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Model studies on the Effect of Aldehyde Structure on Their Selective Trapping by Phenolic Compounds

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ABSTRACT

The reaction among flavor-relevant saturated aldehydes (propanal, 2-methylpropanal, butanal, 2-methylbutanal, 3-methylbutanal, pentanal, hexanal, and glyoxal) and phenolic compounds (resorcinol, 2-methylresorcinol, 2,5-dimethylresorcinol, and orcinol) was studied both to identify and characterize the formed carbonyl-phenol adducts and to understand the differences in the carbonyl-trapping abilities of phenolic compounds. The obtained results showed that carbonyl-trapping by phenolics is selective and the formation of carbonyl-phenol adducts depends on the structure of both phenol and aldehyde involved. In relation to the phenolic derivative, the presence of groups that increase the nucleophilicity of phenolic carbons will increase the carbonyl-trapping ability of these compounds. On the other hand, the presence of groups that increase the steric hindrance of these positions without affecting mucleophilia, will inhibit the reaction. Analogously, the presence of branching at position 2 of the aldehyde will also inhibit the reaction by steric hindrance. All these results suggest that addition of phenolics to foods may change food flavor not only because of their sensory properties but also because they can modify the ratio among food odorants by selective reaction of phenolics with determined carbonyl compounds.

KEYWORDS: *Aldehydes, Carbonyl-phenol reactions, Lipid oxidation, Maillard reaction, Phenols, Reactive carbonyls*

INTRODUCTION

Phenolic compounds protect lipids from oxidation by acting both as free radical scavengers[1,2] and chelators.[3,4] In addition, recent studies have shown that these compounds can also act as lipid-derived carbonyl scavengers,[5–7] which would constitute a third protective barrier of phenolic compounds against the consequences of lipid oxidation in foods.[8] This last barrier is still poorly understood, but it might be playing a major role in the flavor changes observed in foods when phenolics are employed for food protection.[9–11]

These changes might be related to the phenolic-trapping of small and very reactive carbonyl compounds responsible for Maillard-type reactions but also to the lesser known scavenging of flavor aldehydes. Among the small and very reactive carbonyl compounds responsible for Maillard-type reactions, the phenolic-trapping ability of glyoxal and methylglyoxal has been widely studied,[12–14] as well as the phenol-trapping ability of phenolics for short chain aldehydes produced by carbohydrate degradation in the course of Maillard reaction.[15] On the other hand, the scavenging of aldehydes that play a major role in the flavor of foods has been lesser studied, although the reaction of phenolics with phenylacetaldehyde[16,17] or propanal[18] has been described.

In an attempt to understand the differences in the carbonyl-trapping abilities of phenolic compounds, this study describes the reactions produced between phenolic compounds and saturated aldehydes. These aldehydes were selected because they are powerful food odorants and a large series of them with very different structures are available.[19] In addition, analogous reactions with glyoxal were also studied for comparison purposes. As model phenolic compounds, single *m*-diphenols were selected because carbonyl-phenol adducts are mainly produced with phenolic compounds having two hydroxyl groups at *meta* positions,[20] and their single structures facilitate both the

characterization of the produced carbonyl-phenol adducts and the later study of the produced adducts by gas chromatography coupled to mass spectrometry (GC-MS).

MATERIALS AND METHODS

Chemicals. A series of aldehydes having three (propanal, **1**), four (butanal, **2**, and 2-methylpropanal, **3**), five (pentanal, **4**, 2-methylbutanal, **5**, and 3-methylbutanal, **6**), and six carbons (hexanal, **7**) were employed in these studies. In addition, glyoxal (**8**) and 2-methyl-2-pentenal (**9**) were used for comparison purposes. As model phenolic compounds, resorcinol (**10**), 2-methylresorcinol (**11**), 2,5-dimethylresorcinol (**12**), and orcinol (**13**) were employed. The chemical structures of all these compounds are given in Figure 1. All these compounds as well as all other chemicals employed in these studies were purchased from Sigma-Aldrich (St. Louis, MO), Fluka (Buchs, Switzerland), or Merck (Darmstadt, Germany).

Study of the Reaction between Alkanals and Phenolic Compounds. In order to determine the reaction pathway between alkanals and phenolic compounds, a preliminary study of the reaction between pentanal and 2-methylresorcinol was carried out. In this, and in other reactions analyzed in this study, methanol was employed as solvent to facilitate the solubility of both aldehydes and phenols. In addition, reactions were carried out at basic pH. Triethylamine was employed for this purpose. In this first study, reaction products were stabilized by acetylation to avoid both reaction reversion and further polymerizations. The reaction was carried out by mixing pentanal (3 mmol) and 2-methylresorcinol (3 mmol) in methanol (10 mL) containing 400 µL of triethylamine and heating the mixture in a closed test tube under nitrogen for 72 h at 60 ºC. At the end of the heating process, the reaction mixture was cooled at room temperature (22 ºC) for 15 min and taken to dryness. The dried sample was acetylated by adding 100 mL of anhydrous pyridine, and 50 mL of acetic anhydride, and left in the dark for 22 h at room

temperature. After that time, 160 mL of water and 160 mL of chloroform were added, and the organic phase was firstly washed three times with 250 mL of 5% hydrochloric acid until the pyridine was removed and, then, with water to remove the hydrochloric acid. The resulting organic extract was treated with sodium sulfate to eliminate the remaining humidity, then taken to dryness, and, finally, fractionated by column chromatography using mixtures of hexane and diethyl ether as eluent to isolate the produced compounds. The total ion chromatogram of the reaction mixture is shown in Figure 2. Compound **14** was the product of acetylation of 2-methylresorcinol (2-methyl-1,3-phenylene diacetate). The other compounds were isolated and characterized by 1D and 2D nuclear magnetic resonance (NMR) and mass spectrometry (MS). Their structures are shown in Figure 3.

4-(1-Methoxypentyl)-2-methyl-1,3-phenylene diacetate (**15**). Yield: 13.8%. Retention time: 11.21 min. ^{1}H NMR ($CDCl_3$): δ (ppm) 0.90 (t, 3, *J* = 7.1 Hz, H-5'), 1.29 and 1.41 (m, 2, H-3'), 1.32 (m, 2, H-4'), 1.62 and 1.75 (m, 2, H-2'), 1.98 (s, 3, CH_3C2), 2.33 (s, 3, CH_3CO), 2.33 (s, 3, CH_3CO), 3.17 (s, 3, CH_3O), 4.15 (dd, 1, *J* = 5.0 Hz, *J* = 8.0 Hz, H-1'), 7.00 (d, 1, *J* = 8.1 Hz, H-6), and 7.26 (d, 1, *J* = 8.1 Hz, H-5). ^{13}C NMR ($CDCl_3$): δ (ppm) 10.11 ($\underline{C}H_3$C2), 13.97 (C-5'), 20.74 ($\underline{C}H_3$CO), 20.79 ($\underline{C}H_3$CO), 22.52 (C-4'), 28.07 (C-3'), 36.63 (C-2'), 56.97 (CH_3O), 79.05 (C-1'), 119.93 (C-6), 123.66 (C-2), 124.97 (C-5), 132.36 (C-4), 148.03 (C-3), 148.89 (C-1), 168.89 (CO), and 168.92 (CO). MS, *m/z* (%, ion structure): 308 (0.02, M^+), 277 (0.02, $M^+ - CH_3O$), 265 (3, $M^+ - CH_3CO$), 251 (50, $M^+ - CH_3CH_2CH_2$), 234 (19, $C_{13}H_{14}O_4$), 209 (76, 251 – CH_2CO), 192 (70, 234 – CH_2CO), and 167 (100, 209 – CH_2CO).

