Synthesis of hydroxyethyl starch derivatives with phenylpropanoid fragments attached through ester or sulfide bonds*

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Approaches to the synthesis of hydroxyethyl starch derivatives containing phenylpropanoic acid fragments with various degree of substitution were suggested. Esterification of hydroxyethyl starch in heterogeneous aqueous organic medium gave rise to the corresponding esters of ferulic, *p*-coumaric, and sinapinic acids. A reaction of hydroxyethyl starch mercaptodeoxy derivative with acetoxy derivatives of these hydroxycinnamic acids led to the mixed polysaccharides with the oxyphenyl-3-mercaptopropanoic acid fragments.

Key words: phenylpropanoids, hydroxyethyl starch, ferulic acid, bromodeoxypolysaccharides, mercaptodeoxypolysaccharides.

Phenylpropanoids are a large class of plant compounds composed of structural fragments of cinnamic acid and its derivatives, as well as of their esters and glycosides. These compounds like other phenylpropanoids possess high biological activity and are of interest as a starting material for the synthesis of new molecules with predicted antioxidant, antiviral, and adaptogenic properties.[1]

One of the main approaches to the design of new compounds using phenylpropanoids is their combination with carbohydrate components: mono-, oligo-, and polysaccharides. Such an approach to a certain extent is a copying organization of many macromolecular natural structures including phenylpropanoids.

There are known examples when polysaccharides are used for the synthesis of macromolecular compounds containing phenylpropanoic fragments. Earlier, chitosan, inulin, and xylan derivatives esterified with hydroxycinnamic acids were obtained.[2—4] Such compounds exhibit antioxidant properties, therefore, it seems very promising to study synthesis of compounds of polysaccharides and phenylpropanoids which include sulfur-containing groups in lower oxidation states. For the synthesis of such compounds, one can use the reaction of thiols with α,β-unsaturated acids, which proceeds similarly to the reaction of low-molecular-mass thiols and cinnamic acid.[5]

In the present work, we suggested an approach to the synthesis of polysaccharides with oxyphenylpropanoic fragments attached through ester or sulfide bonds, using starch derivative as an example.

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Results and Discussion

We have chosen acetoxyphenylpropenoic acids as compounds from the family of phenylpropanoids for the attachment to 2-hydroxyethyl starch (HES, **1**) and its mercaptodeoxy derivative.

The synthesis of hydroxyphenylpropenoic esters with HES (compounds **2**—**4**) was performed in several steps (Scheme 1). The key step of esterification of polysaccharide with acetoxycinnamoyl chlorides was carried out in the heterogeneous system water—$CHCl_3$ in the presence of NaOH with the formation of polysaccharides **2**Ac—**4**Ac. Acetoxy derivatives of ferulic, *p*-coumaric, and sinapinic acids were synthesized according to the method given in the work;[6] the corresponding acetoxyphenylpropenoyl chlorides were obtained by treatment with $SOCl_2$. In the last step, the acetate groups at the phenol hydroxy groups of polysaccharides **2**Ac—**4**Ac were removed by treatment with CH_3COONH_4 in a mixture of water—DMAc[3] to obtain polysaccharide derivatives **2**—**4** with various degrees of substitution (DS_{Ph}) containing fragments of oxyphenylpropenoic acids: ferulic (**2**), *p*-coumaric (**3**), and sinapinic (**4**).

Degree of substitution in these derivatives can be controlled by the molar excess of acetoxycinnamoyl chloride used relative to a polysaccharide monomeric unit. A twofold excess of chlorides gave the samples with $DS_{Ph} > 1.0$. The molar ratio of acetoxycinnamoyl chloride to a monomeric unit of polysaccharide **1** equal to 0.1 : 1.0 resulted in derivatives with $DS_{Ph} < 0.10$, whereas this ratio equal to 0.5 : 1.0 gave the samples with $DS_{Ph} = 0.27—0.31$ (Table 1). Derivatives of polysaccharide **1** esterified with acetoxyphenylpropenoic acids are well soluble in some polar apro-

