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Associations between Plant-Based Food Intake and COVID-19 Outcomes: A Study Protocol

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Abstract
Introduction: Coronavirus disease 2019 (COVID-19) left a lasting impact on the world where it not only had serious implications to the healthcare system, but also on individuals at a higher risk for other illnesses. In an effort to protect themselves from experiencing severe symptoms of COVID-19, other alternatives to alleviate the risk of the viral infection are being investigated. As documented in previous research, adopting a plant-based diet improves immunity against viruses. Hence, this study aims to investigate the associations between plant-based diets and the effects of COVID-19.
Methods: This is a cross-sectional study of 500 adults between the ages of 18 and 65 who have tested positive for COVID-19 in 2022. Participants are to fill out food-frequency and COVID-19 questionnaires. To detect significance, an ANOVA is to be used to compare the dietary patterns in each COVID-19 severity group (ASYMP, MILD, MOD, SEV, CRI).
Results: Expectedly, participants with the highest alongside most frequent intake of plant-based foods and seafood exhibit mild COVID-19 symptoms based on related studies. Whereas participants with the lowest intake of plant-based foods are expected to display symptoms corresponding to the critical and severe COVID-19 groups.
Discussion: Foods derived from plants are rich in vitamins and minerals that function as anti-inflammatories and boost immunity against COVID-19. Plant-based diets are comprised of low-fat, low-sodium foods, resulting in lower BMIs and risk of comorbidities like heart disease which further reduces the risk of severe COVID-19.
Conclusion: Considering this, a plant-based diet can be proposed as a lifestyle change that may aid in managing COVID-19. Through this protocol, the direct association between diet and COVID-19 will highlight the importance of nutrition for immunity as other infectious diseases continue to emerge.

Keywords: COVID-19; plant-based diet; inflammation; animal-based diet; treatment

Introduction

Coronavirus disease 2019 (COVID-19) is an acute respiratory infection caused by the virus SARS-CoV-2 that resulted in 593 million cases and 6 million deaths [1]. The widespread impact of COVID-19 can be attributed to the person-to-person transmission irrespective of demographic, age and gender [2,3]. As a result of SARS-CoV-2 transmitting through direct contact, public health restricted social contact and recommended a period of quarantine to minimize virus transmission [4]. However, amongst those that were infected, severe COVID-19 outcomes were commonly found in individuals suffering from pre-existing comorbidities such as respiratory diseases, malignancies, obesity, alongside cardiovascular disease [5]. Certain severe outcomes related to COVID-19 include hospitalization, intensive unit care admission, respiratory failure, or in some cases; death [5,6]. Individuals admitted to hospitals were commonly treated with board-spectrum antiviral therapeutics however, there is a lack of specificity against SARS-CoV-2 that make it less effective in treating COVID-19 [7]. Considering the treatment obstacles as well

as the catastrophic effects of the pandemic, other strategies to manage the effects of COVID-19 outcomes are being investigated.

One strategy that has been of interest during the emergence of COVID-19 is modifying dietary patterns [8]. A general shift towards plant-based diets (PBDs) was particularly observed during the pandemic [9]. A PBD is a dietary pattern that consists of increased consumption of plant-based foods such as vegetables, fruits, whole grains, nuts, seeds, and legumes in comparison to the smaller proportions of animal-based foods consumed [9,10]. Factors influencing the shift towards PBDs are related to improved nutrient intake, health outcomes, and sustainability [9,11]. To specify, a body of evidence has suggested that PBDs are associated with a lower body mass index (BMI), reduced systolic blood pressure, low cholesterol and blood sugar levels, as well as a decreased risk of chronic diseases [8, 10, 12,13]. Moreover, the risk of heart disease, obesity and type 2 diabetes are significantly reduced following a PBD [8, 12, 14]. Since COVID-19 is related to a state of inflammation, there is a rationale that

certain foods may modulate the presence of inflammatory agents and as such, influence health outcomes [15-18]. For instance, fruits and vegetables that are increasingly consumed in a PBD contain a variety of bioactive compounds involved in reducing inflammation [19]. Whereas foods not consumed in a typical PBD such as processed, red and white meats are positively associated with inflammatory biomarkers which may increase the risk of severe COVID-19 infections [20].

