PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Fatty liver index and progression to type 2 diabetes: a five-year longitudinal study in Spanish workers with prediabetes
AUTHORS	Busquets-Cortés, Carla; Bennasar-Veny, Miquel; López-González, Angel-Arturo; Fresneda, Sergio; Aguiló, Antoni; Yanez, Aina

VERSION 1 – REVIEW

REVIEWER	Feizi, Awat Isfahan University of Medical Sciences, Department of Epidemiology and Biostatistics, School of Public Health, Isfahan University of Medical Sciences, Isfahan, Iran
REVIEW RETURNED	09-Nov-2020

GENERAL COMMENTS	Dear authors Please see my comments on your study, The following points are suggested Please add "workers" at the title appropriately. Outcome should be clarified along with the predictor and confounders in methods. The association between FLI with diabetes incidence should be evaluated in crude and adjusted models with cox regression and in both situations the HR (95%CI for HR) should be reported. Please follow this point in results section of abstract as well as main results section. your study is prospective then OR is not applicable! Limitations: the third and fourth points should be revised, the third is not relevant to the current study, and the fourth is not applicable; because you evaluated the PA and dietary habits as main component of lifestyle. Introduction Please provide more data and relevant literature regarding the possible clinical pathways between NAFLD and diabetes progression in future. Please enrich the relevant literature about the studies evaluated the association between NAFLD and diabetes incidence particularly among prediabetic patients. Please be clearer about your study novelty, particularly among this specific population. Methods All variables you have reported in results section, should be introduced completely beforehand in methods section (in relevant subsections), which instrument was used for PA, and how about its validity and reliability? I do not see sound and reference-based matters about dietary habits in methods section! please explain it and its instrument. Reference for FPG>126, is needed. Statistical analysis and results

	Statistical analysis in main area has major defects and results based on accordingly. Please see and follow my points in this regard and revise results section extensively based on. First of all, how you have evaluated normality of continuous variables? Your study is prospective cohort then you should have the time of affecting by diabetes for each subject and you Kaplan -Meier, crude cox regression and multivariable cox regression and report the HR and 95% CI for HR. during model fitting you should provide the crude HR for FLI, as your study predictor and you should consider the lowest level as refence category and report the HR for other categories. Adjustment should be made for age, gender, PA, dietary habits and smoking and report the adjusted HR in the presence of mentioned confounders. It is suggest to use predefined cut points for FLI, and use them in regression models, as you did for evaluated the FLI score diagnostic accuracy for diabetes incidence and cut point you found with best predictive value, I strongly suggest to use the predefined cut points that you have mentioned them in methods section for fitting new cox regression models based on above suggested approaches too. Reporting the incidence rate in these categories are more informative Subgroup analysis by gender is strongly recommended, please do it and report both incidence rate and the above suggested regression analysis. strangely I see the protective role for current smoking?!! The reported OR for age in crude is wrong, although as stated Kaplan -Meier and COX should be used. Please footnote the used statistical tests in table 1. You should also a new table for reporting and comparing the variables in table 1 between diabetic, prediabetic and maybe normal glucose tolerance (because prediabetes is reversable condition); this table is necessary. Title table is not correct, people with FLI >60 and FLI <60 are necessarily affected by NAFLD! Study limitations needs revisions, lifestyle variables have been evaluated in your study! Your study results are not generalizable to general population. How about the job related confounders such as job stress , ….please highlight these points as main limitations .

REVIEWER	Mundet Tuduri, Xavier Catedra UAB-Novartis de Docencia e Investigación en Medicina de Familia, Universitat Autònoma de Barcelona
REVIEW RETURNED	02-Dec-2020

GENERAL COMMENTS	a) The authors should describe the criteria of smoker, dietary habits, and phisical activity. Did they use any specific questionary? b) The authors have to explain why they consider that a patient progress to type 2 DM only with one FPG determination. c) It would be useful to describe the association of FLI with HbA1c criteria for Type 2 DM. d) The patients studied were working adults. Justify if they are repreesntative of general population or consider a limitation

REVIEWER	Miyoshi, Hideaki Hokkaido University, Division of Diabetes and Obesity, Faculty of Medicine and Graduate School of Medicine
REVIEW RETURNED	04-Dec-2020