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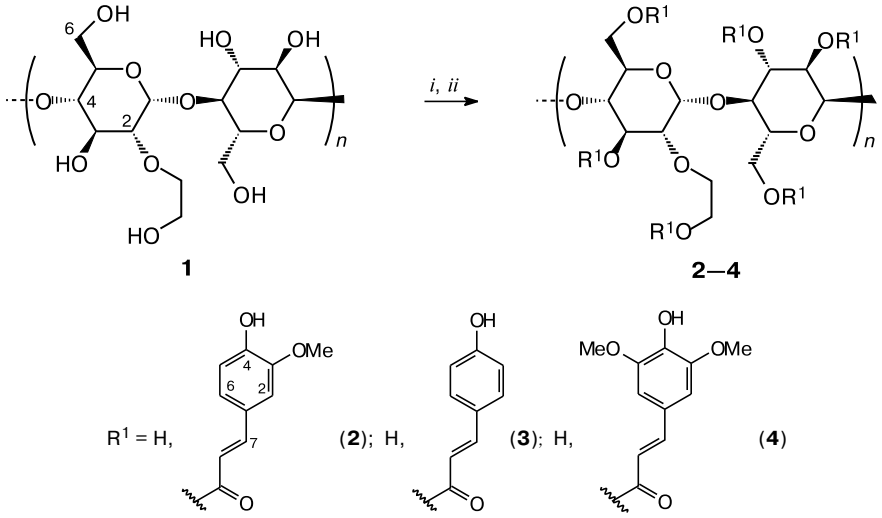
Scheme 1



Reagents and conditions: *i.* 3-(4-acetoxyphenyl)prop-2-enoic chlorides, $CHCl_3/H_2O$, NaOH, 25 °C, 6 h; *ii.* deacetylation: $MeCOONH_4$/DMAc—H_2O, 2 h, 25 °C.

tic solvents (DMSO, DMF, DMAc), whereas those with $DS_{Ph} < 0.15$ are also soluble in water. After removal of the acetyl group from the phenol hydroxyl, polysaccharides **2**—**4** only with $DS_{Ph} < 0.08$ are soluble in water.

To incorporate in the structure of polysaccharide of hydroxyphenyl-3-mercaptopropanoic acid fragments and to form sulfide bonds, we used a thiol-ene reaction, in which the synthesized from bromodeoxy-HES (**5**) HES mercaptodeoxy derivative (**6**) served as the thiol component. Hydroxy- and acetoxycinnamic acids were used as α,β-unsaturated components.

Polysaccharide **5** (Scheme 2) containing bromodeoxyglucose units was synthesized using the triphenylphosphine—NBS brominating system (Ph_3P—NBS). This system was used earlier for the synthesis of 6-bromo-6-deoxyamylose, 6-bromo-6-deoxycellulose and some other bromodeoxypolysaccharides. It acted to selectively substitute a hydroxy group at the primary carbon atom of polysaccharides with the bromine atom.[7,8] Bromodeoxypolysaccharides **5a—c** with various DS_{Br} were obtained based on compound **1** (Table 2).

Table 1. Synthesis of esters **2**—**4***: conditions and results

Compound	Acid moiety	DS_{Ph}
2	Ferulic	0.29
3	*p*-Coumaric	0.31
4	Sinapinic	0.27

* The molar ratio of acetoxyphenolic acyl chloride to polysaccharide monomeric unit was 0.5 : 1.0.

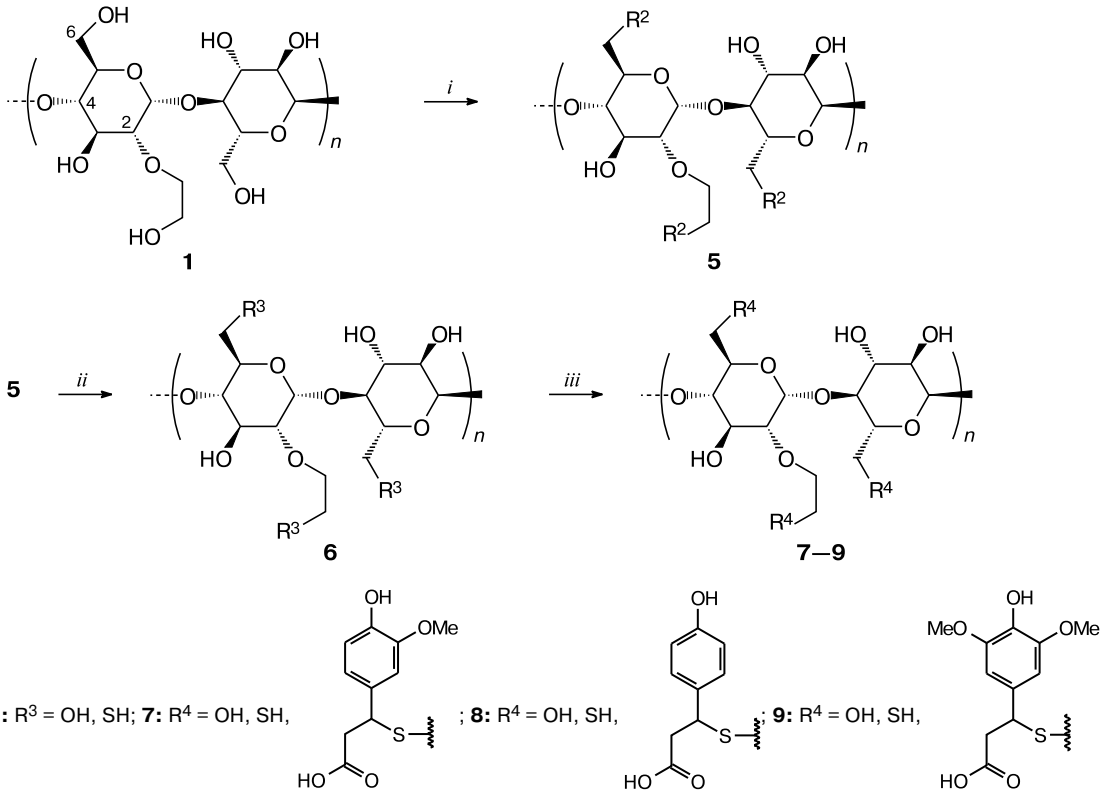
The isolated bromodeoxypolysaccharides **5a—c** are white powders, which are soluble in water when the content of bromine is <15 wt.%. When a two-fold molar excess of the brominating system was used, the sample was obtained with $DS_{Br} = 1.19$, that corresponds to the mass content of bromine ω(Br) = 34.5% (see Table 2, compound **5a**). The content of bromine in HES can be regulated by the change in the ratio of the brominating agent to the elementary unit, considering that about one-half of the brominating agent is consumed in the target reaction (see Table 2).

