Point-by-point response to Editor and Reviewers

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Response to Editor's comments:

We would like to that the Editorial Board for allowing usto re-submit a revised manuscript. We performed all modifications related to the journal style as suggested, including fonts, ORCID numbers, required statements, article highlights and support for the study.

The **language certificate** was provided (uploaded attached) signed by one of the coauthors at McMaster University in Canada (Prof. E. F. Verdu). The manuscript was edited by Dr. Verdu. The **Audio Core Tip** was uploaded by the same coauthor using Core tips as referred in the body of the manuscript.

We hope the manuscript will now be acceptable for publication.

Point by point answer to reviewers' comments:

Reviewer #1: "*In this study, the authors assessed dietary compliance in celiac disease patients treated for more than two years with the gluten-free diet, using a combination of conventional and newly available tools, during the real-life conditions. The assessment included the quantitative determination of excretion of gliadin immunogenic peptides (GIP) in stool which was determined by ELISA and the newly developed point-of-care (PoCT) tests for real-time detection in stool and urine. their study has the novelty of comparing these PoCT tests for use at home or office, with the more conventional dietary assessment and laboratory-based tools. Another point of novelty includes the comparison of performance of these tests in celiac patients presenting with or without symptoms. their results demonstrate these new tools, which can be performed by the patient or by the health care provider, detect dietary indiscretions independently of symptoms*".

Response: We deeply thank the reviewer for the enthusiastic reception of our manuscript.

Reviewer#2:

1) *"...the results do not support their conclusion. The kappa value in this study is 0.317 (value less than 0.4 is not considered good agreement). Also, positive GIP excretion was noted in 17.9% subjects with dietary transgressions (line 239-241) – this suggest that the remaining 82.1% patients who had dietary transgression were not picked up by the test. Hence, this questions the performance of GIP excretion tests."*

Response: Our main conclusions are the following, that: 1)GIP in stool and urine are specific useful ***adjuvant tools*** for monitoring adherence to the GFD in real-life conditions of treated CeD patients, 2) simple, easy to perform, PoCT tools can also detect gluten contaminations, and 3) asymptomatic patients, in whom it is assumed that dietary adherence is high,may also suffer from contaminations.We state in the discussion that one important limitation relates to the lack of gold standard test to evaluate gluten contamination effectively. Thus, although kappa values are not striking, one must realize monitoring of GFD in celiac disease long-term is problematic given there is no single ideal method.Both methods are also not exactly simultaneous, dietary assessment "estimates" gluten intake in the previous weeks, and GIP tests objective excretion in the previous days of ingestion. The key important finding is that although mostly concordant, **GIP can detect gluten at times when dietary assessment suggests these patients are not having contaminations**. This is of key clinical utility.Dietician assessment is the best current approach but is subjective (both by the patient and the dietician), training dependent and with many limitations including the patients inadvertent or voluntary decision not to disclose a transgression. We acknowledge that this is an observational study, and that the lack of gold standard objective test is a limitation. However, for the reasons above, in our opinion, the three conclusions above are supported by our findings.

2) "*The number of symptomatic patients is high considering that they were on GFD for at least 2 years".*

Response: We are confident that the number of symptomatic patients is reflecting what occurs in the daily practice. Selection biasis unlikely since patients enrolled were

consecutive cases attending our specialized clinic. There was no sample size calculation for this study since one of the main objectives was to compare different tools, for the first time.

3) "*Why did the three GIP excretion test perform differently?"*

Response: These aspects were commented in the Discussion Section. Discrepancies between GIP in stool and urine are due to differences in the clearance time. More difficult to explain are the 10% discrepancies between ELISA and PoCT for stools. For the latter, more studies are necessary, and this is acknowledged in the manuscript.

4) "*Table 3 and 4 should be simplified. The language of paper in the results section needs to be more simple and clear. For example, the meaning from sentence 243 is not clear."*

Response: We believe that simplification of Tables 3 and 4 is a very difficult task which could impair understanding of some of our main findings. However, we took the reviewer's suggestion seriously and have edited and simplified the manuscript.

5) "*As significant number of patients had symptoms but were on strict GFD, what was the cause of symptoms in them?"*

Response: The objective of this study was not to explore factors associated with the presence of symptoms. Firstly, what patients consider as strict, is not necessarily strict. Secondly, there are several potential factors associated with symptoms. To determine if dietary indiscretions is associated with symptoms requires a different study design. We clearly state this was not an aim, and was a subgroup analysis that indicated some interesting trends for future research.

Reviewer #3: "*This is an elegant, well designed, performed and written observational, prospective, cross-sectional study for the evaluation of efficiency of ELISA and point-of-care (PoCT) for detection of gluten immunogenic peptides (GIP) excretion in stool and urine in celiac disease (CeD) patients on gluten-free diet (GFD). The authors aim also to*

explore the association of dietary transgressions with symptoms in CeD patients on long-term GFD. The authors investigated altogether a remarkable amount of patients, e.g. 44 of 62 screened CeD adults presenting detailed description of inclusion and exclusion criteria applied to select enrolled patients and samples received and analyzed. The authors give a sufficiently clear overview about the study background and raised clearly the aim of the study, which is fulfilled. The statistical analysis was specified sufficiently well. The Tables and Figures are correct and give a good overview about the results. The material studied is large enough and allows to drawn the conclusions. The Results are presented clearly and have been discussed well. The authors found that the detection of GIP in stool and urine can catch dietary transgressions in celiac disease patients on long-term GFD who are unaware of gluten consumption. The results have also practical application because simple home-based methods may aid in self-assessment of dietary indiscretions in CeD patients on a long-term GFD. Important finding of this study was that using ELISA and PoCT for detection of gluten immunogenic peptides is possible to detect gluten contaminations that do not cause symptoms. I found one misprint in the line 158: it should be added "in stool", I suppose: PoCT in stool and urine samples…"

Response: We thank the reviewer for the enthusiastic reception of our study. We have edited the revised manuscript carefully and we hoped to have corrected misspellings and typos as the one in line 158 (Methods Section; GIP by PoCT detection).