2-Methyl-4-(pent-1-en-1-yl)-1,3-phenylene diacetate (**16**). Yield: 5.2%. Retention time: 11.46 min. ^{1}H NMR ($CDCl_3$): δ (ppm) 0.96 (t, 3, *J* = 7.3 Hz, H-5'), 1.50 (sx, 2, *J* = 7.3 Hz, H-4'), 1.99 (s, 3, CH_3C2), 2.20 (qd, 2, *J* = 7.3 Hz, *J* = 1.2 Hz, H-3'), 2.33 (s, 3,

CH_3CO), 2.37 (s, 3, CH_3CO), 6.18 (dt, 1, *J* = 15.8 Hz, *J* = 6.8 Hz, H-2'), 6.31 (d, 1, *J* = 15.8 Hz, H-1'), 6.94 (d, 1, *J* = 8.5 Hz, H-6), and 7.37 (d, 1, *J* = 8.5 Hz, H-5). ^{13}C NMR ($CDCl_3$): δ (ppm) 10.11 ($\underline{C}H_3C2$), 13.64 (C-5'), 20.49 ($\underline{C}H_3CO$), 20.80 ($\underline{C}H_3CO$), 22.38 (C-4'), 35.37 (C-3'), 119.83 (C-6), 123.10 (C-1'), 123.55 (C-2), 123.94 (C-5), 128.83 (C-4), 133.85 (C-2'), 146.94 (C-3), 148.50 (C-1), 168.58 (CO), and 169.03 (CO). MS, *m/z* (%, ion structure): 276 (17, M^+), 234 (38, $M^+ - CH_2CO$), 192 (100, 234 – CH_2CO), and 163 (97, 192 – CH_3CH_2).

4-(1-Acetoxypentyl)-2-methyl-1,3-phenylene diacetate (**17**). Yield: 3.2%. Retention time: 12.22 min. ^{1}H NMR ($CDCl_3$): δ (ppm) 0.90 (t, 3, *J* = 7.1 Hz, H-5'), 1.24 and 1.34 (m, 2, H-3'), 1.34 (m, 2, H-4'), 1.79 and 1.92 (m, 2, H-2'), 1.98 (s, 3, CH_3C2), 2.04 (s, 3, $\underline{C}H_3COOC1'$), 2.34 (s, 3, CH_3CO), 2.38 (s, 3, CH_3CO), 5.87 (br, 1, H-1'), 6.99 (d, 1, *J* = 8.4 Hz, H-6), and 7.29 (d, 1, *J* = 8.4 Hz, H-5). ^{13}C NMR ($CDCl_3$): δ (ppm) 10.24 ($\underline{C}H_3C2$), 13.90 (C-5'), 20.59 ($\underline{C}H_3COOC1$), 20.82 ($\underline{C}H_3CO$), 21.06 ($\underline{C}H_3CO$), 22.34 (C-4'), 27.78 (C-3'), 34.44 (C-2'), 70.95 (C-1'), 119.92 (C-6), 124.17 (C-2), 125.43 (C-5), 130.46 (C-4), 147.77 (C-3), 149.41 (C-1), 168.60 (CO), and 168.86 (CO). MS, *m/z* (%, ion structure): 336 (0.2, M^+), 277 (1, $M^+ - CH_3COO$), 276 (0.4, $M^+ - CH_3COOH$), 234 (16, 276 – CH_2CO), 192 (100, 234 – CH_2CO), and 163 (62, 192 – CH_3CH_2).

4-Butyl-8-methyl-3-propylchroman-2,7-diyl diacetate (**18**). Yield: 2.9%. Retention time: 13.65 min. ^{1}H NMR ($CDCl_3$): δ (ppm) 0.92 (t, 3, *J* = 7.2 Hz, $C\underline{H}_3CH_2$), 0.95 (t, 3, *J* = 7.0 Hz, $C\underline{H}_3CH_2$), 1.4 (m, 8, $CH_3C\underline{H}_2$, $CH_3C\underline{H}_2$, $CH_3CH_2C\underline{H}_2$, and $CH_3CH_2C\underline{H}_2$), 2.02 (s, 3, CH_3C8), 2.06 (s, 3, CH_3CO), 2.33 (s, 3, CH_3CO), 2.55 (dd, 1, *J* = 5.5 Hz and *J* = 9.9 Hz, H-3), 2.80 (dt, 2, *J* = 5.3 Hz and *J* = 7.7 Hz, $C\underline{H}_2C4$), 2.95 (dt, 1, *J* = 4.8 Hz and *J* = 7.5 Hz, H-4), 6.37 (dd, 1, *J* = 1.3 Hz and *J* = 1.9 Hz, H-2), 6.67 (d, 1, *J* = 8.3 Hz, H-6), and 6.98 (d, 1, *J* = 8.3 Hz, H-7). ^{13}C NMR ($CDCl_3$): δ (ppm) 9.23 ($\underline{C}H_3C8$), 14.01 ($\underline{C}H_3CH_2$), 14.23 ($\underline{C}H_3CH_2$), 20.13 ($CH_3\underline{C}H_2$), 20.83 (CH_3CO), 21.19 (CH_3CO), 23.01

121 ($CH_3\underline{C}H_2$), 27.48 ($CH_3CH_2\underline{C}H_2$), 29.97 ($CH_3CH_2\underline{C}H_2$), 33.83 (C-4), 36.72 ($\underline{C}H_2$C4),
122 37.45 (C-3), 93.90 (C-2), 114.86 (C-6), 118.60 (C-8), 122.55 (C-4a), 127.31 (C-5),
123 148.54 (C-7), 150.45 (C-8a), 169.35 (CO), and 169.64 (CO). MS, *m/z* (%, ion structure):
124 362 (7, M^+), 320 (8, $M^+ - CH_2CO$), 303 (3, $M^+ - CH_2COO$), 302 (1, $M^+ - CH_2COOH$),
125 260 (9, 302 – propene), 259 (22, 302 – $CH_3CH_2CH_2$), 245 (19, 302 – $CH_3CH_2CH_2CH_2$),
126 217 (32, 260 – $CH_3CH_2CH_2$), and 203 (100, 260 – $CH_3CH_2CH_2CH_2$).