The synthesis of mixed mercaptodeoxypolysaccharide **6** (see Scheme 2) was carried out by the reaction of compound **5** with thiocarbamide and subsequent hydrolysis of formed thiouronium salts in alkaline medium. Polysaccharide **6** even with a low degree of substitution is poorly soluble in water, but can be dissolved in alkalis or DMF.

The thiol-ene reaction of polysaccharide **6** and acetoxyphenylcarboxylic acids was carried out in DMF at different temperatures, as well as in the presence of fluorine ions. Table 3 shows the results of the thiol-ene reaction of polysaccharide **6** with $DS_{SH} = 0.52$ with ferulic (**7a**), acetoxyferulic (**7b—e**), acetoxycoumaric (**8**), and acetoxysinapinic (**9**) acids.

Hydroxyphenylpropenoic acids used as α,β-unsaturated components do not give the thiol-ene reaction with the thiol groups of polysaccharide **6** (see Table 3, compound **7a**). Addition of acetoxyphenylpropenoic acids to polysaccharide **6** with the formation of sulfide bond requires prolonged heating of the reaction mixture. It was found that an increase in temperature to 100 °C provides the highest conversion of the thiol groups to the sulfide ones without significant decrease in the yield of the modi-

Scheme 2



Reagents and conditions: *i*. DMAc, Ph_3P—NBS, 80 °C, 4 h; *ii*. DMF, $(NH_2)_2CS$, 80 °C, 24 h; NaOH. *iii*. 1) DMF, 4-acetoxyferulic, 4-acetoxycoumaric, or 4-acetoxysinapinic acid, 100 °C, 24 h; 2) 0.5 *N* NaOH.

fied polysaccharide. Under the selected conditions, the greater part of phenylpropanoic fragments is incorporated in the structure of polymer within 24 h (see Table 3, compounds **7c** and **7d**) with obtaining polysaccharides containing the hydroxyphenyl-3-mercaptopropanoic acid fragments. However, the DS_{SPh} values obtained indicate that under these conditions only ≤70% of thiol groups of compound **6** are involved in the thiol-ene reaction. Degree of substitution calculated according to the data of acid-base titration of the carboxy groups in the addition product is lower as compared to the data of elemental analysis, that is apparently explained by poor availability of some acid groups for the ion exchange.

Earlier, NBu_4F was suggested for the use as a catalyst of thiol-ene reactions involving phenylpropanoids. However, in our studies addition of NBu_4F as a catalyst (**7e**)

Table 2. Bromination of compound **1** with a mixture of Ph_3P—NBS: conditions and results

Product	[Br] : [PS]* (mol)	DS_{Br}	Yield (%) (on the mass of starting polymer)
5a	2.0	1.19	52
5b	1.0	0.57	90
5c	0.5	0.28	93

* The molar ratio of brominating agent (Br) to polysaccharide (PS) unit.

