson WM. Re: Diverse components of the oral environment in attention-deficit hyperactivity disorder (ADHD) make it difficult to establish whether ADHD is a risk factor for dental caries. JEBD 2005;5:39-40-review of Broadbent et al (2004). J Evid Based Dent Pract. 2006;6(3):250-252. doi:10.1016/j.jebdp.2006.06.001

[19] Akirav I, Maroun M. Ventromedial prefrontal cortex is obligatory for consolidation and reconsolidation of object recognition memory. Cereb Cortex. 2006;16(12):1759-1765. doi:10.1093/cercor/bhj114

[20] Ennaceur A, Delacour J. A new one-trial test for neurobiological studies of memory in rats. 1: Behavioral data. Behav Brain Res. 1988;31(1):47-59. doi:10.1016/0166-4328(88)90157-x

[21] Frye CA, Lacey EH. Posttraining androgens' enhancement of cognitive performance is temporally distinct from androgens' increases in affective behavior. Cogn Affect Behav Neurosci. 2001;1(2):172-182. doi:10.3758/cabn.1.2.172

[22] Luine VN, Jacome LF, Maclusky NJ. Rapid enhancement of visual and place memory by estrogens in rats. *Endocrinology*. 2003;144(7):2836-2844. doi:10.1210/en.2003-0004

[23] Popa-Wagner A, Dumitrascu DI, Capitanescu B, et al. Dietary habits, lifestyle factors and neurodegenerative diseases. Neural Regen Res. 2020;15(3):394-400. doi:10.4103/1673-5374.266045

[24] Giacalone M, Di Sacco F, Traupe I, Topini R, Forfori F, Giunta F. Antioxidant and neuroprotective properties of blueberry polyphenols: a critical review. Nutr Neurosci. 2011;14(3):119-125. doi:10.1179/14768305 11Y.0000000007

[25] Wood E , Hein S , Heiss C , Williams C , Rodriguez-Mateos A . Blueberries and cardiovascular disease prevention. Food Funct. 2019;10(12):7621-7633. doi:10.1039/c9fo02291k

[26] Dinstel RR, Cascio J, Koukel S. The antioxidant level of Alaska's wild berries: high, higher and highest. Int J Circumpolar Health. 2013;72:10.3402/ijch.v72i0.21188. Published 2013 Aug 5. doi:10.3402/ijch.v72i0.21188

[27] L. Wang, B. Waltenberger, E.M.Pferschy-Wenzig, M. Blunder, X. Liu, C. Malainer, *et al.*Natural product agonists of peroxisome proliferator-activated receptor gamma (PPARγ): a review Biochem. Pharmacol., 92 (2014), pp. 73-89

[28] M. Botta, M. Audano, A. Sahebkar, C.R. Sirtori, N. Mitro, M. RuscicaPPAR agonists and metabolic syndrome: an established role? Int. J. Mol. Sci., 19 (4) (2018)

[29] D. Mozaffarian, J.H.Y. WuFlavonoids, dairy foods, and cardiovascular and metabolic health: a review of emerging biologic pathways. Circ. Res., 122 (2) (2018), pp. 369-384

[30] A.B. Ward, H. Mir, N. Kapur, D.N.Gales, P.P. Carriere, S. SinghQuercetin inhibits prostate cancer by attenuating cell survival and inhibiting anti-apoptotic pathways. World J. Surg. Oncol., 16 (1) (2018), p. 108

[31] Mallik S.B., Mudgal J., Nampoothiri M., Hall S., Anoopkumar-Dukie S., Grant G. Caffeic acid attenuates lipopolysaccharide-induced sickness behaviour and neuroinflammation in mice. Neurosci. Lett. 2016;632:218-223.

[32] Ward et al., 2018 A.B. Ward, H. Mir, N. Kapur, D.N. Gales, P.P. Carriere, S. Singh Quercetin inhibits prostate cancer by attenuating cell survival and inhibiting anti-apoptotic pathwaysWorld J. Surg. Oncol., 16 (1) (2018),

[33] Wang et al., 2018J. Wang, P. Um, B.A. Dickerman, J. Liu Zinc, magnesium, selenium and depression: a review of the evidence, potential mechanisms and implications Nutrients, 10 (5) (2018)

[34] Matrisciano F, Pinna G. PPAR and functional foods: Rationale for natural neurosteroid-based interventions for postpartum depression. Neurobiol Stress. 2020 Apr 19;12:100222. doi: 10.1016/j.ynstr.2020.100222. PMID: 32426424; PMCID: PMC7226878.

[35] D. Eyles, T. Burne, J. McGrath Vitamin D in fetal brain development. Semin. Cell Dev. Biol., 22 (6) (2011), pp. 629-636

[36] S. Amini, R. Amani, S. Jafarirad, B.Cheraghian, M. Sayyah, A.A. HemmatiThe effect of vitamin D and calcium supplementation on inflammatory biomarkers, estradiol levels and severity of symptoms in women with postpartum depression: a randomized double-blind clinical trial Nutr. Neurosci., 3 (2020), pp. 1-11

[37] M.J. SzpunarAssociation of antepartum vitamin D deficiency with postpartum depression: a clinical perspective Publ. Health Nutr. (2019), pp. 1-6

[38] D.M. Zhu, W. Zhao, B. Zhang, Y. Zhang, Y. Yang, C. Zhang, et al.The relationship between serum concentration of vitamin D, total intracranial volume, and severity of depressive symptoms in patients with major depressive disorder Front. Psychiatr., 10 (2019), p. 322

[39] J.J. McGrath, D.W. Eyles, C.B.Pedersen, C. Anderson, P. Ko, T.H.Burne, et al.Neonatal vitamin D status and risk of schizophrenia: a population-based case-control study Arch. Gen. Psychiatr., 67 (2010), pp. 889-894