Supplement

Utilization of Industrial *Rosa damascena* Mill. By-products and Cocoa Pod Husks as Natural Preservatives in Muffins

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| Compound           | RI  | RDCO2       | CPH       |
|--------------------|-----|-------------|-----------|
|                   |     | % of TIC 926 ± 0.21 | % of TIC 7.06 ± 0.33 |
| Hexanal            | 800 | -           | 9.26 ± 0.21 |
| α-Pinene           | 940 | 0.80 ± 0.05\(^a\) | 7.06 ± 0.33\(^a\) |
| Benzaldehyde       | 961 | -           | 1.42 ± 0.08 |
| Sabinene           | 976 | -           | 7.12 ± 0.12 |
| β-Pinene           | 980 | 0.57 ± 0.06\(^a\) | 2.53 ± 0.010\(^a\) |
| β-Myrcene          | 991 | 0.31 ± 0.10  | -         |
| 3-Octanol          | 993 | -           | 0.60 ± 0.06 |
| 2-Octanol          | 998 | -           | 0.34 ± 0.08 |
| α-Phellandrene     | 1005| -           | 7.40 ± 0.15 |
| α-Cymene           | 1022| -           | 2.85 ± 0.13 |
| Limonene           | 1029| -           | 1.50 ± 0.10 |
| β-Phellandrene     | 1031| -           | 5.32 ± 0.18 |
| γ-Terpinene        | 1062| 0.83 ± 0.08  | -         |
| Terpinolene        | 1087| 0.62 ± 0.11  | -         |
| β-Linalool         | 1097| 2.47 ± 0.10\(^a\) | 3.25 ± 0.21\(^a\) |
| Phenethyl alcohol  | 1110| 17.22 ± 0.16 | -         |
| cis-Rose oxide     | 1112| 0.40 ± 0.07  | -         |
| trans-Rose oxide   | 1127| 0.23±0.04   | -         |
| Terpin-4-ol        | 1178| 1.24 ± 0.10  | -         |
| β-Citronellol      | 1228| 6.45 ± 0.15  | -         |
| Nerol              | 1230| 3.47 ± 0.09  | -         |
| Geraniol           | 1255| 2.50 ± 0.12  | -         |
| Thymol             | 1289| -           | 1.62 ± 0.06 |
| Eugenol            | 1356| 0.18 ± 0.04  | -         |
| Geranyl acetate    | 1383| 2.60 ± 0.14  | -         |
| Compound          | RI  | RDCO2          | CPH |
|-------------------|-----|----------------|-----|
| Neryl acetate     | 1365| 1.92 ± 0.18    | -   |
| Methyl eugenol    | 1401| 0.40 ± 0.04    | -   |
| β-Bourbonene      | 1383| 2.90 ± 0.09    | -   |
| β-Cubebene        | 1389| 5.41 ± 0.12    | -   |
| β-Elemene         | 1390| 0.46 ± 0.06    | -   |
| α-Caryophyllene   | 1419| 1.45 ± 0.11    | -   |
| α-Humulene (α-Caryophyllene) | 1454| 0.31 ± 0.07    | -   |
| Germacrene D      | 1479| 0.36 ± 0.09    | -   |
| α-Farnesene       | 1508| 0.52 ± 0.10    | -   |
| β-Bisabolene      | 1510| 0.17 ± 0.02    | -   |
| trans-Nerolidol   | 1564| 4.39 ± 0.06    | -   |
| Spathulenol       | 1575| 1.50 ± 0.14    | -   |
| Caryophyllene oxide | 1580| 0.30 ± 0.07    | 0.15 ± 0.04 |
| γ-Eudesmol        | 1631| 0.27 ± 0.05    | -   |
| β-Eudesmol        | 1649| 0.23 ± 0.06    | -   |
| α-Eudesmol        | 1652| 0.81 ± 0.11    | -   |
| Farnesol          | 1714| 0.31 ± 0.03    | -   |
| n-Nonadecane      | 1901| 16.69 ± 0.16   | -   |
| n-Eicosane        | 2000| 0.12 ± 0.04    | -   |
| 10-Heneicosene    | 2093| 3.51 ± 0.08    | -   |
| n-Heneicosane     | 2100| 5.25 ± 0.15    | -   |
| n-Docosane        | 2200| 0.68 ± 0.18    | -   |
| n-Tricosane       | 2300| 4.09 ± 0.21    | -   |
| n-Tetracosane     | 2400| 1.47 ± 0.14    | -   |
| n-Pentacosane     | 2500| 1.30 ± 0.10    | -   |
| n-Hexacosane      | 2600| 1.30 ± 0.09    | -   |

RDCO2 - waste from CO₂ extracted *Rosa damascena* Mill.