Table 3. Thiol-ene reaction of polysaccharide **6** and α,β-unsaturated acids: conditions and results[a]

Product	Starting acid	τ/h	DS_{SPh}[c]	
			I	II
7a	Ferulic	24	0.00	0.00
7b	4-Acetoxyferulic	10	0.19	0.12
7c	4-Acetoxyferulic	24	0.34	0.26
7d	4-Acetoxyferulic	48	0.38	0.27
7e[b]	4-Acetoxyferulic	24	0.11	0.08
8	4-Acetoxycoumaric	24	0.37	0.30
9	4-Acetoxysinapinic	24	0.32	0.26

[a] The results for the synthesis at 100 °C and a 1.5 molar excess of phenolic acid relative to thiol group.
[b] 20 mol.% of NBu_4F was added.
[c] Degree of substitution (DS_{SPh}) from the results of elemental analysis (I) and titration (II).

led to the decrease in the content of phenylpropanoic structures in the polymer after the reaction of polysaccharide **6** and acetoxyferulic acids as compared to the results of this reaction without source of fluoride ions. Analysis of the products of the reaction between compounds **6** and acetoxyferulic acid with NBu_4F showed the presence of ferulic acid among the low-molecular-mass products, that allowed us to draw a conclusion on the deacetylation of acetoxyphenylpropenoic acid under these conditions. The hydroxyphenylpropenoic acid formed does not react with thiol, as shown for compound **7a**, that leads to the formation of derivatives with low DS_{SPh}.

In conclusion, we have synthesized hydroxyethyl starch derivatives containing phenylpropanoic fragments, which are grafted to the polysaccharide chain with ester and sulfide bonds. For the preparation of the target modified polysaccharides, we used esterification in aqueous organic medium and thiol-ene reaction involving mercaptodeoxypolysaccharide and acetoxyphenylpropenoic acids. Bromodeoxypolysaccharide synthesized as a precursor for obtaining mercaptodeoxypolysaccharide has a significant potential for further modification, including that which suggests addition to this polysaccharide of various phenol-containing compounds.

The macromolecular compounds obtained in these studies and containing phenylpropanoic structures, both water-soluble and insoluble in water, are of interest primarily as macromolecular antioxidants with the structure close to the natural representatives. Sulfur-containing polysaccharide-phenols are promising as components capable of modifying synthetic polymeric matrix in the technologies of preparation of thermoplastic polymeric materials (plastic, rubber). The studies on the selection of conditions and catalysts for the reaction of α,β-unsaturated phenolic acids with macromolecular thiols should be continued, since this method is promising for obtaining new compounds and materials on their basis with wide possibilities of application.

Experimental

The starting HES (DS = 0.5, MW = $200 \cdot 10^3$) (Serumwerk Bernburg AG), ferulic (3-(4-hydroxy-3-methoxyphenyl)prop-2-enoic), *p*-coumaric (3-(4-hydroxy)prop-2-enoic), and sinapinic (3-(4-hydroxy-3,5-dimethoxyphenyl)prop-2-enoic) acids, as well as $NBu_4F \cdot 3H_2O$, NBS, Ph_3P, $SOCl_2$ (Sigma-Aldrich) were used without preliminary purification. Ferulic and *p*-coumaric acid acetates were obtained according to the method described earlier.[6] The solvents DMAc and DMF were purified and dried before used according to the known procedure.[9]

^1H and ^{13}C NMR spectra were recorded on a Bruker Avance II 300 spectrometer (300.17 (^1H) and 75.5 MHz (^{13}C), solvent DMSO-d_6). IR spectra were recorded on a Shimadzu IR Prestige 21 Fourier-transform IR spectrometer. Elemental analysis was carried out on a Vario Micro Cube Elementar CHNS analyzer. Bromine in derivatives was determined according to the method described earlier.[10] Content of phenolic acids in polysaccharide (ω, wt.%) was determined spectrophotometrically according to the method described earlier,[11] using a UV-1700 spectrophotometer (Shimadzu). For the HES esters containing 4-acetoxyphenylcarboxylic acid fragments, as well as for esters insoluble in water, this analysis was carried out after preliminary hydrolysis of the ester bond and extraction of phenolic acids with AcOEt (see Ref. 2). Degree of substitution (DS_{Ph} is the number of added phenylpropanoids per one polysaccharide unit) in the esterified polysaccharide derivatives were calculated using the formula:

$$DS_{Ph} = \frac{M_{eu}\omega_{pa}}{M_{pa} \cdot 100 - (M_{pa} - 1)\omega_{pa}},$$

where M_{eu} is the average molecular mass of the polysaccharide elementary unit (182 g mol^{-1}); ω_{pa} is the content of the corresponding phenolic acid, wt.%; M_{pa} is the molecular mass of the phenolic acid fragment (which was 193, 163, and 223 g mol^{-1} for ferulic, coumaric, and sinapinic acid, respectively).