While the relationship between PBDs and chronic diseases has been widely researched, studies investigating the benefits of PBDs in relation to COVID-19-related outcomes are limited. In view of this, this study aims to investigate the association between plant-based food intake and COVID-19-related outcomes including hospitalization, intensive care unit admission, mechanical ventilation, and mortality. Doing so further highlights the role of nutrition in viral illnesses and how dietary choices may impact health. Moreover, insights into nutritional implications to support immune functionality are explored.

Methods
Study Design & Participant Selection

To evaluate the relationship between PBDs and the severity of COVID-19 infections, this cross-sectional study collects data on immunocompetent adults aged 20-65 with a minimum of two positive COVID-19 PCR or antibody test results in 2022. Participants are to complete an online-based questionnaire and are entered in a draw to win a $100 Amazon gift card. From the total number of participants screened, those who have a primary immunodeficiency disease such as; Bruton disease, DiGeorge Syndrome or Hyper-IgM syndrome are excluded. Additionally, participants with incomplete and inconsistent responses on COVID-19-related symptoms, disease severity and dietary assessment are excluded from the final study population.

Dietary Assessment

Dietary assessments of 500 participants are to be collected using a modified self-reported Food Frequency Questionnaire (FFQ) utilized by the European Prospective Investigation into Cancer and Nutrition Norfolk previously. The FFQ will be completed once by each participant to examine their dietary intake from 2021-2022. To modify

this questionnaire, additional questions making queries on COVID-19 symptoms, pre-existing medical conditions, intermediate family members deceased due to a COVID-19 infection, alongside potential treatments administered during the pandemic are incorporated into the FFQ. Besides focusing on the participants' diet and well-being during the pandemic, the questionnaire also focused on parameters such as age, income, occupation, gender, and ethnicity. The participants are divided into three groups based on their intake of plant-based foods: high consumption of plant-based foods (T4), equal consumption of plant- and animal-based foods (T2 & T3), as well as high consumption of animal-based foods (T1). This classification is based on the dietary assessment of the study sample and participants are organized into quartiles such that (T1=lowest consumption of plant-based foods; T2 = lower than average consumption; T3 = higher than average consumption; T4 = highest consumption of plant-based foods).