CPH - Cocoa Pod Husks

RI - Relativ Index (Kovats retention index)

% of TIC - percent of Total Ion Current

The results were expressed as mean ±SD (n = 3)

*a, b* - Values with different letters in superscript (a, b) in a column are statistically significant (ANOVA, Tuckey’s post hoc test, *p* < 0.05).

With the letter a are denoted the highest determined value, and with the letter b – the lowest value. The values denoted with different letters (a, b) are different with level of significance *p* < 0.05, meaning that 95 % of the determined results differ.

Table S2 GC-MS analysis of RDCO2 and CPH extracts: non-volatile substances

| Compound          | RI  | RDCO2          | CPH |
|-------------------|-----|----------------|-----|
| L-Valine          | 1228| 0.67 ± 0.12    | 1.83 ± 0.14 |
| Glycerol          | 1266| 0.80 ± 0.14    | 2.91 ± 0.09 |
| L-Leucine         | 1272| 0.44 ± 0.08    | 2.00 ± 0.10 |
| Phosphoric acid   | 1278| 7.56 ± 0.32    | -    |
| L-Isoleucine      | 1299| 0.31 ± 0.04    | 1.55 ± 0.08 |
| L-Proline         | 1307| 1.74 ± 0.09    | 2.88 ± 0.15 |
| Succinic acid     | 1310| 5.02 ± 0.11    | 9.94 ± 0.19 |
| o-Hydroxybenzoic acid | 1326| 7.76 ± 0.12    |      |
| Fumaric acid      | 1355| 1.11 ± 0.06    | 5.10 ± 0.10 |
| Serine            | 1362| 0.81 ± 0.10    | 1.53 ± 0.11 |
| L-Threonine       | 1390| 0.37 ± 0.07    | 1.19 ± 0.09 |
| L-Homoserine      | 1446| 0.19 ± 0.03    | -    |
| Malic acid        | 1488| 8.38 ± 0.15    | 6.68 ± 0.19 |
| Salicylic acid    | 1516| 0.51 ± 0.10    | -    |
| Compound                  | RI  | RDCO2       | CPH       |
|---------------------------|-----|-------------|-----------|
| Pyroglutamic acid         | 1512| 0.63 ± 0.06 | 8.49 ± 0.08 |
| L-Aspartic acid           | 1531| 0.69 ± 0.09 | 7.84 ± 0.12 |
| L-Threonic acid           | 1528| 0.33 ± 0.04 | -         |
| 4-Aminobutyric acid       | 1542| -           | 15.43 ± 0.18 |
| p-Hydroxybenzoic acid     | 1621| -           | 7.66 ± 0.21 |
| L-Glutamic acid           | 1629| -           | 10.41 ± 0.16 |
| L-Phenylalanine           | 1646| 1.17 ± 0.11 | 6.18 ± 0.19 |
| L-Asparagine              | 1682| 0.11 ± 0.02 | -         |
| L-Lysine                  | 1737| 0.17 ± 0.05 | -         |
| Ribonic acid              | 1756| 0.71 ± 0.09 | -         |
| Vanillic acid             | 1758| 0.53 ± 0.10 | 16.11 ± 0.17 |
| Protocatechuic acid       | 1813| 0.65 ± 0.08 | 32.51 ± 0.28 |
| Isocitric acid            | 1839| -           | 7.22 ± 0.17 |
| Quinic acid               | 1843| 0.34 ± 0.07 | -         |
| Fructose isomer           | 1862| 25.06 ± 0.23 | 7.78 ± 0.34 |
| Fructose isomer           | 1868| 38.55 ± 0.41 | 4.49 ± 0.47 |
| Fructose isomer           | 1875| 13.04 ± 0.20 | -         |
| Galactose isomer          | 1884| 26.37 ± 0.19 | 6.55 ± 0.29 |
| Syringic acid             | 1888| 8.65 ± 0.11 | -         |
| Glucose isomer            | 1896| 90.09 ± 0.28 | 17.77 ± 0.34 |
| Galactose isomer          | 1907| 14.42 ± 0.21 | 3.67 ± 0.35 |
| Glucose isomer            | 1916| 12.63 ± 0.19 | 13.40 ± 0.30 |
| p-Coumaric acid           | 1920| -           | 15.91 ± 0.18 |
| Glucitol                  | 1930| 64.65 ± 0.24 | 9.72 ± 0.29 |
| Gallic acid               | 1968| 35.96 ± 0.18 | -         |
| Gluconic acid             | 1991| 26.57 ± 0.26 | -         |
| Palmitic acid             | 2039| 5.61 ± 0.17 | 8.46 ± 0.26 |
| Glucaric acid             | 2069| 10.58 ± 0.15 | 8.75 ± 0.24 |
| Ferulic acid              | 2069| -           | 23.16 ± 0.31 |
| Myo-Inositol              | 2090| 19.42 ± 0.20 | -         |
| Stearic acid              | 2132| 0.99 ± 0.14 | 7.43 ± 0.19 |
| Caffic acid               | 2140| 4.61 ± 0.16 | 16.32 ± 0.24 |
| Linoleic acid             | 2209| 8.41 ± 0.26 | -         |
| Linolenic acid            | 2217| 6.73 ± 0.12 | -         |
| Sucrose isomer; α-D-Glc-(1,2)-β-D-Fru | 2649 | 2.17 ± 0.14 | 39.01 ± 0.31 |
| Sucrose isomer; α-D-Glc-(1,2)-β-D-Fru | 2660 | 4.24 ± 0.10 | 15.76 ± 0.29 |
| Sucrose isomer; α-D-Glc-(1,2)-β-D-Fru | 2674 | 1.31 ± 0.11 | -         |
| Catechin                  | 3222| 3.64 ± 0.14 | 55.38 ± 0.41 |
| Epicatechin               | 3228| -           | 45.71 ± 0.21 |
| Stigmasterol              | 3315| 1.91 ± 0.12 | -         |
| β-Sitosterol              | 3355| 2.05 ± 0.13 | -         |