Degree of substitution in the products of the thiol-ene reaction (DS_{SPh}) was determined using the elemental analysis data and the results of the acid-base titration. To carry out the titration, a weighed amount of the sample (0.08—0.12 g) was left to swell in DMSO (2 mL) for 3 h, then 0.1 *N* aqueous NaCl was added. The concentration of the samples in solutions before titration was 2.0 mg mL^{-1}. The titration was carried out with 0.05 *N* aqueous NaOH. The medium pH in all the experiments was measured using an Ekspert-pH ionometer (Ekonix, Russia) with an ESK-10601/7 combination electrode. Degree of substitution was calculated using the formula:

$$DS_{Ph} = \frac{M_{eu}n(COOH)}{m - (M_{pa} - 1)n(COOH)},$$

where M_{eu} = 190 g mol^{-1} is the average molecular mass of the polysaccharide **6** elementary unit; n(COOH) is the content of carboxy groups obtained from titration, mol; M_{pa} is the molecular mass of the oxyphenylpropanoic acid fragment (M_{pa} = 195, 165, and 225 g mol^{-1} for the products of the reaction of **6** with derivatives of ferulic, coumaric, and sinapinic acid, respectively).

3-(4-Acetoxy-3,5-dimethoxyphenyl)prop-2-enoic acid (4-acetoxysinapinic acid). Sinapinic acid (2.0 g, 8.9 mmol) was dissolved in dioxane (10 mL), followed by addition of aqueous solution (50 mL) containing NaOH (0.92 g, 23 mmol). The solution was cooled to 4 °C and the first portion of Ac_2O (4.5 mmol) was added. The temperature of the reaction mixture was raised to 25 °C over next 30 min and the rest of Ac_2O (7.7 mmol) was added. After standing at 25 °C for 1 h, the reaction mixture was acidified with 2 *N* aqueous H_2SO_4 to pH 4.5, a precipitate formed was filtered off, washed with H_2O until neutrality, and recrystallized from aqueous (80%) EtOH. The yield of the product was 78%. IR (KBr), ν/cm^{-1}: 2944 (OH), 1765 (AcOPh), 1664 (COOH). ^1H NMR (DMSO-d_6), δ: 2.268 (s, 3 H); 3.816 (s, 6 H); 6.61—6.67 (d, 1 H, J = 15.9 Hz); 7.12 (s, 2 H); 7.56—7.61 (d, 1 H, J = 15.9 Hz); 12.39 (s, 1 H). ^{13}C NMR (DMSO-d_6), д: 20.58 (Me); 56.44 (OMe); 105.62 (C(2), C(6)); 120.18 (C(8)); 129.94 (C(4)); 133.11 (C(1)); 144.23 (C(7)); 152.44 (C(3), C(5)); 168.06 (COOH);168.14 (Me<u>C</u>(O)OPh).

Synthesis of acyl chlorides was accomplished by reflux of the corresponding acetoxycinnamic acid with a three-fold excess of

$SOCl_2$ in $CHCl_3$ in the presence of a catalytic amount of DMF for 4 h with subsequent evaporation of the solvent *in vacuo*. The chlorides were used in syntheses immediately after preparation without additional purification.

3-(4-Acetoxy-3-methoxyphenyl)prop-2-enoyl chloride. The yield was 94%. ^{1}H NMR ($CDCl_3$), δ: 2.35 (s, 3 H); 3.88 (s, 3 H); 6.57 (d, 1 H, J = 15.9 Hz); 7.09—7.12 (m, 3 H); 7.79 (d, 1 H, J = 15.9 Hz); 12.39 (s, 1 H). ^{13}C NMR ($CDCl_3$), δ: 20.6 (Me); 56.4 (OMe); 111.9 (C(6)); 120.2 (C(8)); 123.6 (C(4)); 131.2 (C(1)); 142.8 (C(7)); 149.8 (PhC=C); 165.9 (COCl); 168.5 (MeC(O)OPh).

3-(4-Acetoxyphenyl)prop-2-enoyl chloride. The yield was 96%. ^{1}H NMR ($CDCl_3$), δ: 2.28 (s, 3 H); 6.78 (d, 1 H, J = 15.9 Hz); 7.22—7.24 (m, 2 H); 7.85—7.89 (d, 2 H). ^{13}C NMR ($CDCl_3$), δ: 21.0 (Me); 122.4 (C(2), C(6)); 129.6 (C(3), C(5)); 142.8 (C(7)); 148.8 (PhC=C); 166.6 (COCl); 169.0 (MeC(O)OPh).