127 **Synthesis and Characterization of Saturated Aldehyde-Phenol Adducts.**
128 Reactions were carried out as described above but formed compounds were isolated
129 without acetylation. Briefly, a mixture of the saturated aldehyde (3 mmol) and the
130 phenolic compound (3 mmol) in methanol (10 mL) containing 400 µL of triethylamine
131 was heated under nitrogen at 60 ºC. At the end of the heating process, reactions mixtures
132 were fractionated by column chromatography using mixtures of hexane and diethyl ether
133 as eluent. Different reactions were studied and the formed compounds were isolated and
134 characterized by 1D and 2D NMR and MS. The structures of all these compounds are
135 shown in Figure 3. NMR and MS data for these compounds are given in the Supporting
136 Information. Selected mass spectral data are given in Table 1.

137 The reaction between resorcinol and propanal produced 4-(1-methoxypropyl)benzene-
138 1,3-diol (**19**). The reaction was heated for 48 h and the compound was isolated using
139 hexane-diethyl ether (7:3) as eluent. Retention time: 18.18 min.

140 The reaction between resorcinol and pentanal produced 4-(1-methoxypentyl)benzene-
141 1,3-diol (**20**) and 2-(1-hydroxypentyl)benzene-1,3-diol (**21**). The reaction was heated for
142 72 h and the compounds were isolated using hexane-diethyl ether (3:1) as eluent.
143 Retention time of compound **20**: 20.04 min. Retention time of compound **21**: 18.87 min.

144 The reaction between resorcinol and 2-methylbutanal produced 4-(1-methoxy-2-
145 methylbutyl)benzene-1,3-diol (**22**). This compound has two chiral centers at C-1' and C-

2’. Therefore, it was produced as two pairs of diasteromers, which have been named **22a** and **22b**. Their retention times were 18.64 and 18.86 min. The reaction was heated for 48 h and the compounds were isolated using hexane-diethyl ether (7:3) as eluent.

The reaction between resorcinol and hexanal produced 4-(1-methoxyhexyl)benzene-1,3-diol (**23**). The reaction was heated for 3 h at 100 ºC and the compound was isolated using hexane-diethyl ether (75:25) as eluent. Retention time: 12.08 min.

The reaction between 2-methylresorcinol and propanal produced 4-(1-methoxypropyl)-2-methylbenzene-1,3-diol (**24**). The reaction was heated for 72 h and the compound was isolated using hexane-diethyl ether (9:1) as eluent. Retention time: 18.52 min.

The reaction between 2-methylresorcinol and pentanal produced 4-(1-methoxypentyl)-2-methylbenzene-1,3-diol (**25**) and 2-methyl-4-(pent-1-en-1-yl)benzene-1,3-diol (**26**). The reaction was heated for 72 h and the compounds were isolated using hexane-diethyl ether (85:15) as eluent. Retention time of compound **25**: 20.28 min. Retention time of compound **26**: 20.38 min.

The reaction between 2-methylresorcinol and 2-methylbutanal produced 4-(1-methoxy-2-methylbutyl)-2-methylbenzene-1,3-diol (**27**). Analogously to compound **22**, this compound also has two chiral centers at C-1’ and C-2’. Therefore, it was produced as two pairs of diasteromers, which have been named **27a** and **27b**. Their retention times were 19.05 and 19.27 min. The reaction was heated for 72 h and the compounds were isolated using hexane-diethyl ether (85:15) as eluent.

The reaction between 2-methylresorcinol and 3-methylbutanal produced 4-(1-methoxy-3-methylbutyl)-2-methylbenzene-1,3-diol (**28**). The reaction was heated for 48

h and the compound was isolated using hexane-diethyl ether (85:15) as eluent. Retention time: 19.97 min.

The reaction between 2-methylresorcinol and hexanal produced 4-(hex-1-en-1-yl)-2-methylbenzene-1,3-diol (**29**). The reaction was heated for 24 h at 100 ºC and the compound was isolated using hexane-diethyl ether (80:20) as eluent. Retention time: 13.55 min.

For comparison purposes the reactions of glyoxal with both resorcinol and 2-methylresorcinol, and the reaction of 2-methyl-2-pentenal with resorcinol were also studied. The reaction between resorcinol and glyoxal produced 1-(2,4-dihydroxyphenyl)-2-hydroxyethan-1-one (**30**). The reaction, using a glyoxal-resorcinol ratio of 4:1, was heated for 3 h at 100 ºC and the compound **30** was isolated using hexane-diethyl ether (1:3) as eluent. Retention time: 12.18 min.

The reaction between 2-methylresorcinol and glyoxal produced 1-(2,4-dihydroxy-3-methylphenyl)-2-hydroxyethan-1-one (**31**). The reaction, using a glyoxal-resorcinol ratio of 4:1, was heated for 3 h at 100 ºC and the compound **31** was isolated using hexane-diethyl ether (2:5) as eluent. Retention time: 12.57 min.

The reaction between resorcinol and 2-methyl-2-pentenal produced 2-ethyl-3-methyl-2*H*-chromen-7-ol (**32**). The reaction was heated for 72 h at 100 ºC and the compound **32** was isolated using hexane-diethyl ether (3:1) as eluent. Retention time: 11.65 min.

Effect of Aldehyde Chain Length on the Formation of Alkanal-Phenol Adducts. Mixtures of one phenolic compound (80 µmol) and four lineal alkanals (propanal, butanal, pentanal, and hexanal, 20 µmol of each) in 500 µL of methanol containing 20 µL of triethylamine were heated at 60 ºC under nitrogen. At different reaction times, reaction mixtures were cooled at room temperature (15 min), 15 µL of the internal

standard added (a solution of 54.8 mg of methyl heptanoate in 25 mL of methanol), and studied by GC-MS. The phenolic compounds assayed were resorcinol, 2-methylresorcinol, 2,5-dimethylresorcinol, and orcinol.

Effect of Aldehyde Branching on the Formation of Alkanal-Phenol Adducts. Two different studies were carried out. Firstly, mixtures of 2-methylresorcinol (40 μmol) and two alkanals (butanal and 2-methylpropanal, 20 μmol of each) in 500 μL of methanol containing 20 μL of triethylamine were heated at 60 ºC under nitrogen. In the second study, mixtures of 2-methylresorcinol (60 μmol) and three alkanals (pentanal, 2-methylbutanal, and 3-methylbutanal, 20 μmol of each) in 500 μL of methanol containing 20 μL of triethylamine were heated at 60 ºC under nitrogen. At different reaction times, reaction mixtures were cooled at room temperature (15 min), 15 μL of the internal standard added (a solution of 54.8 mg of methyl heptanoate in 25 mL of methanol), and studied by GC-MS.

