**RESPONSE TO REVIEWERS**

Dear Editor,

Authors would like to thank Reviewers on very constructive comments.

All corrections in the Manuscript text proposed by Reviewers are marked by track changes option, while in this document are marked by blue colour letters.

Further in the text are responses to all reviewers' comments.

**Reviewer: E**

General notifications:
- The manuscript needs to be checked by native speaker, because linguistic
quality is unsatisfactory. Some sections are hard to understand.

**Auhtor response:**

Manuscript is corrected by English native speaker

Abstract is rewritten.

**Reviewer E:**

In the experimental section more details are required.

**Auhtor response:**

Spelt flour was used, so in the experimental section following text is changed to:

For whole meal bread production, flour from spelt, grown in the year 2018 in Serbia was used,

**Reviewer E:**

This lipid content seems to be very low. In most studies this value is about 2%. Why this value is so low?

**Auhtor response:**

This value of lipid content of spelt flour is determined as incorrect. New set of samples of whole meal spelt flour and bread were subjected to the lipid content determination. New data is entered in the tables.

**Reviewer E:**

Since there is a similar characteristic of spelt and yeast extract it woult be better to put this results into a table.

**Auhtor response:**

New table 1 is formed. Other tables are renumbered

**Reviewer E:**

This reference does not refer to AACC method.

**Auhtor response:**

Reference 17 is changed to:

1. AACC. Approved Methods of Analysis *(11th ed.). St. Paul: MN: AACC International,* Method No. 10-09.01 (1999).

**Reviewer E:**

Please add more details about bread ingredients, such as amount of flour, water and yeast. How the dough was prepared? The fermentation time, temperature, humidity of the proofing chamber, are also required. As well as the baking procedure.

**Auhtor response:**

Following text is added:

Bread was baked according to the slightly modified AACC method 18. The composition of bread dough was: spelt flour (in amount of 100g), yeast extract, salt and sugar (in amount determined by experimental plan in table 2), instant dry yeast, ascorbic acid and water in amount of 0.5 g, 0.6 ml (1g ascorbic acid in 100 ml water) and 60 g, respectively.

Table 2 shows design matrix of varied bread formulas, with coded values, where -1, 0 and +1, represents added quantities of yeast extract, salt and sugar in amounts of 0, 2 and 5; 1.0, 1.5 and 2; 0, 5 and 10% d.m. on flour basis, respectively.

bread formulas with varied added quantities of yeast extract, salt and sugar.

Measured ingredients were placed in farinograph bowl and mixed until peak time, previously determined by farinograph curve. Temperature ingredients yialded final dough temperature of 30°C. After mixing dough was rounded by hand and placed in the fermentation cabinet at 30°C. Bulk fermentation lasted 150 minutes, while punching was done by hand, after 60 and 120 minutes from the beginning of the bulk fermentation. After bulk fermentation, dough was moulded by hand and placed in greased pan. Proofing was done at 32°C, at relative humidity of 75%, during 60 minutes. Baking was done for 25 minutes, at 230°C, at humidity provided by the oven.

**Reviewer E:**

Please add more details about the scale. 10 points are for the best quality, and 1 for poor?

**Auhtor response:**

Following text is added:

(1 – lowest, 10 - highest intensity of descriptors)

High values of different descriptors does not mean high quality of bread. It depends of the type of descriptor, if is it favourable sensory characteristic.

**Reviewer E:**

What kind of test was used?

**Auhtor response:**

Following text is added:

Tukey HSD

**Auhtor response:**

Following sub-section is added to the Experimental section:

*Z-Score analysis*

Z-Score analysis uses min-max normalisation of bread quality parameters transforming them from their original unit system in new dimensionless system where further mathematical calculations with different types of quality parameters are applicable 25. Maximum value of normalised score presents optimum value of all combined analysed parameters, indicating on optimum total quality:

, *k*=1-3, *j*=1-2 (3)

where *xk* are: proteins, starch, and cellulose, and *xj* are: fat, total sugars;

, *l*=1-5 (4)

where *xl* are: Zn, Cu, Mg, Ca and Fe;

, *m*=1-5 (5)

where *xm* are: L\*, a\*, b\*, C\* and bread crumb quality;

, *n*=1-8, *o*=1-8 (6)

where *xn* are: characteristic appearance, taste and aroma, crust colour intensity, colour uniformity, sweet taste, elasticity and pores uniformity, and *xo* are: crumb colour intensity, sour and salty taste, sour, yeast and pungent aroma, firmness and wall thickness;

, *p*=1-4 (7)

where *Sp* are *S1*, *S2*, *S3* and *S4*;

$max\left[Score\_{i}\right]\rightarrow optimum$ (8)

**Reviewer E:**

The section “RESULTS AND DISCUSSION” is mostly composed of result
description. Appropriate discussion should be added. Mostly I am interested
in knowing how yeast extract affect bread preparation, fermentation and
baking. Does it affect bread volume and other basic parameters? Discuss it
with already published literature.