3-(4-Acetoxy-3,5-dimethoxyphenyl)prop-2-enoyl chloride. The yield was 96%. ^{1}H NMR (DMSO-d_6), δ: 2.26 (s, 3 H); 3.87 (s, 6 H); 6.59—6.63 (d, 1 H, J = 15.9 Hz); 7.13 (s, 2 H); 7.74—7.78 (d, 1 H, J = 15.9 Hz). ^{13}C NMR (DMSO-d_6), δ: 20.2 (Me); 56.4 (OMe); 105.4 (C(2), C(6)); 120.2 (C(8)); 129.31 (C(4)); 134.5 (C(1)); 144.9 (C(7)); 153.0 (C(3), C(5)); 166.64 (COCl); 168.14 (MeC(O)OPh).

Synthesis of 2Ac—4Ac (general method). Compound **1** (500 mg, 2.74 mmol) was dissolved in water (12 mL) with stirring under nitrogen, then NaOH (an equimolar amount to the acyl chloride) was added. A solution of phenylcarboxyl chloride in $CHCl_3$ (10 mL) was added to the reaction mixture cooled to 4 °C with vigorous stirring. Then, the temperature of the reaction mixture was raised to 25 °C for 1 h, and the mixture was stirred for another 5 h. To isolate the ester, the heterogeneous mixture was acidified with 0.5 *N* HCl to pH 4.5 and poured into Pr^{i}OH (200 mL). A precipitate formed was separated by centrifugation, washed with 70% aqueous EtOH and then subjected to freeze drying.

***O*-[3-(4-Acetoxy-3-methoxyphenyl)prop-2-enoyl]-*O*-(2-hydroxyethyl)-(1→4)-α-D-glucan** (ester of 4-acetoxyferulic acid and HES, **2**Ac). Polysaccharide (380 mg) was obtained as a slightly colored finely grained powder. IR (KBr), ν/cm^{-1}: 3396 (OH); 2934 (CH_2); 1761 (C=O, AcOPh); 1724 (C=O); 1634, 1510 (C—H, Ar); 1033 (C—O, carbohydrate skeleton). ^{13}C NMR (DMSO-d_6), δ: 20.8 (Me); 56.5 (OMe); 60.8 (C(6), CH_2CH_2OH, HES); 72.6 (C(2), C(3), CH_2CH_2OH, HES); 73.6 (C(5), HES); 80.3 (C(4), HES); 96.8 (C(1), HES, hydroxyethylated unit); 100.5 (C(1), HES); 112.2 (C(2)); 116.0 (C(8)), 121.6 (C(6)); 123.8 (C(5)); 133.3 (C(1)); 141.5 (C(4)); 146.6 (C(7)); 151.7 (C(3)); 168.7 (C=O); 168.9 (MeC(O)OPh).

***O*-[3-(4-Acetoxyphenyl)prop-2-enoyl]-*O*-(2-hydroxyethyl)-(1→4)-α-D-glucan** (ester of HES and 4-acetoxy-*p*-coumaric acid, **3**Ac). Polysaccharide (366 mg) was obtained as a slightly colored finely grained powder. IR (KBr), ν/cm^{-1}: 3477 (OH); 2943 (CH); 1762 (C=O, AcOPh); 1719 (C=O); 1631, 1506 (C—H, Ar). ^{13}C NMR (DMSO-d_6), δ: 60.8 (C(6), CH_2CH_2OH, HES); 72.20—72.73 (C(2), C(3), CH_2CH_2OH, HES); 73.5 (C(5), HES); 80.3 (C(4), HES); 96.8 (C(1), HES, hydroxyethylated unit); 100.7 (C(1), HES), 116.22 (C(8)); 122.9 (C(3), C(5)); 129.85—130.80 (C(2), C(6)); 152.2 (C(4)), 160.0 (MeC(O)OPh).

***O*-[3-(4-Acetoxy-3,5-dimethoxyphenyl)prop-2-enoyl]-*O*-(2-hydroxyethyl)-(1→4)-α-D-glucan** (ester of HES and 4-acetoxy-sinapinic acid, **4**Ac). Polysaccharide (370 mg) was obtained as a slightly colored finely grained powder. IR (KBr), ν/cm^{-1}: 3415 (OH); 2931 (CH); 1780 (C=O, AcOPh); 1701 (C=O); 1627, 1597, 1506 (C—H, Ar). ^{13}C NMR (DMSO-d_6), δ: 20.6 (Me); 56.7 (OMe); 60.9 (C(6), CH_2CH_2OH, HES); 72.18—72.62 (C(2), C(3), CH_2CH_2OH, HES); 73.5 (C(5), HES); 79.2 (C(4), HES); 79.7 (C(4), HES); 96.8 (C(1), HES, hydroxyethylated unit); 100.7 (C(1), HES); 105.6 (C(2), C(6)); 120.2 (C(8)); 129.9 (C(4)); 133.1 (C(1)); 144.2 (C(7)); 152.4 (C(3), C(5)); 168.0 (C=O); 168.4 (MeC(O)OPh).