GC-MS Analyses. GC-MS analyses were conducted with an Agilent 6890 GC Plus coupled to an Agilent 5973 MSD (mass selective detector, quadrupole type). Separations were carried out on a fused-silica DB5-MS capillary column (30 m × 0.25 i.d.; coating thickness, 0.25 μm) and 1 μL of sample was injected in the pulsed splitless mode. Most working conditions were described previously.[21] Two oven temperatures were programmed. For acetylated derivatives **15**–**18**, the oven temperature was programmed from 100 ºC (1 min) to 300 ºC at 15 ºC/min and, then, 5 min at 300 ºC. For non-acetylated derivatives **19**–**32**, oven temperature was programmed from 40 ºC (1 min) to 240 ºC at 12 ºC/min, then to 300 ºC at 20 ºC/min, and, finally, 5 min at 300 ºC. The flow velocities employed were 30 cm/s for acetylated compounds and 37 cm/s for non-acetylated compounds.

Determination of Carbonyl-Phenol Adducts Content. Isolated carbonyl-phenol adducts were chromatographically pure and were employed for the quantitation of carbonyl-phenol adducts produced in aldehyde/phenol reaction mixtures. The quantitation was carried out by preparing standard curves of the different adducts in the 515 µL of solution prepared for GC-MS injection. Seven different concentration levels of the adducts were used. Adduct content was directly proportional to the aldehyde/internal standard area ratio ($r > 0.98$, $p < 0.001$). The coefficients of variation were less than 10 %. When one adduct was not available, such as the adducts derived from either orcinol or butanal, the calibration curve used was that of the adduct with the closest chemical structure.

NMR Spectroscopy. All NMR spectra were obtained by a Bruker Advance III spectrometer operating at 500 MHz for protons. Acquisition parameters were described previously.[7] For structural determinations, COSY, HMQC and HMBC experiments were carried out.

RESULTS AND DISCUSSION

The Reaction between Phenolic Compounds and Saturated Aldehydes. Phenolic compounds have atoms with a high nucleophilicity that are able to react with the carbonyl carbon of saturated aldehydes as a consequence of the low electron density of this last carbon. In addition, under the conditions required for the formation of carbonyl-phenol adducts, the aldol condensation of saturated aldehydes occurs and the corresponding 2-alkenals are produced. As described previously,[6] phenolics compounds are added to these 2-alkenals at the carbon-carbon double bond of the aldehyde.

Figure 4 proposes a reaction pathway to explain the products formed in the reaction between pentanal and 2-methylresorcinol. As described in the Materials and Methods

section, compounds **15**–**18** were isolated and characterized from acetylated samples and compounds **25**-**26** were isolated and characterized from non-acetylated samples. On the other hand, the acetate of compound **33** was not detected under the employed conditions. Nevertheless, compound **32** (Figure 3), which is the analogous to compound **33** when propanal is involved, was isolated in the reaction between 2-methyl-2-pentenal and resorcinol. The absence of compound **33** is likely a consequence of its instability. Thus, previous studies have suggested that adducts similar to compound **33** are more unstable than adducts similar to compound **37**, and disappear upon prolonged heating.[6]

As shown in Figure 4, under the reaction conditions employed, saturated aldehydes can either react with methanol to produce the corresponding hemiacetal **34** or take place an aldol condensation to produce the corresponding 2-alkenal **35**. Both compounds react then with the phenolic compound. In the case of the hemiacetal **34**, this reaction can be produced with the formation of either methanol or water. If methanol is produced, the carbonyl-phenol adduct **36** is formed. The presence of a hydroxyl group in this adduct makes possible its dehydration to produce the corresponding olefin **26**. After acetylation, adduct **26** produces compound **16**, and adduct **36** produces compound **17**.

If water is formed after the attack of the phenolic compound, the carbonyl-phenol adduct **25** will have a methoxy group and, after acetylation, compound **15** will be produced. The loss of methanol in adduct **25** would also be another route to produce compound **26** in a first step and, after acetylation, compound **16**.

The reaction of the product of aldol condensation **35** with the phenolic compound is more complex because one of the phenolic carbons and, also, one of the phenolic hydroxyl groups are involved.[6] Compound **18** is produced by addition of one phenolic carbon to the carbon-carbon double bond of the product of aldolization followed by the

addition of one phenolic hydroxyl group to the carbonyl carbon to produce the corresponding hemiacetal **37**. On the other hand, compound **33** is produced by addition of one phenolic hydroxyl group to the carbon-carbon double bond of the aldehyde and then addition of one phenolic carbon to the carbonyl carbon to produce a hydroxyl derivative **38**. The dehydration of this compound is the origin of compound **33**. The presence of a carbon-carbon double bond in this compound increases its susceptibility to further reactions, including polymerizations, and explains its relative instability.[6]

Although all these compounds can be produced, some compounds are produced to a higher extent than others depending on the structure of the reactants and the reaction conditions. In addition, some of the reaction products are more stable than others. Figure 3 shows the main reaction products formed in the different reactions. In all these reactions, major reaction products were isolated and characterized. As can be observed in Figure 3, when resorcinol was involved (compounds **19**-**23**), most reactions occurred at the C-4 of the phenolic compound, and only when using pentanal, the corresponding adduct at C-2 could be isolated and characterized (compound **21**). In addition, all adducts involving C-4 were similar and corresponded to the methoxylated adduct (the analogous to compound **25** in Figure 4). Neither the corresponding hydroxylated derivative (the analogous to compound **36** in Figure 4) nor the olefin (the analogous to compound **26** in Figure 4) could be isolated. On the contrary, the adduct at C-2 was the hydroxylated adduct and neither the methoxylated adduct nor the olefin could be isolated.

When 2-methylresorcinol was involved, the methoxylated derivative adduct (the analogous to compound **25** in Figure 4) was also the compound isolated in all assayed reactions with the exception of its reaction with hexanal. In addition, when using this phenolic compound, two olefins were also produced to a significant extent: those corresponding to the reactions with pentanal and hexanal (compounds **26** and **29**,

290 respectively). On the other hand, the hydroxylated derivative could not be isolated in any
291 reaction.

292 All these reactions always implied the formation of a new chiral center and the
293 corresponding racemic mixture was produced. Therefore, when the aldehyde had a chiral
294 center, like in the case of 2-methylbutanal, the formed adduct had two chiral centers and
295 2 pairs of disateroisomers were produced.

296 Glyoxal is a dialdehyde, but it reacts similarly to other saturated aldehydes and the
297 addition of the phenolic C-4 to one of the carbonyl carbons was produced to form
298 compounds **30** and **31**. However, differently to most suggested structures in many
299 previous studies between phenolics and glyoxal, the structure determined by NMR
300 indicated that the carbonyl group was conjugated with the aromatic ring and the adduct
301 was a primary alcohol. The formation of this product implies that an isomerization has
302 occurred. This isomerization also makes it difficult to add a second molecule of phenol
303 to the carbonyl compound. For that reason, adducts involving two molecules of phenolics
304 and one of glyoxal were not isolated.