**Reviewer E:**

What kind of reaction occurs to make changest in bread colour? Add some discussion.

**Auhtor response:**

Due to the increased Fe content of bread samples with yeast extract addition (table 4), positive effects of yeast extract on bread colour characteristics can be probably attributed to catalyzation of oxidative reaction of ascorbic acid to dehydro-ascorbic acid 11, thus acting as dough improver.

**Reviewer E:**

How yeast extract influence bread fermentation and overall quality of crumb? What kind chemical reactions are observed in the matrix? Discuss results with literature.

**Auhtor response:**

While addition of sugar statistically insignificantly decreased L\* and increased C\* values, probably related to requirements for the initiation of colour formation during bread baking 28.

Together with proposed mechanism of dough improvement by increased Fe content, yeast extract (containing dead yeast cells) acts as yeast food source 11, improving fermentation tolerance, loaf volume and overall bread crumb quality.

**Reviewer E:**

Add more detail

**Auhtor response:**

The interaction of salt with dough components such as gluten is very important due to inhibitory effect on proteolytic enzymes and direct interaction with flour proteins, providing better dough handling and oven spring 11,16. Proposed effects are contributing to higher bread crumb quality, as it can be seen form the statistically significant increase of the bread crumb quality results in the samples with higher salt addition (comparison of the samples 5 and 8; 10 and 13, and also from the figure S15a).

**Reviewer E:**

What about other changes, for example between samples 0 and 5? There is a significant change with the same addition of salt and sugar.

**Auhtor response:**

In the text section:

Analyzing yeast extract, as a salt substitution in the bread formulas, on texture characteristics, by comparing bread samples 4 and 10, it can be seen that addition of yeast extract only statistically insignificantly decreased bread crumb quality (for about 2.5 %).

the intent was to analyze only the effect of yeast extract, as a salt substitution on bread crumb quality, hence the choosing these samples for comparison (4 and 10, with different quantities of added salt), where samples 0 and 5 have the same quantity of added salt, and the effect of yeast extract addition as a salt substitution can not be analyzed.

**Reviewer E:**

Please discuss this in more details and add references.

**Auhtor response:**

Following text is rewritten:

Bread samples with sugar added in the highest quantities to their recipes had the highest bread crumb quality values, figure S15b, indicating that sugar acted as improver of textural characteristics of bread with added yeast extract. Added sugar promoted vigorous yeast fermentative activity, contributed to delayed gelatinization of starch and protein denaturation, consequently improving oven spring and bread crumb and pores quality 11. In case of bread crumb quality model, only linear term for sugar has shown statistical significance.

**Reviewer E:**

The column with “bread crumb quality” from table 4 should be
transferred to the Table 5 or the table caption must be changed

**Auhtor response:**

Table 4 caption is changed to:

Average values and standard deviations of the instrumental colour and bread crumb quality analysis of the bread with yeast extract.

**Reviewer E:**

What about other samples?

**Auhtor response:**

Following section is added and modified:

Lysine from yeast extract could be the major source of primary amines in proteins in Maillard reactions of condensation between reducing sugars and amino acids 28, providing the increased crumb colour intensity of the bread samples containing yeast extract.

By comparing the effectiveness on sensory characteristics of yeast extract as a salt substitution in the bread samples recipes, by analyzing bread samples 4 and 10; 1 and 7; 3 and 6; it can be seen that addition of yeast extract improved appearance descriptors, without high deterioration of texture descriptors.

In the Maillard reactions the type of flavor compound formed depends on the type of sugars and amino acids involved 28, so the taste of the samples with yeast extract became more complex, but without increasing salty taste.

**Reviewer E:**

Conclusions should be rewritten, pleas add more details in there, such as
which recipe obtained in general best scores in sensory evaluation.

Rewrite this section, adding more details, such as which bread with the added yeast extract was the best

**Auhtor response:**

Conclusions are rewritten, following text is added:

Although addition of yeast extract deteriorated overall sensory characteristics, combination of sugar and yeast extract addition, improved most sensory characteristics, enhancing overall acceptability of bread samples.