Characterization of COVID-19 Cases

From the divided participant groups, the participants are to report the number of days symptoms were experienced and are further categorized based on COVID-19 disease severity (asymptomatic, mild, moderate, severe, critical) as defined in the National Institute of Health guidelines [22]. Participants grouped into asymptomatic (ASYMP) tested positive for a virologic test, but did not exhibit significant physiological indications of an infection over a span of 14 days [21,22]. In the mild (MILD) group, participants who had expressed fever, cough, malaise, nausea, diarrhea, and body pain for less than 14 days in addition to a positive virologic test are included [22]. The moderate (MOD) group is comprised of participants who experienced the previously mentioned symptoms more severely for 14 days or more but not in a life-threatening manner [21,22]. The severe (SEV) group includes participants who were admitted into the hospital along with those who exhibited clinical signs of a respiratory infection such as a lower respiratory rate, reduced oxygen saturation of 94% at room temperature and overall respiratory distress [21,22]. In the critical (CRI) group, patients who were admitted into the intensive care unit, experienced organ dysfunction, respiratory failure or sepsis were analyzed [22].
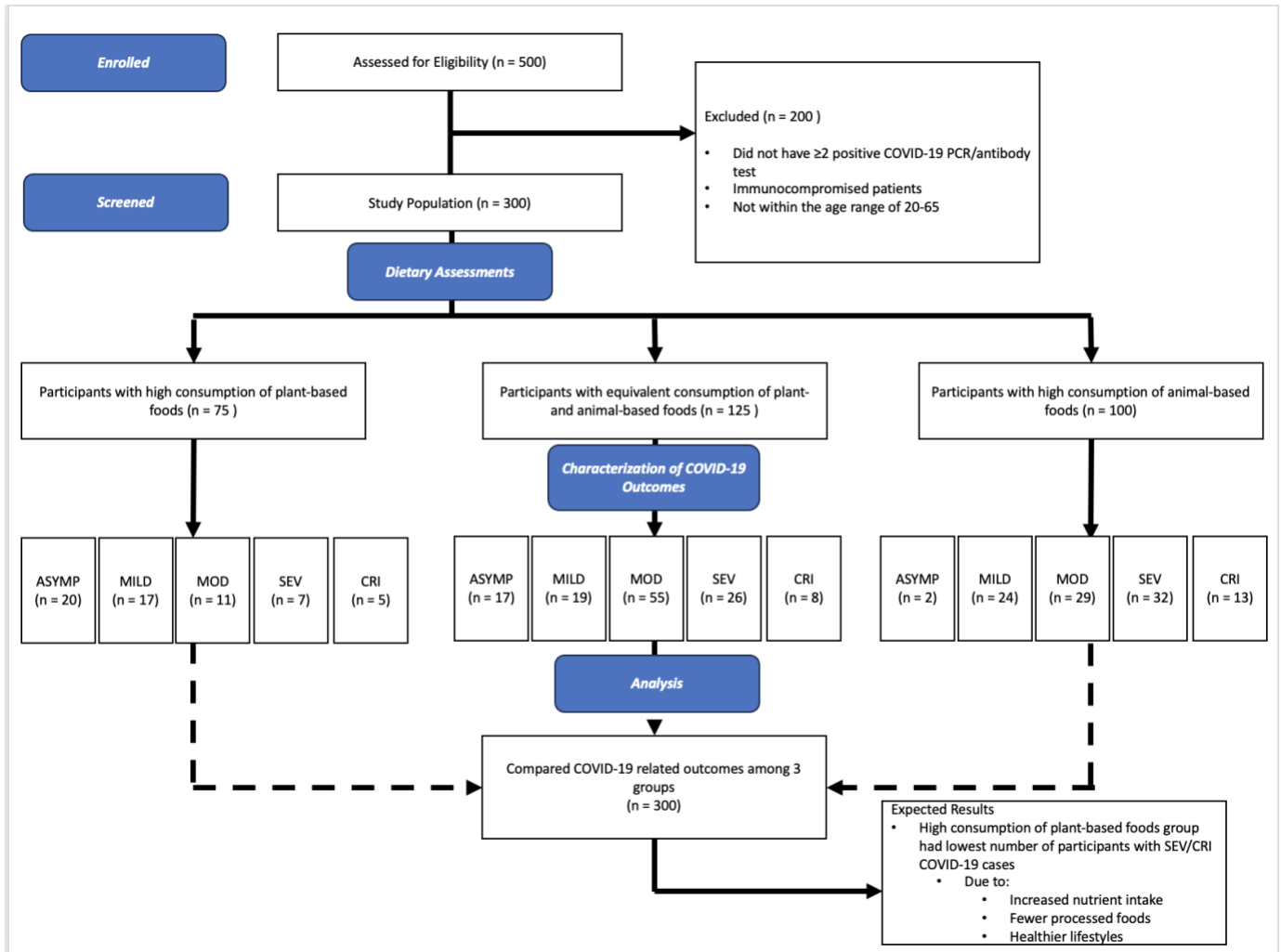
Figure 1. Schematic diagram outlining the study design. Figure made using Microsoft Powerpoint.

Data & Statistical analyses

To analyze the association between PBDs and its benefits for COVID-19-related outcomes, a one-way analysis of variance (ANOVA) test is performed to compare the COVID-19 severity scores in each of the dietary assessment groups. A p-value of 0.05 is considered the threshold for statistical significance.