305 **Effect of Phenol and Aldehyde Structures on the Formation of Alkanal-Phenol**
306 **Adducts.** Above described results showed that alkanals reacted similarly with phenolic
307 compounds and the preferred adduct was formed at position C-4. This is likely a
308 consequence of being C-4 the carbon atom of the phenolic compound with the lowest
309 steric hindrance among those with a high nucleophilicity in the phenolic ring. In order to
310 know the role of the structures of both the phenol and the carbonyl compound on the
311 formation of carbonyl-phenol adducts, the comparative reaction of different phenolic
312 compounds with different linear and branched aldehydes was studied. As shown in
313 Figures 5 and 6 the amount of carbonyl-phenol adducts always increased linearly as a

function of time for the first 96 h of incubation at 60 ºC. However, some adducts were produced to a higher extent than others.

Figure 5 shows the formation of carbonyl-phenol adducts in the reaction between linear alkanals (propanal, butanal, pentanal, and hexanal) and phenolic compounds: resorcinol (Figure 5A), 2-methylresorcinol (Figure 5B), 2,5-dimethylresorcinol (Figure 5C), and orcinol (Figure 5D). Adduct concentration always increased linearly for the first 96 h. Reaction rates were calculated by using the equation

$$[\text{adduct}] = k \cdot t$$

where [adduct] is the concentration of the adduct, k is the rate constant, and t is the time. Reaction rates for the formation of adducts of Figure 5 are collected in Table 2.

As can be observed, resorcinol had a lower reactivity than other phenolics. In addition, the highest amount of adducts were formed with orcinol. These results are likely a consequence of the electronic and steric effects of methyl groups in the phenolic compound. Thus, the introduction of a methyl group at C-2 activates the phenolic ring and, for that reason, 2-methylresorcinol was more reactive than resorcinol. When a new methyl group was introduced at position C-5, the phenolic ring should be further activated but this group also introduced a steric hindrance. For that reason, reaction yields for 2-methylresorcinol and 2,5-dimethylresorcinol were very similar. However, when the methyl group at C-2 was eliminated, as occurred in orcinol, the yield increased because the electronic effects of methyl groups are higher at *ortho* and *para* positions than at *meta* position.[22]

Independently of the assayed phenol, the formation of carbonyl-phenol adducts mostly followed the order: propanal < butanal < pentanal ≈ hexanal. This behavior seemed to be

related to an easier formation of aldol products in shorter aldehydes under the employed reaction conditions. Thus, carbonyl-phenol adducts of 2-methyl-2-pentenal (the product of aldol condensation of propanal) with phenolic compounds were detected to a high extent, although they were not quantified (data not shown).

Figure 6 shows the effect of aldehyde branching on the formation of carbonyl-phenol adducts. In addition, reaction rates for adduct formation were calculated as described above and are collected in Table 3. The presence of a methyl group at position 2 of the aldehyde always decreased the formation of the carbonyl-phenol adduct. Thus, a higher amount of butanal/2-methylresorcinol adduct than of 2-methylpropanal/2-methylresorcinol adduct was produced when incubating aldehydes having four carbon atoms (Figure 6A). Analogous results were obtained for the aldehydes having five carbon atoms (Figure 6B). Thus, pentanal produced a higher amount of adducts than 2-methylbutanal. On the other hand, similar amounts of adducts were produced by both pentanal and 3-methylbutanal. This behavior is likely a consequence of the steric hindrance introduced by the methyl group at position 2 of the aldehyde. In addition, this methyl group inhibited the formation of the 2-alkenal produced by aldol condensation and no carbonyl-phenol adducts involving 2 molecules of aldehyde were observed.

All these results confirm that flavor-relevant saturated aldehydes can be trapped by phenolics. However, this trapping ability is not the same for all kinds of aldehydes and the formation of carbonyl-phenol adducts depends on the structures of both phenol and aldehyde involved. In relation to the phenolic derivative, the nucleophilicity of phenolic carbons should be high and the presence of groups that contribute to an increase of this nucleophilicity will increase the carbonyl-trapping ability of these compounds. On the other hand, any group that increases the steric hindrance of these positions without affecting electron density, will inhibit the reaction. Analogously, the presence of

362 branching at C-2 of the aldehyde will also inhibit the reaction by steric hindrance.
363 Therefore, obtained results suggest that addition of phenolics to foods may change food
364 flavor not only because of their sensory properties[23] or because they can trap the
365 intermediates responsible for the development of off-flavors in processed foods such as
366 ultrahigh-temperature-processed bovine milk.[9] Obtained results suggest that food
367 phenolics can also trap the produced off-flavors and modify the ratio among them by
368 selective reaction with some determined carbonyls.

369

370 **ASSOCIATED CONTENT**

371 **Supporting Information**

372 The Supporting Information is available free of charge on the ACS Publications website
373 at DOI:

374 NMR and MS data of compounds **19**-**32** (PDF)

375

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Notes

The authors declare no competing financial interest.

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FIGURE CAPTIONS

Figure 1. Chemical structures of the aldehydes and phenolics employed in this study.

Figure 2. Total ion chromatogram of the reaction between pentanal and 2-methylresorcinol after acetylation. The compounds that appear in the chromatogram are: 2-methyl-1,3-phenylene diacetate (**14**), 4-(1-methoxypentyl)-2-methyl-1,3-phenylene diacetate (**15**), 2-methyl-4-(pent-1-en-1-yl)-1,3-phenylene diacetate (**16**), 4-(1-acetoxypentyl)-2-methyl-1,3-phenylene diacetate (**17**), and 4-butyl-8-methyl-3-propylchroman-2,7-diyl diacetate (**18**). Chemical structures for these compounds are collected in **Figure 3**.

Figure 3. Chemical structures of the compounds isolated and characterized in the present study.

Figure 4. Reaction pathway proposed for the reaction between saturated aldehydes and phenolics. Compounds **15**–**18** were isolated in acetylated reaction mixtures. Compounds **25** and **26** were isolated in non-acetylated reaction mixtures. All other compounds are proposed intermediates.

Figure 5. Time-course of carbonyl-phenol adduct formation in the reactions of: A, resorcinol; B, 2-methylresorcinol; C, 2,5-dimethylresorcinol; and D, orcinol; with propanal (□), butanal (○), pentanal (△), or hexanal (▽).

Figure 6. Time-course of carbonyl-phenol adduct formation in the reactions of: A, butanal (□) and 2-methylpropanal (○) with 2-methylresorcinol; and B; pentanal (□), 2-methylbutanal (○), and 3-methylbutanal (△) with 2-methylresorcinol.