Bread samples with addition of 5% of yeast extract, 1.5% of salt and 0% sugar were determined as the best from the aspect of overall quality.

**Reviewer E:**

Statistical analysis should be revised. In my opinion Tables S1-S4 are not
necessary, and the description in the section of “RESULTS AND
DISCUSSION” does not add any value. At the same time, Tables S5-S8 are not
discussed well enough.

**Auhtor response:**

Tables S1-S4 are deleted from supplementary material, and former Tables S5-S8 are supplemented with R2 values and critical values.

**Reviewer E:**

RSM is a method with a great potential to optimize
parameters. When prepared with appropriate care it should give the answer to
researchers about optimal addition of tested additives (yeast extract, salt,
sugar). Therefore, this section should be rebuilt.

Change this section according to my comments in the Word file

**Auhtor response:**

New literature citations are added:

1. G. Dąbrowski, S. Czaplicki, I. Konopka, *‎J. Food Compos. Anal*., **83**, (2019) 103261 (<https://doi.org/10.1016/j.jfca.2019.103261>).
2. E. Purilis, *J. Food Eng.* **99** (2010) 239-249 (https://doi.org/[10.1016/j.jfoodeng.2010.03.008](http://dx.doi.org/10.1016/j.jfoodeng.2010.03.008))

Whole results and discussion section is rewritten and discussed together with supplemented figures.

Z-Score analysis is added, in purpose of determining optimal quantities of additions from the aspect of total quality.

**Reviewer E:**

You have to add a table with, coded values of your samples and design matrix, using coded values.

**Auhtor response:**

Following text is added:

Table 1 shows design matrix of varied bread formulas, with coded values, where -1, 0 and +1, represents added quantities of yeast extract, salt and sugar in amounts of 0, 2 and 5; 1.0, 1.5 and 2; 0, 5 and 10% d.m. on flour basis, respectively.

Table 1 is rewritten.

**Reviewer E:**

Also, add graphical presentation with response surface. It is well described
in Dąbrowski, G., Czaplicki, S., & Konopka, I. (2019). Fractionation of
sterols, tocols and squalene in flaxseed oils under the impact of variable
conditions of supercritical CO2 extraction. Journal of Food Composition and
Analysis, 83, 103261.

**Auhtor response:**

Graphical presentations of modeled dependencies of tested responses from yeast extract, salt and sugar addition, are added to the supplementary material, due to their number (there are 31 responses and for each there are 2 graphics).

**Reviewer: G**

This is paper presents very extensive research, with a lot of experimental
and modeling data.
I found it appropriate for publication in the Journal of the Serbian
Chemical Society, but only after some minor modifications and clarification
from the Authors. The paper is well written and the topic is appropriate for
the journal.

The aim of the paper is well described and the discussion was well
approached, its results and discussion are correlated to the cited
literature data. The novelty of the work is clearly demonstrated.

The significance of the Work: Given a large number of analyzed data, this is
an interesting study with a significant impact in this area.

Statistical interpretation of the analytical is properly presented.

Other Specific Comments: The work is properly presented in terms of the
language. The work presented here is very interesting and well done, it is
presented in a compact manner.

Some typographic errors should be corrected before the acceptance of the
paper.

**Auhtor response:**

Manuscript is corrected for all typographic errors

Some specific comments are written within the attached document.

REPORT:
        Some typographic errors should be corrected before the acceptance of the
paper.

Some specific comments are written within the attached document.

In my opinion, this manuscript should:
        be published after minor revision without additional review

If manuscript is suitable for publishing, referees recommendation :
        Original scientific paper

**Reviewer G:**

Introduction lacks evidences about nutritive attributes of yeast extract proteins. Due to their composition they could be a valuable enriching supplement. This beneficial composition of yeast cells should be also stressed in the Conclusion.

**Auhtor response:**

Following text is added to the introduction:

Comparing composition of yeast protein and muscle protein, in terms of essential amino acids composition, results revealed striking similarity. Yeast protein is shown to contain all of the essential amino acids and essential amino acids and to be a biologically complete protein 11.

Following text is added to the conclusion:

Bread samples with added yeast extract were characterized by improved nutritional profile, due to **increased high quality protein** from yeast, Zn, Mg and Ca content.

Following text is added to the refernces:

E. J. Pyler, L. A. Gorton, Baking Science and Technology, Sosland Publishing Company, Kansas City, USA, 2008, p. 272-295.

**Auhtor response:**

Section “Извод“ is correctted and rewritten.

Best regards,

Corresponding author:

Prof. Vesna Vučurović, PhD

In Novi Sad, Serbia

3.12.2019.