Moreover, logistic regression is performed to examine the influence of PBDs on COVID-19 irrespective of other potentially confounding factors like age, gender, ethnicity and demographic. Through using multivariable models such as multiple logistic regressions as well as intraclass correlation coefficient, these potential confounders are held constant which allows the dietary patterns to be assessed closely.

Results

Participants

Study demographics include 500 total participants, with an expected 75 participants in the high intake of plant-based foods, 125 participants who consume relatively equivalent quantities of plant-based and animal-based foods and 100 participants in the high intake of animal-based foods. The expected median age of participants is 42 years old with a higher proportion of male participants.

Dietary Assessment

The ASYMP and MILD groups' participants expectedly demonstrate greater intakes of low-carbohydrate, high-protein foods such as legumes, nuts and tofu. In contrast, participants in the MOD to SEV groups' are anticipated to follow a pattern typical of a "Western diet" that consists of high portions of processed foods and refined grain products, with emphasis on foods like bacon, deli meats, white rice and white bread. Participants in the ASYMP to MILD groups, are expected to report eating habits associated with PBDs or pescatarian diets where higher intakes of fruits and vegetables, whole grains and dairy alternatives are consumed. Notably, the male participants surveyed expressed a dramatically higher

consumption of meat and alternatives compared to the female participants that are expected to have significantly increased intakes of fruits and vegetables. Older participants ranging from 50-65 are expected to demonstrate increased intake of fruits, vegetables and whole grains while the younger participants ranging from 18-30 are expected to exhibit slightly increased intakes of other food groups such as meats, poultry and seafood.

COVID-19 Outcomes

The diets of the screened participants can be correlated to their typical diets between 2021-2022. From examining dietary patterns and COVID-19-related outcomes simultaneously, the low-impact COVID-19 groups; ASYMP and MILD, reported little to no symptoms alongside had lower reports of hospitalization, ICU admission, and death. Meanwhile, the SEV and CRI groups are expected to exhibit significantly increased reports of symptoms associated with COVID-19 like difficulty breathing, pain in chest and inability to stay conscious alongside the need for additional medical care following positive virological tests. Despite this, the vast majority of screened participants are expected to demonstrate outcomes aligned with the MILD and MOD COVID-19 groups with very few reporting symptoms associated with the ASYMP and SEV to CRI groups. Throughout all the surveys, the highest overall incidence of COVID-19 reported by the participants is in those aged 20-29, with individuals over 55 years of age demonstrating increasingly severe disease outcomes.

It is anticipated that the associations between diets and COVID-19-related outcomes did not differ substantially when adjusted for confounding factors like BMI, age, gender, demographic and the presence of pre-existing conditions.

Discussion

The purpose of this study is to examine the variable COVID-19-related outcomes in those who consume high intakes of plant-based foods. The majority of participants who experienced little to no COVID-19-related symptoms had dietary patterns characterized by a high intake of plant-based foods while the moderate to severe groups typically consumed lower portions of plant-based foods and higher intakes of animal-based foods.

Similar to this research protocol, Kim et al. examined the relationship of COVID-19 outcomes and diets but in a study population of healthcare workers in France, Germany, Italy, Spain, UK and the USA [23]. Their participants that followed PBDs had lower odds of moderate to severe COVID-19 outcomes while the participants consuming low carbohydrate, high protein diets had greater odds of moderate to severe outcomes [23]. Interestingly, they also found that participants following pescatarian diets, a diet in which seafood and dairy are regularly consumed, have similar odds of moderate to severe COVID-19 outcomes as those who follow PBDs [23]. Alternatively, a study by Cobre et al. found that animal food products greatly contributed to relieving COVID-19-related outcomes as they are high-caloric, proteinaceous foods with essential micronutrients [24].

