PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Camu Camu effects on microbial translocation and systemic immune activation in ART-treated people living with HIV: protocol of the single-arm non-randomised Camu Camu prebiotic pilot study (CIHR/CTN PT032)
AUTHORS	Isnard, Stéphane; Fombuena, Brandon; Ouyang, Jing; Royston, Léna; Lin, John; Bu, Simeng; Sheehan, Nancy; Lakatos, Péter; Bessissow, Talat; Chomont, Nicolas; Klein, Marina; Lebouché, Bertrand; Costiniuk, Cecilia T.; Routy, Bertrand; Marette, André;Routy, Jean-Pierre

VERSION 1 – REVIEW

REVIEWER	Patel, Vainav National Institute for Research in Reproductive Health, Indian Council of Medical Research
REVIEW RETURNED	04-Oct-2021

GENERAL COMMENTS	Dear Authors, when selecting participants with 2 years of ART and >200 CD4 counts you would need to also document what 'response' or rebound CD4 count/ratio is from the initiation of therapy. This would enable stratifying results on the basis of whether CC effects occur in the context of CD4 rebound or not. Another factor to consider is the initial CD4 count and period of ART initiation following diagnosis as these have been shown to influence both rebound of CD4 counts as well as restoration of immune dysfunction. Further an age range is not specified and this is important as microbiome diversity as well as colonising ability is known to change significantly with age. Together with these specified criteria, a defined age range and a larger sample size, n=50 if possible, the quality of the study would be enhanced further.

REVIEWER	Serrano-Villar, Sergio Hosp Univ Ramon
REVIEW RETURNED	19-Oct-2021

GENERAL COMMENTS	Thank you for the opportunity to review the protocol for this study aimed at exploring the effects of camu camu, a fruit with antioxidant properties in people with HIV with incomplete immune recovery. This is a single arm, non-randomized, interventional pilot trial to assess the tolerance and effects of supplementation with camu camu on markers of gut damage, microbial translocation, inflammation, and HIV latent reservoir. The sample size is too small to assess efficacy, includes a substudy with colon biopsies, and the number of assessed

	outcomes will be high, so the study is exploratory, as it is expressed in the protocol where it is stated that the study will generate data to allow the sample size calculation of future larger studies. The study design, the intervention and the methods are well described in detail. The methodology is appropriate to ensure reproducibility and the SPIRIT 2013 checklist is appropriately reported.

VERSION 1 – AUTHOR RESPONSE

Reviewer: 1
Dr. Vainav Patel, National Institute for Research in Reproductive Health
Comments to the Author:
Dear Authors, when selecting participants with 2 years of ART and >200 CD4 counts you would need to also document what 'response' or rebound CD4 count/ratio is from the initiation of therapy. This would enable stratifying results on the basis of whether CC effects occur in the context of CD4 rebound or not. Another factor to consider is the initial CD4 count and period of ART initiation following diagnosis as these have been shown to influence both rebound of CD4 counts as well as restoration of immune dysfunction. Further an age range is not specified and this is important as microbiome diversity as well as colonising ability is known to change significantly with age. Together with these specified criteria, a defined age range and a larger sample size, n=50 if possible, the quality of the study would be enhanced further.

> We thank Reviewer 1 for the insightful comments and suggestions.
We agree that demographics such as age and sex should be taken into account in the analysis of the results. In addition to sexual practice, we will surely perform such analysis.
CD4 count, CD4 nadir and CD4/CD8 ratio will be available as part of the HIV medical history data, and will be taken into account in univariate (correlations with changes before-after CC) and multivariable analyses.
We have discussed those points in the “Statistical analyses“ section of the manuscript.
Finally, due to the pilot status and the limiting funds for this study, we were not able to increase the sample size, although we agree it would have given more definitive answers. We expect that results from this pilot study will allow sample size calculation for a more definitive, randomized placebo-controlled study.

Reviewer: 2
Dr. Sergio Serrano-Villar, Hosp Univ Ramon
Comments to the Author:
Thank you for the opportunity to review the protocol for this study aimed at exploring the effects of camu camu, a fruit with antioxidant properties in people with HIV with incomplete immune recovery. This is a single arm, non-randomized, interventional pilot trial to assess the tolerance and effects of supplementation with camu camu on markers of gut damage, microbial translocation, inflammation, and HIV latent reservoir.

The sample size is too small to assess efficacy, includes a substudy with colon biopsies, and the number of assessed outcomes will be high, so the study is exploratory, as it is expressed in the protocol where it is stated that the study will generate data to allow the sample size calculation of future larger studies.

The study design, the intervention and the methods are well described in detail. The methodology is appropriate to ensure reproducibility and the SPIRIT 2013 checklist is appropriately reported.

> We thank reviewer 2 for his distinctive and positive comments on the study design and methodology. We acknowledge that our study is exploratory and hope that it will allow the conduction of larger studies.

VERSION 2 – REVIEW

REVIEWER	Patel, Vainav National Institute for Research in Reproductive Health, Indian Council of Medical Research
REVIEW RETURNED	16-Nov-2021

GENERAL COMMENTS	I have no further comments.

REVIEWER	Serrano-Villar, Sergio Hosp Univ Ramon
REVIEW RETURNED	16-Nov-2021

GENERAL COMMENTS	I have no further comments.

VERSION 2 – AUTHOR RESPONSE

We thank Reviewer 1 for noticing this mistake. We have added in the Methods, in the section describing the study design, a paragraph describing the medical and medication history collected at screening. We have also added in the Statistical analysis section that those data will be included in multivariable analyses. We have checked that the medical history data collection has been included in the schedule of events, table 1.

We thank Reviewer 2 as well.