RDCO2 - waste from CO\textsubscript{2} extracted \textit{Rosa damascena} Mill.
CPH - cocoa pod husks
RI - Relativ Index (Kovats retention index)
% of TIC - percent of total ion current
The results were expressed as mean ±SD (n = 3)

\*\* - Values with different letters in superscript (a, b) in a column are statistically significant (ANOVA, Tuckey’s post hoc test, *p* < 0.05).
With the letter a are denoted the highest determined value, and with the letter b – the lowest value. The values denoted with different letters (a, b) are different with level of significance *p* < 0.05, meaning that 95 % of the determined results differ.
Table S3 Inhibition of microorganisms’ development in presence of extracts of RDCO2 and CPH waste

| Microorganism                        | Control | RDCO2 | CPH |
|--------------------------------------|---------|-------|-----|
| **Escherichia coli** ATCC 25922, 1.0 × 10¹² cfu/cm³ | IZ, mm  | 10.0 ± 0.3⁸ | 9.0 ± 0.5⁸ |
|                                      | MIC, µg/ml | 60 | 600 |
| **Proteus vulgaris** ATCC 6380, 5.0 × 10¹¹ cfu/cm³ | IZ, mm  | 10.0 ± 0.3⁷ | 9.0 ± 0.0⁰ |
|                                      | MIC, µg/ml | > 60 | 600 |
| **Pseudomonas aeruginosa** NBIMCC 1370, 7.5 × 10¹⁰ cfu/cm³ | IZ, mm  | 9.0 ± 0.0⁰ | 10.0 ± 0.2² |
|                                      | MIC, µg/ml | 600 | 600 |
| **Staphylococcus aureus** ATCC 25923, 4.0 × 10⁸ cfu/cm³ | IZ, mm  | 11.0 ± 1.0⁰ | 9.0 ± 0.5⁰ |
|                                      | MIC, µg/ml | < 600 | 600 |
| **Enterococcus faecalis** ATCC 19433, 8.0 × 10¹¹ cfu/cm³ | IZ, mm  | 10.5 ± 0.5⁷ | 10.0 ± 0.7⁰ |
|                                      | MIC, µg/ml | 60 | 600 |
| **Listeria monocytogenes** ATCC 19111, 4.9 × 10⁹ cfu/cm³ | IZ, mm  | 17.0 ± 1.0⁰ | 10.0 ± 0.2² |
|                                      | MIC, µg/ml | 600 | 600 |
| **Salmonella abony** NTCC 6017, 2.0 × 10⁸ cfu/cm³ | IZ, mm  | 10.0 ± 0.2⁰ | 9.0 ± 0.5⁰ |
|                                      | MIC, µg/ml | 600 | 600 |
| **Candida albicans** NBIMCC 74, 2.0 × 10¹⁰ cfu/cm³ | IZ, mm  | 9.5 ± 0.5⁰ | 9.0 ± 0.0⁰ |
|                                      | MIC, µg/ml | 600 | 600 |
| **Candida utilis** ATCC 42402, 4.6 × 10⁸ cfu/cm³ | IZ, mm  | 13.5 ± 0.2² | 10.0 ± 0.2² |
|                                      | MIC, µg/ml | < 600 | 600 |
| **Aspergillus niger** ATCC 1015, 1.4 × 10⁷ cfu/cm³ | IZ, mm  | 10.0 ± 0.0⁰ | 9.5 ± 0.5⁰ |
|                                      | MIC, µg/ml | 600 | 600 |