The discrepancies in literature could be related to the fact that PBDs are poorly defined within research [25]. From a nutrient-poor PBD, individuals do not acquire sufficient levels of nutrients essential for decreasing the risk of severe infections [24]. Studies hypothesize that while an unhealthy dietary pattern comprised of processed meats alongside refined sugars are pro-inflammatory and have negative health implications, individuals following a poorly balanced PBD are correspondingly also at an increased risk of severe COVID-19 infection [10,23,26]. Ongoing studies indicate that vegetarians and vegans who largely consume these foods are at a higher risk of deficiencies for vitamin B12 and D, iron, zinc and calcium [27]. As a result, there are not enough biological factors to modulate a COVID-19 infection and hence, these individuals consuming an unbalanced PBD are also at risk of severe COVID-19 outcomes [28,29].

Plant-based Diets and Immune Function

While micronutrient deficiencies increase the risk of severe COVID-19-related outcomes, food derived from plant sources provide individuals with various nutrients and phytochemicals responsible for mitigating the effects of a COVID-19 infection [17]. Specifically, PBDs consist of foods primarily regarded as anti-inflammatory [30]. Nuts which are high in polyunsaturated fats (PUFA) modulate immune pathways and work to inhibit the production of cytokines, a pro-inflammatory chemical agent which rapidly accumulates during a COVID-19 infection [15,31]. Subsequently, the immune system is not suppressed as expected which aids in the body's response to viruses and preventing organ damage [15]. Similar to the effects of PUFA, seafood also exerts anti-inflammatory effects [23]. A plausible explanation for the reduced risk of severe COVID-19 outcomes with a pescatarian diet in the previously mentioned study can be explained by the increased intake of omega-3 fatty acids [32,33]. PBDs are poor in eicosapentaenoic (EPA) and docosahexaenoic acids (DHA) while animal sources like fish and seafood are high in these omega-3, polyunsaturated fatty acids [33]. While nuts can be a great source of PUFA, plant-based foods do not supply individuals with sufficient levels of EPA and DHA which exert a greater inhibitory effect on the production of cytokines [32,33].

Through following a balanced PBD, high proportions of fruits and vegetables are also consumed which are major sources of vitamins A, C and E [27,34]. In comparison, individuals with a high intake of animal-based foods have lower levels of these micronutrients [27,35]. Prior research examining an elderly population found that they are also often deficient in vitamins C and E alongside magnesium,

subsequently increasing their susceptibility to COVID-19 [36]. These vitamins and minerals play a vital role in the innate immune system where they act as anti-inflammatories [27,37]. In a meta-analysis and systematic review by Panda et al., a deregulated immune response and increased inflammation contribute to high COVID-19 severity [38]. Through consuming foods high in magnesium as well as vitamins A, C and E, including plant-based foods, these micronutrients can reduce COVID-19 severity by improving the immune response [27,37]. Moreover, these vitamins further contribute to lower COVID-19 outcome severities by inhibiting the expression of receptors responsible for mediating pathogenesis hence, preventing an immuno-inflammatory reaction in individuals [39,40].

In spite of this, numerous studies have revealed that animal-based food products are also beneficial for reducing the severity of COVID-19. Particularly, animal-based foods are highly nutritious and consist of several essential nutrients in copious amounts relative to plant-based foods [41]. Notably, there are also certain nutrients, including vitamin B12, that are exclusively found in animal-based food products [7]. Vitamin B12 plays a pivotal role in modulating the gut microbiota in order to maintain the release of compounds such as methylmalonic acid and homocysteine which elevate inflammation [42]. Considering vitamin B12's role in decreasing inflammation, a cohort study in Singapore assessed the effects of vitamin B on the progression of severe COVID-19 patients in patients above 50 years old [43]. Subsequent to giving the elderly patients dietary supplements, Tan et al. observed a decrease in COVID-19 symptom severity to the point where the administration of oxygen alongside intensive care support was significantly reduced [43]. Likewise, another study surveying African American women for their experiences with COVID-19 infections found that there was a 69% increased reporting of testing positive alongside greater incidence of hospitalization in those deficient in vitamin D, a vitamin abundant in animal-based food products [17]. SARS-CoV-2 invokes an aggressive inflammatory response hence, adequate levels of vitamin D, which is responsible for modulating the innate immune system, may inhibit the production of inflammatory cytokines and as a result, decrease the severity of COVID-19 infections [44,45]. However, similar to vitamin B12, vitamin D deficiencies are common among individuals consuming a high proportion of plant-based food [10]. An explanation for this is the reduced intake of calcium which simultaneously decreases plasma vitamin D levels due to the exclusion of numerous foods [27].

