Manuscropt**ID**: 7174

**Title**: “Extraction of polyphenols and nicotine and cellulase production using tobacco waste”

**Reviewers G comments**:

The paper deals with (a) the optimization of the microwave-assisted extraction of polyphenols and nicotine from a tobacco wasteusing the response surface methodology (RSM) combined a central composite design (CCD) with respect to the liquid/solid ratio (v/w) and extraction time;(b) the effect of the fermentation pretreatment of the tobacco waste by *Streptomyces fulvissimus*CKS7) the effect of fermentation by *Streptomyces* 199 *fulvissimus*CKS7 on the extraction yields of polyphenols and nicotine; and (c) the cellulase production from the pre-fermented and unfermented residues by *Paenibacilluschitinolyticus*CKS1.The paper presented new experimental data.

The paper is recommended for publication after major revision.

English must be improved.

The authors would like to thank the Reviewer G for the suggestions. The manuscript language has been corrected, as appropriate. All of misspells and uncertainties have been checked in detail, thus were cleared in the revised manuscript.

Title:

1. It should be clearer (in the present form, it might mean that the extraction of polyphenols and nicotine was done using tobacco waste). Maybe, „Production of polyphenols, nicotine and cellulase from tobacco waste“.

The authors have reviewed the suggestions of the reviewer about the title of the manuscript and suggest the following correction: „Extraction of polyphenols and nicotine and the production of cellulase using tobacco waste“.

Running title:

1. The authors are suggested to include the term „tobacco“, for instance: FROM TOBACCO WASTE TO VALUE-ADDED PRODUCTS.

The authors agree with the reviewer suggestion and propose the following running title (taking into account the maximum of 5 words as prescribed in the Journals instructions for authors): VALUE-ADDED PRODUCTS FROM TOBACCO WASTE.

Abstract:

1. The first sentence is superfluous and should be deleted.

The whole abstract has been corrected and rephrased.

1. The extracting solvent should be mentioned at an appropriate place.

Done, the extracting solvent was added in Abstract section (page 1, line 19).

Introduction

1. Please place reference #1 at the more proper location, i.e. „nicotine,1“.

Done, the reference was placed at the appropriate place.

Experimental:

1. This part should be reorganized in order to make the experimental work clearer. For instance, 1) Materials: a) Tobacco materials; b) Chemicals; 2) Polyphenols and nicotine extraction: a) Equipment and procedure, b) Optimization of extraction (or Experimental design); 3) Pretreatment by bacterial fermentation; 4) Cellulase production; and 6) Analytical methods: a) Determination of total polyphenols content, b) Determination of nicotine content, c) Cellulase assay.

Done, experimental section was reorganized in suggested order.

1. The origin of the used tobacco waste should be described more specifically. The used grinder and screens should be defined.

The authors provided as much information about the tobacco waste as manufacturer allowed. The sample material was collected from IQOS units, originated from Philip Morris International company. Further treatment of the material was described in detail, as recommended by the reviewer.

1. The sentence „This residue was used as a low-cost material for nicotine and polyphenol extraction and bacterial solid state fermentation without pretreatment“ should be left out or moved to a proper place.

The sentence has been rephrased.

1. The used chemicals should be specified (producers and purity should be given).

Done, the used chemicals (with producers and purity) were listed in chemicals section.

1. Bacterial fermentation experiments: The reasons for selecting the residues obtained in the extraction runs 1, 2 and 11 should be explained.

Done. The runs 1, 2 and 11 were selected randomly and it is mentioned in page 4, line 116.

1. Bacterial fermentation experiments: The extraction of polyphenols and nicotine after bacterial pretreatment should be explained more clearly because the last sentence is too general. For instance, what the liquid/solid ratio and extraction time were applied.

The extraction of polyphenols and nicotine after bacterial pretreatment has been performed in the same manner as for unfermented samples, as described above, thus it was referenced to that section, in order to avoid the repeating of the procedures. Also, the liquid/solid ratio and extraction time were referenced to Table 2.

Results and discussion:

1. Table 2: Only significant digits should be given. It seems to me that three significant digits are too many.

Done. The values in Table 2 are presented with two significant digits.

1. Eqs. (1) and (2): It should be stated whether the independent variables are coded or uncoded. One can conclude that these equations are based on the actual A and B values, whereas they are based on the coded values. Also, Eq. (1) should be checked as it gives values larger for an order. For instance, for (A,B) = (-1, -1), the calculated value of Y1 is 635 mg/g.

The independent variables are based on coded values, and it has been stated in line 171, page 6. According to the Eq. 1, it was found that the mistake occurred, thus it has been now corrected. Also, during the calculation of the equations, the standard deviation value should be included, in order to obtain the correct result.

1. The authors are expected to give the physical explanations of the effects of individual process factors and their interactions on the responses.

The authors provided additional physical explanations about the effects of individual process factors and their interactions on the responses in the Second revised manuscript, pages 8-9, lines 195-200 and 203-209.

1. Values of the coefficients of determination should be given with three digits.

The suggestion has been adopted and corrected in lines 176 and 179, page 7.

1. Table 3: “s” and “ns” should be explained in the footnote of the table. In the heading, P-value should be *p*-value.

Done. The subscriptions were explained in the footnote of the Table 3.

1. Table 5: The authors are suggested to replace “Tobacco residues before extraction” with “Tobacco waste”

The suggestion has been adopted.