Removal of the acetyl protecting group from the phenol hydroxyl in the esters of HES and acetoxycinnamic acids (general method). Esters **2**Ac—**4**Ac (200 mg) were dissolved in DMAc (3.0 mL), followed by addition of an aqueous solution of $MeCOONH_4$ (3.0 mL; 8 mol. equiv. of $MeCOONH_4$ per 1 mol. equiv. of the acetyl group). The mixture was stirred for 2 h at 25 °C. Polysaccharide was precipitated by addition of Pr^{i}OH (25 mL). The precipitate was separated, washed with EtOH acidified with AcOH, then with EtOH, and subjected to freeze drying.

***O*-[3-(4-Hydroxy-3-methoxyphenyl)prop-2-enoyl]-*O*-(2-hydroxyethyl)-(1→4)-α-D-glucan** (ester of HES and ferulic acid, **2**). Polysaccharide (170 mg) was obtained as a slightly colored finely grained powder (DS_{Ph} = 0.29). IR (KBr), ν/cm^{-1}: 3408 (OH); 2934 (CH); 1719 (C=O); 1631, 1514 (C—H, Ar). ^{13}C NMR (DMSO-d_6), δ: 56.5 (OMe); 60.7 (C(6), CH_2CH_2OH, HES); 71.59—72.61 (C(2), C(3), CH_2CH_2OH, HES); 73.6 (C(5), HES); 79.4 (C(4), HES); 96.7 (C(1), HES, hydroxyethylated unit); 100.7 (C(1), HES); 115.9 (C(8)); 126.1 (C(1)); 148.33 (C(3)); 149.6 (C(4)); 170.7 (C=O).

***O*-[3-(4-Hydroxyphenyl)prop-2-enoyl]-*O*-(2-hydroxyethyl)-(1→4)-α-D-glucan** (ester of HES and *p*-coumaric acid, **3**). Polysaccharide (180 mg) was obtained as a slightly colored finely grained powder (DS_{Ph} = 0.31). IR (KBr), ν/cm^{-1}: 3406 (OH); 2935 (CH_2); 1709 (C=O); 1627, 1606, 1514 (C—H, Ar). ^{13}C NMR (DMSO-d_6), δ: 61.0 (C(6), CH_2CH_2OH, HES); 70.37—72.76 (C(2), C(3), CH_2CH_2OH, HES); 73.7 (C(5), HES); 80.6 (C(4), HES); 97.3 (C(1), HES, hydroxyethylated unit); 100.8 (C(1), HES); 116.4 (C(8)); 123.0 (C(3), C(5)); 130.8 (C(2), C(6)); 160.3 (C(4)).

***O*-[3-(4-Hydroxy-3,5-dimethoxyphenyl)prop-2-enoyl]-*O*-(2-hydroxyethyl)-(1→4)-α-D-glucan** (ester of HES and sinapinic acid, **4**). Polysaccharide (167 mg) was obtained as a slightly colored finely grained powder (DS_{Ph} = 0.27). IR (KBr), ν/cm^{-1}: 3396 (OH); 2933 (CH); 1724 (C=O); 1634, 1587, 1510 (C—H, Ar). ^{13}C NMR (DMSO-d_6), δ: 56.7 (OMe); 61.0 (C(6), CH_2CH_2OH, HES); 70.23—72.71 (C(2), C(3), CH_2CH_2OH, HES); 73.7 (C(5), HES); 81.1 (C(4), HES); 96.8 (C(1), HES, hydroxyethylated unit); 100.7 (C(1), HES); 106.8 (C(2), C(6)); 116.4 (C(8)); 139.8 (C(4)); 149.2 (C(3); C(5)); 167.8 (C=O).