Table 1. Selected Mass Spectral Data of Compounds 19–32[a]

Compound	Ion structure				
	M^+	$M^+ - CH_3OH$	$M^+ - CH_3OH - R$	$M^+ - CH_2OH$	$M^+ - R$
19	-	150 (100)			
20	-	178 (38)	149 (100)		
21	178 (48)	-	149 (100)		
22a	-	178 (100)	163 (71)		
22b	-	178 (100)	163 (80)		
23		192 (34)	149 (100)		
24		164 (100)			
25		192 (36)	163 (100)		
26	192 (35)		163 (100)		
27a[b]		192 (82)	177 (48)		
27b[b]		192 (90)	177 (45)		
28		192 (67)	177 (83)		
29	206 (39)		163 (100)		
30	168 (15)			137 (100)	
31	182 (15)			151 (100)	
32	190 (12)				161 (100)

[a]Values are *m/z* (%). R is the alkyl chain with the exception of C-1 to C-3. [b]The base ion for these compounds was $M^+ - CH_3OH - CH_3CH_2$.

Table 2. Effect of Aldehyde Chain Length on the Rate Constants of Carbonyl-Phenol Adduct Formation

Phenol	Aldehyde	Rate constant [(mmol adduct)·(mol aldehyde)$^{-1}$·h^{-1}]
Resorcinol	Propanal	0.40 ± 0.01
	Butanal	0.43 ± 0.01
	Pentanal	0.48 ± 0.01
	Hexanal	0.41 ± 0.01
2-Methylresorcinol	Propanal	1.69 ± 0.08
	Butanal	2.89 ± 0.13
	Pentanal	3.65 ± 0.17
	Hexanal	3.75 ± 0.21
2,5-Dimethylresorcinol	Propanal	1.52 ± 0.05
	Butanal	2.41 ± 0.04
	Pentanal	3.05 ± 0.06
	Hexanal	3.49 ± 0.04
Orcinol	Propanal	2.62 ± 0.04
	Butanal	4.29 ± 0.07
	Pentanal	6.06 ± 0.07
	Hexanal	5.56 ± 0.06

Table 3. Effect of Aldehyde Branching on the Rate Constants of Carbonyl-Phenol Adduct Formation

Phenol	Aldehyde	Rate constant [(mmol adduct)·(mol aldehyde)$^{-1}$·h^{-1}]
2-Methylresorcinol	Butanal	3.44 ± 0.20
	2-Methylpropanal	1.87 ± 0.03
2-Methylresorcinol	Pentanal	3.68 ± 0.11
	2-Methylbutanal	1.02 ± 0.05
	3-Methylbutanal	3.34 ± 0.11