| **Penicillium chrysogenum** ATCC 28089, 1.5 × 10⁷ cfu/cm³ | IZ, mm  | 9.5 ± 0.5⁰ | 9.5 ± 0.5⁰ |
|                                      | MIC, µg/ml | 600 | 600 |
| **Bacillus subtilis** ATCC 19659, 1.0 × 10⁹ cfu/cm³ | IZ, mm  | 10.5 ± 0.5⁰ | 11.0 ± 0.0⁰ |
|                                      | MIC, µg/ml | 60 | 60 |
| **Fusarium moniliforme** ATCC 38932, 1.0 × 10⁷ cfu/cm³ | IZ, mm  | 10.0 ± 0.0⁰ | 11.0 ± 0.2² |
|                                      | MIC, µg/ml | 600 | < 600 |
| **Rhizopus arrhizus** ATCC 11145, 4.0 × 10⁹ cfu/cm³ | IZ, mm  | 9.5 ± 0.5⁰ | 9.5 ± 0.5⁰ |
|                                      | MIC, µg/ml | 600 | 600 |

RDCO2 - waste from CO₂ extracted Rosa damascena Mill.
CPH - Cocoa Pod Husks
IZ - Inhibition Zone
MIC - Minimal Inhibition Concentration
cfu - colony forming units

The results were averaged of 4 repetitions ±SD

⁸, ⁹ - Values with different letters in superscript (a, b) in a column are statistically significant (ANOVA, Tuckey’s post hoc test, p < 0.05).
With the letter a are denoted the highest determined value, and with the letter b – the lowest value. The values denoted with different letters (a, b) are different with level of significance p < 0.05, meaning that 95 % of the determined results differ.

Table S4 Shear stress range (D); yield stress (τ₀), consistency index (k), flow index (n) and coefficient of determination (R²) of muffin batters

| Sample | D, s⁻¹ | τ₀, Pa | k, Pa · sⁿ | n | R², % |
|--------|--------|--------|-------------|---|------|
| C1     | 0.17 ± 3.4 | 5.45 ± 0.11⁷ | 68.53 ± 0.26⁷ | 0.49 ± 0.08 | 99.8 |
| C2     | 0.17 ± 3.4 | 3.26 ± 0.08⁸ | 46.21 ± 0.33³ | 0.43 ± 0.07 | 99.9 |
| V1     | 0.17 ± 3.4 | 3.23 ± 0.10⁰ | 44.31 ± 0.35³ | 0.44 ± 0.06 | 99.9 |
| V2     | 0.17 ± 3.4 | 6.39 ± 0.11⁷ | 51.43 ± 0.29³ | 0.45 ± 0.09 | 99.9 |
| V3     | 0.17 ± 3.4 | 7.18 ± 0.12⁴ | 50.75 ± 0.28³ | 0.51 ± 0.08 | 99.8 |

The results were expressed as mean ±SD (n = 3)

⁷, ⁸, ⁹, ¹⁰ - Values with different letters in superscript (a, b, c, d) in a column are statistically significant (ANOVA, Tuckey’s post hoc test, p < 0.05). With the letter a are denoted the highest determined value, and with the letter d – the lowest value; the others denotes values in between a and d. The values denoted with different letters (a, b, c, d) are different with level of significance p < 0.05, meaning that 95 % of the determined results differ.
Fig. S1 Image analysis of muffins: gas pore area distribution pattern