6-Bromo-6-deoxy-[2-*O*-(2-bromoethyl)]-(1→4)-α-D-glucan (bromodeoxy HES, **5**). Compound **1** (5.00 g, 27.5 mmol) was dissolved with heat in DMAc (100 mL) under argon. A solution of the brominating system was prepared separately by mixing Ph_3P (7.21 g, 27.5 mmol) and NBS (4.90 g, 27.5 mmol) in DMAc (100 mL) with cooling. The solution of the brominating system was added to the solution of compound **1** and the homogeneous mixture was heated for 3 h at 80 °C. Then, it was cooled down, polysaccharide was precipitated with Pr^{i}OH (1.0 L), the precipitate was separated, sequentially washed with acetone and 70% aqueous EtOH and dried *in vacuo* at 60 °C to obtain polysaccharide **5** (4.60 g) with ω(Br) = 20.10 wt.%, DS_{Br} = 0.57. IR (KBr), ν/cm^{-1}: 3423 (OH); 2931 (CH, CH_2); 1452 (C—O—C); 1060 (C—O—C); 538 (C—Br). ^{13}C NMR (75 MHz, DMSO-d_6), δ: 30.0 (CH_2CH_2Br); 35.0 (C(6), Br); 61.0 (C(6), CH_2CH_2OH,

HES); 70.20—72.77 (C(2), C(3), $\underline{C}H_2CH_2OH$, HES); 73.6 (C(5), HES); 80.5 (C(4), HES); 96.9 (C(1), HES, hydroxyethylated unit); 100.9 (C(1), HES).

6-Deoxy-6-mercapto-6-(2-*O*-(2-mercaptoethyl)-(1→4)-α-D-glucan (mercaptodeoxy HES, **6**). Polysaccharide **5** (3.00 g, 13.8 mmol) was dissolved in DMF (25 mL). Thiourea (5.25 g, 69.0 mmol) in DMF (25 mL) was added to the solution. The mixture obtained was thermostated over 24 h at 80 °C under argon atmosphere. After cooling the mixture, polysaccharide was precipitated with EtOH (1.0 L), washed with EtOH, and poured into 2 *N* solution of NaOH in 70% aqueous EtOH. The mixture was stirred for 3 h at 20 °C. Then, the solution was acidified with 2 *N* HCl, a precipitate was separated, washed with 70% aqueous EtOH, and subjected to freeze drying to obtain polysaccharide **6** (2.20 g) with ω(S) = 8.46 wt.%, DS_S = 0.52. Found (%): C, 44.70; H, 5.43; S, 8.46. Calculated for DS_{SH} = = 0.52: C, 44.79; H, 5.60; S, 8.46. IR (KBr), ν/cm^{-1}: 3442 (OH); 2927 (CH_2); 2563 (SH).

Synthesis of compounds 7—9. Polysaccharide **6** (0.50 g, 2.6 mmol) was placed in DMF (3 mL). A solution of acetoxyphenylpropenoic acid in DMF (3 mL; 1.5 molar excess relative to the thiol group) was added to the solution under argon. The reaction mixture was stirred for 24 h at 100 °C under argon. Homogenization of the reaction mixture was observed within first 60 min the reaction. Then, the solvent was evaporated *in vacuo* (65 °C), the residue was washed with Pr^iOH and poured into 0.5 *N* aqueous NaOH (5 mL). The solution was allowed to stand for 3 h, then the mixture was neutralized with 1.0 *N* HCl to pH 2—3. A precipitate was sequentially washed with H_2O and 70% aqueous EtOH and dried *in vacuo* at 60 °C.

***O*-{[1-(4-Hydroxy-3-methoxyphenyl]-2-carboxyethylthio]-ethyl}-(1→4)-α-D-glucan (7).** The product (0.36 g) was obtained as a yellowish powder, DS_{SPh} = 0.34. Found (%): C, 48.11; H, 6.23; S, 6.36. Calculated (%): C, 48.16; H, 6.26; S, 6.39. IR (KBr), ν/cm^{-1}: 3391 (OH); 2929 (CH_2); 1701 (COOH); 1629, 1597, 1514 (C—H, Ar).

***O*-{[1-(4-Hydroxyphenyl)-2-carboxyethylthio]ethyl}-(1→4)-α-D-glucan (8).** The product (0.39 g) was obtained as a yellowish powder, DS_{SPh} = 0.37. Found (%): C, 49.24; H, 5.48; S, 6.54. Calculated (%): C, 49.20; H, 5.52; S, 6.60. IR (KBr), ν/cm^{-1}: 3441 (OH); 2927 (CH); 1712 (COOH); 1565, 1514 (C—H, Ar).

***O*-{[1-(4-Hydroxy-3,5-dimethoxyphenyl)-2-carboxyethylthio]ethyl}-(1→4)-α-D-glucan (9).** The product (0.34 g) was obtained as a yellowish powder, DS_{SPh} = 0.32. Found (%): C, 48.11; H, 5.37; S, 6.34. Calculated (%): C, 48.13; H, 5.39; S, 6.34. IR (KBr), ν/cm^{-1}: 3441 (OH); 2927 (CH); 1712 (COOH); 1565, 1514 (C—H, Ar).

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