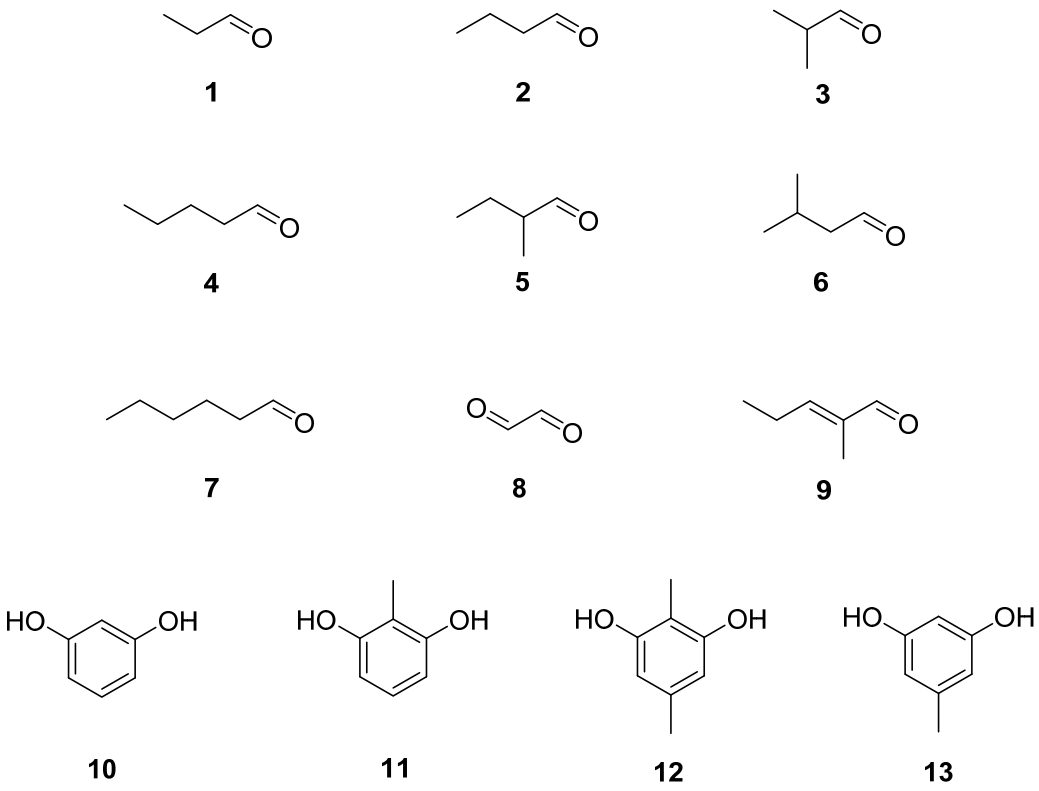

Figure 1

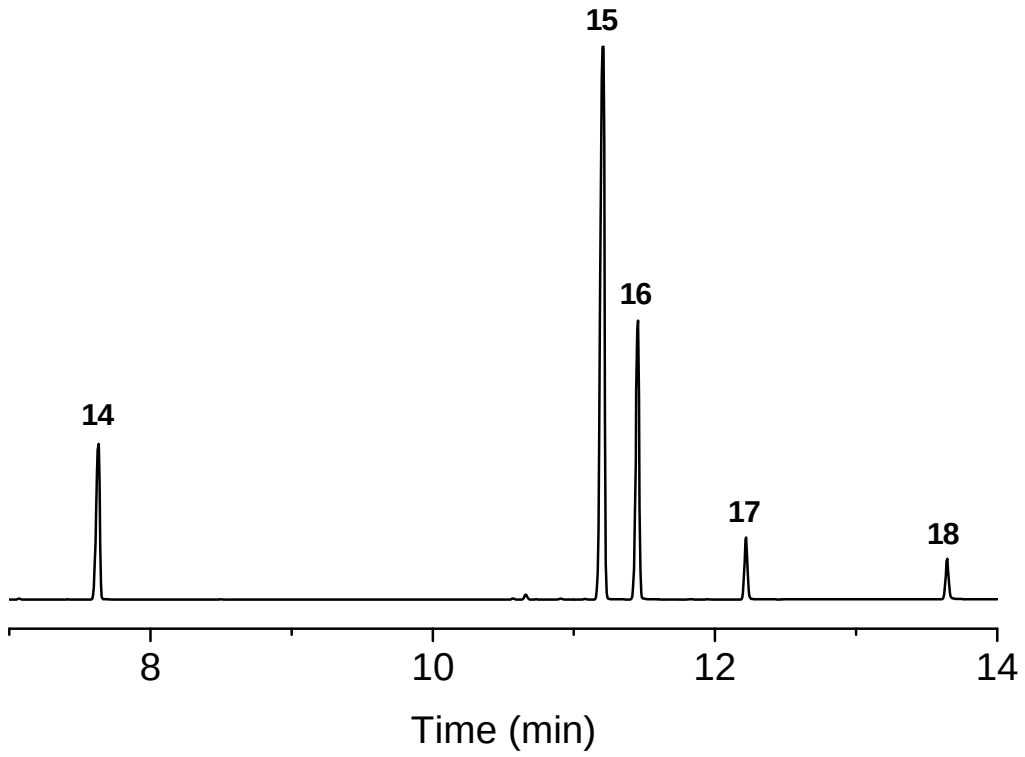

Figure 2

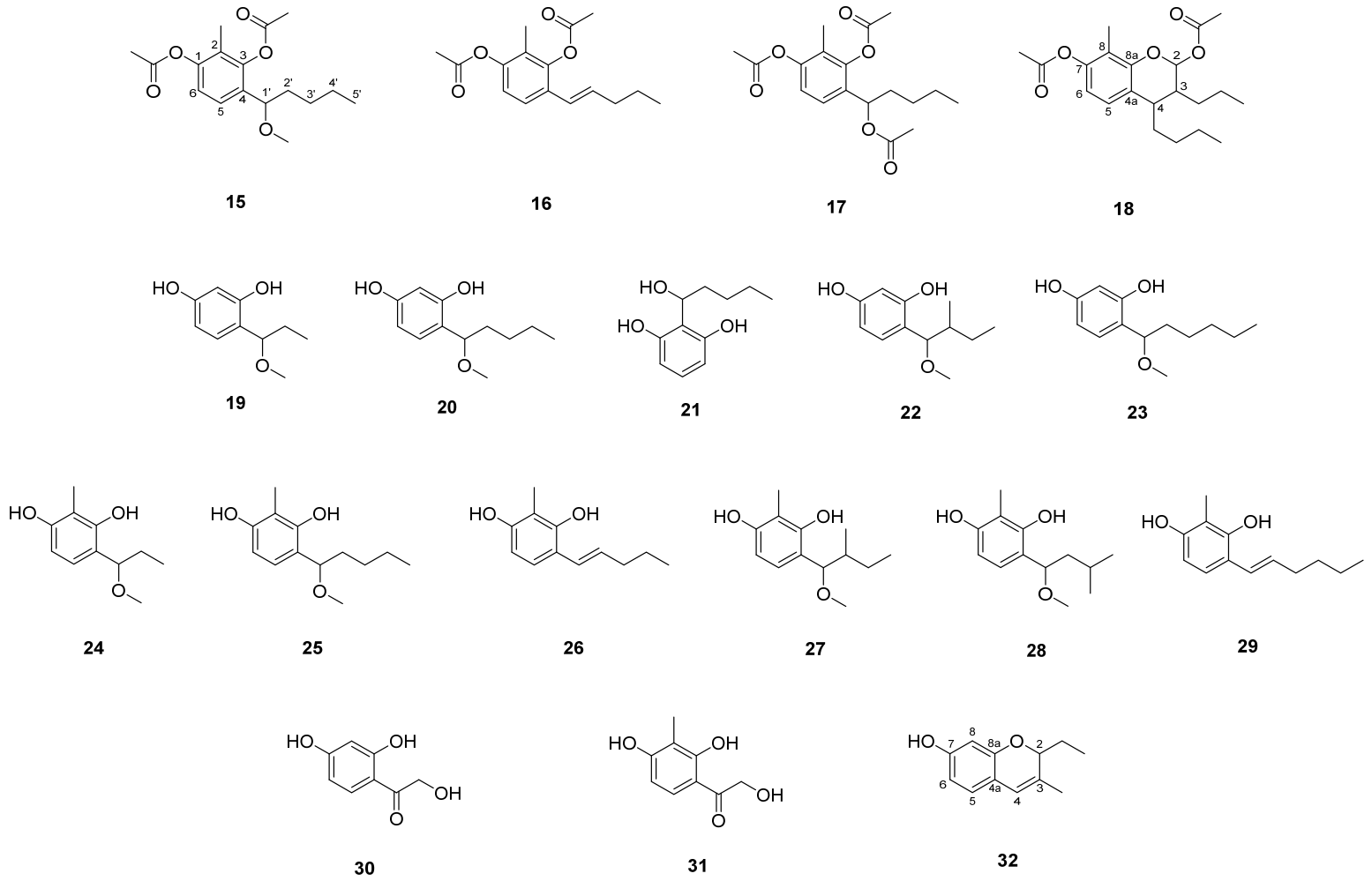

Figure 3

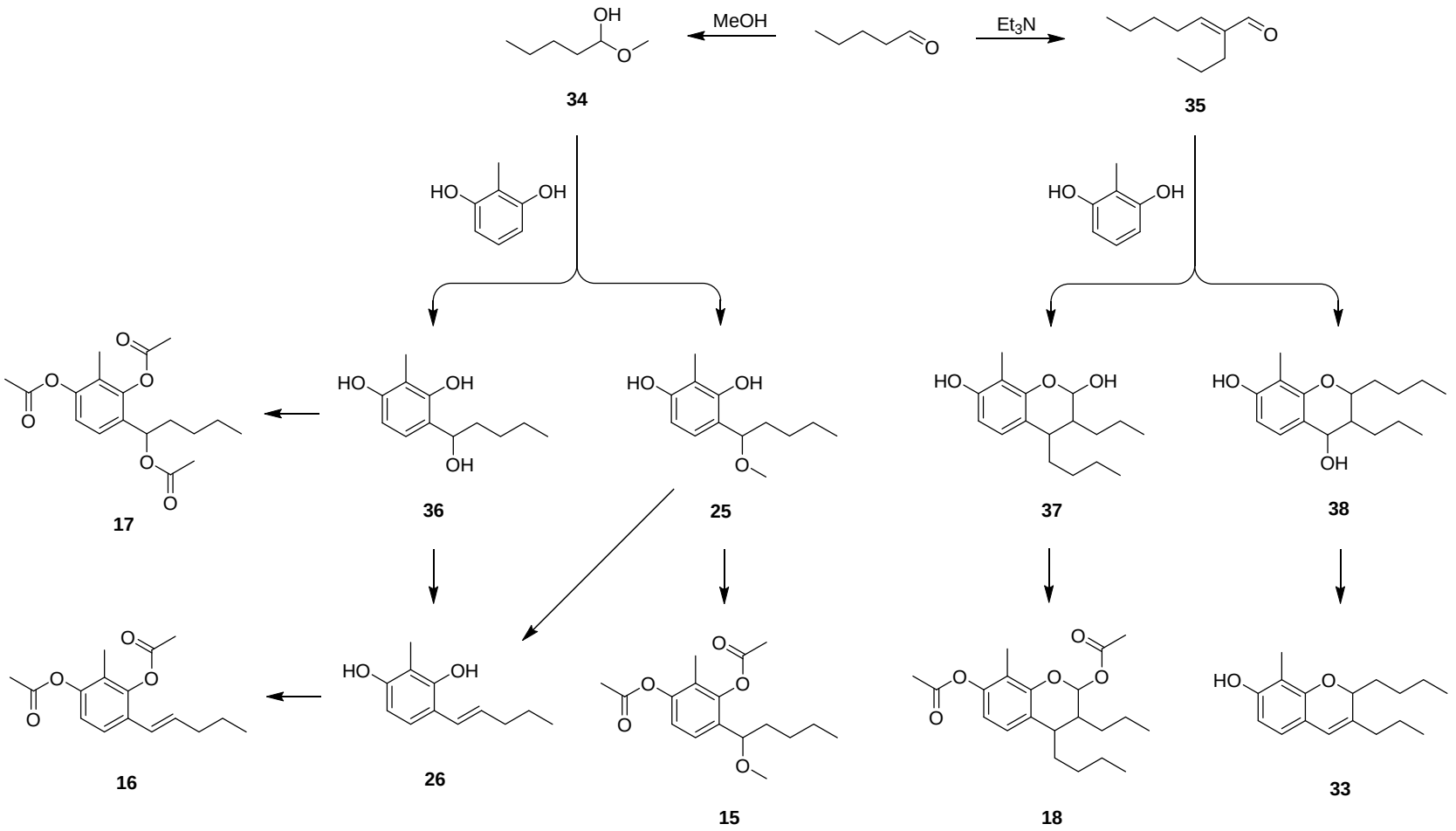

Figure 4