The findings of these studies have further highlighted the importance of a balanced diet for reducing the severity of COVID-19 infections through consuming a plethora of different micronutrients. Contrary to the initial hypothesis, optimal nutrition obtained from a balanced diet strengthens the immune system against COVID-19 which also explains the contradictory findings in previous literature [46].

However, the lower odds of moderate to severe COVID-19 outcomes observed in this study may be explained by external factors such as the adoption of healthier, active lifestyles among participants who followed a PBD [47].

Strengths and Limitations

Despite this, a strength of this study is that it identifies biological mechanisms by which nutrition influences COVID-19 infections. Furthermore, the large sample size, adjustment for specific potential confounding factors, alongside plausible mechanisms by which a PBD could potentially be beneficial for COVID-19-related outcomes are also strengths of this study.

However, the findings should also be interpreted within the context of limitations. This study relied on self-reported dietary patterns and COVID-19 symptoms in which, the possibility of recall bias occurs. Additionally, the questionnaire did not account for how external factors may impact the findings. Participants consuming a PBD life tend to have healthier lifestyles which can be potential confounders when interpreting results [48]. Residual confounding factors may also be present due to improper assessment of the variables. Moreover, the protocol examining a small demographic of individuals aged 18-65 could reduce the external validity of this research.

Despite these limitations, this study provides important insights on the role of nutrition in COVID-19 management however, further research is necessary to confirm these results.

Future Developments

A balanced diet comprised of meats, fruits, vegetables, dairy along with grains is expected to be increasingly beneficial in reducing the risk of severe COVID-19-related outcomes instead of a PBD [49]. Nutrient inadequacies are expected to arise through dietary patterns consisting of nutrient-poor, staple foods, further increasing the risk of diseases such as COVID-19 [17]. To investigate this, future studies can examine the influence of a healthy, nutrient-dense diet in opposition to unhealthy PBDs comprised of low nutrient intake on COVID-19-related outcomes. Moreover, additional research may be conducted on COVID-19-related outcomes in individuals residing in regions associated with food insecurity or undernourishment to further solidify the findings of this study.

Conclusions

The intent of this protocol is to investigate the potential benefits of PBDs for COVID-19-related outcomes. By demonstrating such, this study highlights how nutrition impacts COVID-19 outcomes. Following a diet rich in whole grains, fruits, vegetables, dairy, seafood and other nutrient-dense foods can contribute to a well-functioning, effective immune system and be protective against viral illnesses. Despite the findings of this study, further

investigations are required to verify the association between PBDs and COVID-19 in addition to the duration of its therapeutic effects to guarantee its efficacy against this novel virus.

List of Abbreviations Used (If Any)
ANOVA: analysis of variance
ASYMP: asymptomatic
BMI: body mass index
CRI: critical
DHA: docosahexaenoic acids
EPA: eicosapentaenoic acids
FFQ: food frequency questionnaire
COVID-19: coronavirus disease 2019
MOD: moderate
MILD: mild
PBD: plant-based diet
PCR: polymerase chain reaction
SEV: severe
URNCST: Undergraduate Research in Natural and Clinical Science and Technology

Conflicts of Interest
The author declares there are no conflicts of interest.

Ethics Approval and Participant Consent
This study will require Brock University's Research and Ethics boards ethics approval and participant consent which will be conducted in accordance with the Tri-Council Policy. Prior to beginning the survey, the contents of the survey will be briefly summarized and permission to participate in the study will be sought through an online platform.

Authors' Contributions
IS: Contributed to the planning and design of the study, interpretation of data, drafting and critically revising of the manuscript, and provided the final approval of the version to be published.

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