GENERAL COMMENTS	This is a prospective cohort study, with 5-years follow-up to evaluate the association between NAFLD and the development of T2D in a large cohort of South-European Mediterranean workers with prediabetes. They indicated the FLI was the strongest

	predictor of progression to T2D and the optimal cut-off score for maximum accuracy for FLI was 59.5 with high sensitivity and specificity. The authors proposed that FLI might be useful in routine clinical practice as an additional screening tool to identify subjects with prediabetes who were at high risk of progression and could benefit from early interventions. Although the study was well organized and the results were reasonable, there are several comments for revision. Major 1. What is the primary endpoint of this prospective cohort study in the study protocol at first? The authors should include it in the Methods section. 2. Excess intake of fruits is also known as the risk of NAFLD. The analysis of daily fruits and vegetables should be divided and reanalyzed each. 3. OGTT is more informative and important compared with just measuring FPG. Please show the sources for evidence regarding the sentence: '(OGTT), which is considered more sensitive but less specific than FPG for identifying people at risk of developing T2D. (page 15)'. What's the 'specific'? Minor 1. The way of writing for 'progression to T2D' is mixed-up (T2D conversion, development of T2D,,,). That should have consistency in the manuscript. 'progression to T2D' would be the best. 'prediabetes reversion' should be arranged (page 14). 2. The sentence should be arranged: 'This would benefit patients at greater risk for T2D, allowing more careful monitoring and providing an opportunity for early interventions to prevent and reduce both the progression of hepatic disease. (line 7-9, page 15)' 3. Discussion is bit too long. Please focus more.

VERSION 1 – AUTHOR RESPONSE

Reviewer: 1
Dr. Awat Feizi, Isfahan University of Medical Sciences
Comments to the Author:
1) Please add "workers" at the title appropriately.
R: We are grateful for the suggestion. We added the word "workers" in the title of the manuscript.

2) Outcome should be clarified along with the predictor and confounders in methods.
R: We agree with the reviewer. We have now clarified the outcome in the methods section and the specified the confounders in the statistical analysis section.

3) The association between FLI with diabetes incidence should be evaluated in crude and adjusted models with cox regression and in both situations the HR (95%CI for HR) should be reported. Please follow this point in results section of abstract as well as main results section. Your study is prospective then OR is not applicable!
R: We thank the reviewer for the suggestion. We have fitted and reported Cox regression models. Accordingly, changes were made to the results section.

4) Limitations: the third and fourth points should be revised, the third is not relevant to the current study, and the fourth is not applicable; because you evaluated the PA and dietary habits as main component of lifestyle.
R: We agree with the reviewer about the non-relevance of the third and fourth points. We have now deleted those points and added an extra one on generalizability of results.

5) Introduction: Please provide more data and relevant literature regarding the possible clinical pathways between NAFLD and diabetes progression in future.
R: As suggested by the reviewer, we included in the introduction new relevant literature about the clinical association between NAFLD and diabetes progression.

6) Please enrich the relevant literature about the studies evaluated the association between NAFLD and diabetes incidence particularly among prediabetic patients.
R: More information on the association between NAFLD and diabetes in prediabetic patients was added in the introduction.

7) Please be clearer about your study novelty, particularly among this specific population.
R: We have now clarified the importance and novelty of our study conducted in a cohort of workers with prediabetes.

8) Methods
All variables you have reported in results section, should be introduced completely beforehand in methods section (in relevant subsections), which instrument was used for PA, and how about its validity and reliability? I do not see sound and reference-based matters about dietary habits in methods section! please explain it and its instrument.
R: We administered ad hoc questionnaires that include questions about adherence to diet and PA WHO recommendations.

Reference for FPG>126, is needed.
R: The reference has been added.

Statistical analysis and results
Statistical analysis in main area has major defects and results based on accordingly. Please see and follow my points in this regard and revise results section extensively based on. First of all, how you have evaluated normality of continuous variables? Your study is prospective cohort then you should have the time of affecting by diabetes for each subject and you Kaplan -Meier, crude cox regression and multivariable cox regression and report the HR and 95% CI for HR. during model fitting you should provide the crude HR for FLI, as your study predictor and you should consider the lowest level as refence category and report the HR for other categories. Adjustment should be made for age, gender, PA, dietary habits and smoking and report the adjusted HR in the presence of mentioned confounders. It is suggest to use predefined cut points for FLI, and use them in regression models, as you did for evaluated the FLI score diagnostic accuracy for diabetes incidence and cut point you found with best predictive value, I strongly suggest to use the predefined cut points that you have mentioned them in methods section for fitting new cox regression models based on above suggested approaches too.
R: We thank the reviewer for the valuable comments. The main analysis has been changed according to the suggestions. We have now used predefined cutoff points for FLI along the manuscript.