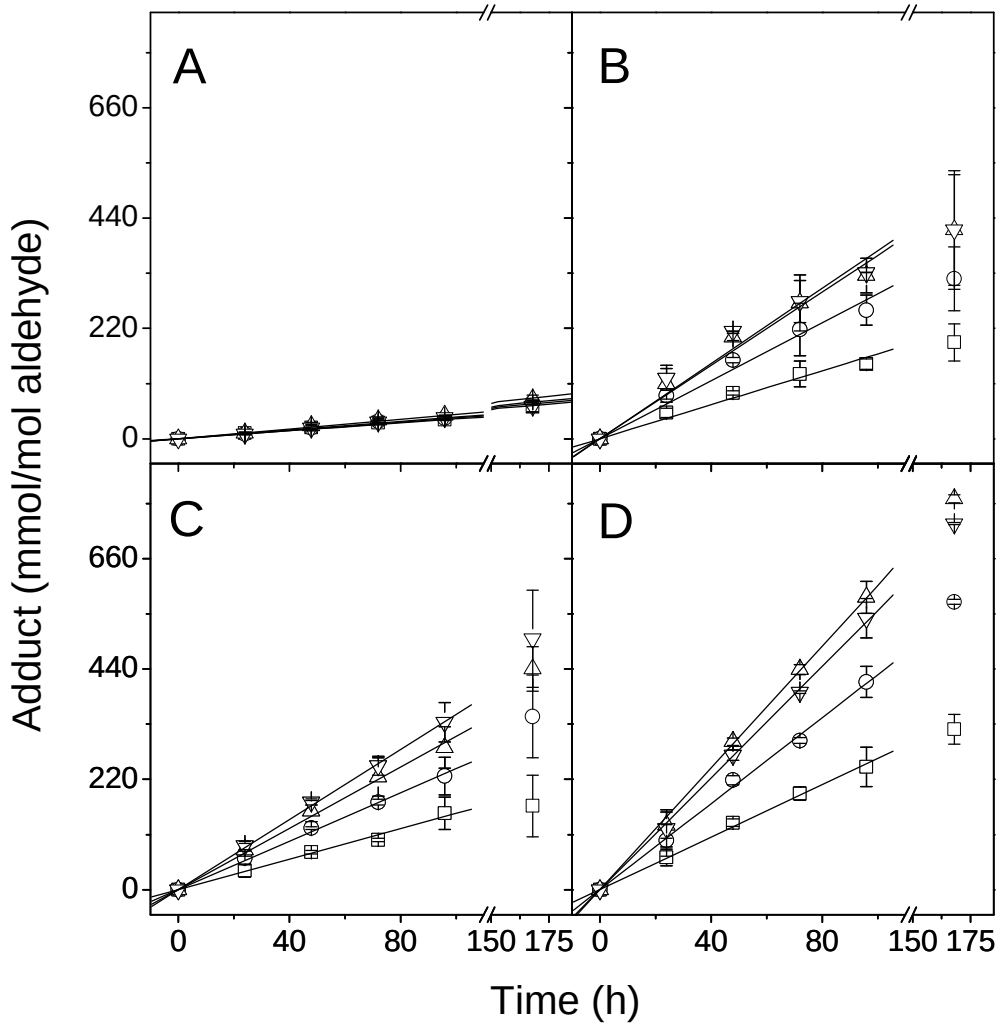
A
B
C
D
660
440
220
0
0
40
80
150
175
Adduct (mmol/mol aldehyde)
Time (h)

Figure 5

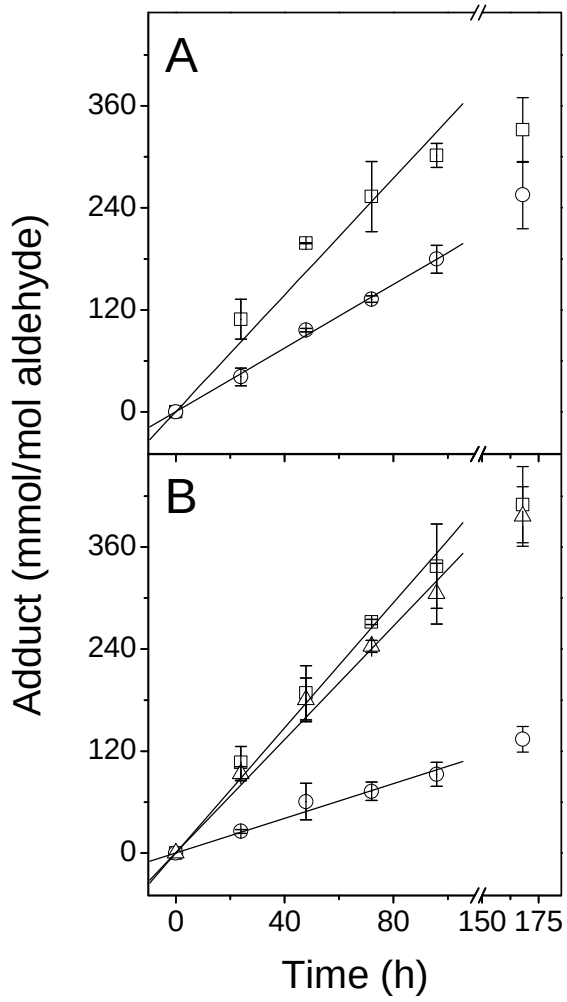

Figure 6

GRAPHIC FOR TABLE OF CONTENTS