Reporting the incidence rate in these categories are more informative Subgroup analysis by gender is strongly recommended, please do it and report both incidence rate and the above suggested regression analysis. strangely I see the protective role for current smoking?!! The reported OR for age in crude is wrong, although as stated Kaplan -Meier and COX should be used.

R: Following the reviewer's comments, we performed subgroup analysis by gender. We reported incidence rate, as well as Cox regression results expressed as HR, for men and women. According to the Cox regression there is a statistically significant difference between genders.
As for the apparently protective effect of smoking on progression to diabetes, we were also surprised. By reading the literature however we believe that this could be due to the anorexigenic effect of tobacco, more than tobacco consumption itself. We discussed this point in the discussion section of the manuscript.

Please footnote the used statistical tests in table 1.
R: We have added a footnote in table 1 about the statistical tests.

You should also a new table for reporting and comparing the variables in table 1 between diabetic, prediabetic and maybe normal glucose tolerance (because prediabetes is reversable condition); this table is necessary. Title table is not correct, people with FLI >60 and FLI <60 are necessarily affected by NAFLD!
R: We thank the reviewer for the comment, however, we already added new tables showing results by gender, and since the main aim of the study is to evaluate the progression of prediabetes to diabetes according to FLI scores, we prefer not to create further tables, which could possibly confuse the reader.
The table title has been corrected. Please note that Table 1 has been replaced with Tables 2 (for men) and 3 (for women).

Study limitations needs revisions, lifestyle variables have been evaluated in your study! Your study results are not generalizable to general population. How about the job related confounders such as job stress.... please highlight these points as main limitations.
R: We thank the reviewer for the recommendations. We have reviewed the section on study limitations and clarified all points.

Reviewer: 2
Dr. Xavier Mundet Tuduri, Catedra UAB-Novartis de Docencia e Investigación en Medicina de Familia, Universitat Autònoma de Barcelona
Competing interests of Reviewer: None declared
Comments to the Author:
a) The authors should describe the criteria of smoker, dietary habits, and physical activity. Did they use any specific questionary?
R: Smoking habits were collected according to WHO criteria. This information was not in the text and we thank the reviewer for pointing out the omission. We have now added it in the text.
As for PA and dietary habits, we used ad hoc questionnaires that included questions about adherence to diet and PA WHO recommendations.

b) The authors have to explain why they consider that a patient progress to type 2 DM only with one FPG determination.
R: We thank the reviewer for the comment. With only one measurement of FPG available, the possibility to regression toward the mean phenomenon can take place, thus possibly affecting the regression rate. We included such limitation in the text.

c) It would be useful to describe the association of FLI with HbA1c criteria for Type 2 DM.
R: We agree with the reviewer that analysis including HbA1c would add valuable information. Unfortunately, this variable was not collected for all subjects. The study sample includes data from occupational routine health assessments, during which HbA1c measurements are done on workers with previously identified elevated FPG. Furthermore, HbA1c was not collected at 5-years follow up.

d) The patients studied were working adults. Justify if they are representative of general population or consider a limitation
R: We thank the reviewer for the comment. We considered the above point as a limitation and discussed it in the Strengths and limitations section.

Reviewer: 3
Dr. Hideaki Miyoshi, Hokkaido University
Competing interests of Reviewer: None declared
Comments to the Author:
This is a prospective cohort study, with 5-years follow-up to evaluate the association between NAFLD and the development of T2D in a large cohort of South-European Mediterranean workers with prediabetes. They indicated the FLI was the strongest predictor of progression to T2D and the optimal cut-off score for maximum accuracy for FLI was 59.5 with high sensitivity and specificity. The authors proposed that FLI might be useful in routine clinical practice as an additional screening tool to identify subjects with prediabetes who were at high risk of progression and could benefit from early interventions. Although the study was well organized and the results were reasonable, there are several comments for revision.

Major
1. What is the primary endpoint of this prospective cohort study in the study protocol at first? The authors should include it in the Methods section.
R: According to the reviewer suggestion, we included the primary endpoint of our study in the Methods section.

2. Excess intake of fruits is also known as the risk of NAFLD. The analysis of daily fruits and vegetables should be divided and reanalyzed each.
R: We agree with the reviewer that excessive fruits intake can increase the risk of NAFLD. Unfortunately, we cannot discern intakes of fruit from that of vegetables as during data collection patients were simply asked whether they consumed “fruits and vegetables” daily. We believe this is a limitation and discussed it the in Strengths and limitations section.

3. OGTT is more informative and important compared with just measuring FPG. Please show the sources for evidence regarding the sentence: ‘(OGTT), which is considered more sensitive but less specific than FPG for identifying people at risk of developing T2D. (page 15)’. What’s the ‘specific’?
R: We thank the reviewer for the comment. We clarified the sentence in the manuscript and added two references to support this information.

Minor
1. The way of writing for ‘progression to T2D’ is mixed-up (T2D conversion, development of T2D,,,). That should have consistency in the manuscript. ‘progression to T2D’ would be the best. ‘prediabetes reversion’ should be arranged (page 14).
R: According to the reviewer’s comment, we corrected this inconsistency and replaced all the former expressions by ‘progression to T2D’.

2. The sentence should be arranged: ‘This would benefit patients at greater risk for T2D, allowing more careful monitoring and providing an opportunity for early interventions to prevent and reduce both the progression of hepatic disease. (line 7-9, page 15)’
R: We thank the reviewer for the useful suggestion. We changed the sentence accordingly.

3. Discussion is bit too long. Please focus more.

R: We agree with the reviewer's comment. We have reduced the length of the discussion and rephased some parts to give it more focus.

VERSION 2 – REVIEW

REVIEWER	Feizi, Awat Isfahan University of Medical Sciences, Department of Epidemiology and Biostatistics, School of Public Health, Isfahan University of Medical Sciences, Isfahan, Iran
REVIEW RETURNED	09-Jul-2021

GENERAL COMMENTS	Dear authors, Thanks for good jobs did on the requested revisions, majority have been addressed. However, some points should be addressed All data in table 2-4 should be presented in total sample beside of genders. i.e. you have a table such as tables 2 and three for total sample to, and it should be presented before 2&3, and in table 4 you should have column for total sample similar as those you have for both genders you should add a straight line (regression line) to figure scatter plot. Current smokers has lower risk of progression to diabetes!?

REVIEWER	Miyoshi, Hideaki Hokkaido University, Division of Diabetes and Obesity, Faculty of Medicine and Graduate School of Medicine
REVIEW RETURNED	14-Mar-2021

GENERAL COMMENTS	The authors appropriately responded to my comments in the revision. Minor 1. Need a comma between FPGA and blood pressure in the sentence newly added in Abstract. There are several minor typos in the manuscripts.

VERSION 2 – AUTHOR RESPONSE

Reviewer: 3

Dr. Hideaki Miyoshi, Hokkaido University

Comments to the Author:

The authors appropriately responded to my comments in the revision.

Minor

1) Need a comma between FPGA and blood pressure in the sentence newly added in Abstract. There are several minor typos in the manuscripts.

R: We are grateful for the suggestion. We added the missing comma, and we corrected the typos in the manuscript.

Reviewer: 1

Dr. Awat Feizi, Isfahan University of Medical Sciences

Comments to the Author:

Dear authors,

Thanks for good jobs did on the requested revisions, majority have been addressed.

However, some points should be addressed

1) All data in table 2-4 should be presented in total sample beside of genders. i.e. you have a table such as tables 2 and 3 for total sample to, and it should be presented before 2&3, and in table 4 you should have column for total sample similar as those you have for both genders

R: Following the reviewer's comments, we present all data in table 2-4 in relation to the total sample. We added a new table (table 2) for total sample. Accordingly, we renamed table 2 as table 3, table 3 as table 4, and table 4 as table 5. We added a column for total sample in former table 4 (renamed as table 5).

2) you should add a straight line (regression line) to figure scatter plot.

R: We previously added a regression line to the scatter plot in the main manuscript. We have colored the regression line in red in order to highlight it.

3) Current smokers has lower risk of progression to diabetes!?

R: We already justified in the main text the alleged protective effect of smoking on progression to diabetes due to the anorexigenic effect of tobacco, more than tobacco consumption itself, according to scientific reported data (please, see page 17, paragraph 3).