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Processing of Rice Husk Autohydrolysis Liquors for Obtaining Food Ingredients

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Rice husks are of a lignocellulosic nature, with a hemicellulose fraction made up of substituted arabinoxylan. Rice husks were treated with hot, compressed water (autohydrolysis reaction) under optimized conditions to cause the hydrolytic degradation of arabinoxylan. The reaction products contained volatile components and nonvolatile components (NVC), which were made up of hemicellulose-derived products (substituted oligosaccharides and monosaccharides) and other nonvolatile solutes (ONVS). To decrease the content of ONVS, concentrated autohydrolysis liquors were first subjected to ethyl acetate extraction and then subjected to various alternative treatments (solvent precipitation, freeze-drying solvent extraction, or ion exchange). The resulting liquors were assayed for composition and yield determination. Material balances are presented for the several processes considered. The best results in terms of purification were obtained with sequential stages of ethyl acetate extraction and ion exchange, which led to concentrates with hemicellulose-derived compounds (sugars and substituted sugar oligomers) accounting for 92 wt % of the NVCs.

KEYWORDS: Autohydrolysis; rice husks; hemicelluloses; purification; refining; xylooligosaccharides

INTRODUCTION

Lignocellulosic materials are mainly made up of cellulose, hemicelluloses, lignin, extractives, proteins, and ashes. In a worldwide context defined by the shortening of nonrenewable resources, lignocellulosic materials can replace dwindling traditional raw materials of the chemical industry for many product applications.

Hemicelluloses are the second most abundant polysaccharide in nature, with an estimated annual world production of 60 billion tons (*1*). They are alternative sources of chemicals, including food-related products.

From a chemical point of view, hemicelluloses are heteropolymers made up of pentoses (xylose, arabinose), hexoses (mannose, glucose, and galactose), and uronic acid units. The composition of hemicelluloses depends on the lignocellulosic material considered; hardwoods contain mostly xylan, softwoods contain mostly glucomannans, and many agricultural wastes or byproducts are made up of backbone chains of 1−4-linked β-D-xylopyranose units substituted with arabinose, uronic acid moieties (or its 4-*O*-methyl ether), and acetic, ferulic, or coumaric acids (*2*).

Starting from xylan-containing lignocellulosic materials, the breakdown of hemicellulose chains leads to xylooligomers with food, medical, and pharmaceutical applications (*3*). As food ingredients, xylooligosaccharides have prebiotic actions (*4*, *5*), improving the intestinal function by enhancing the growth of healthy *Bifidobacteria* (*6*), suppressing the growth of *Clostridium*, and causing bacteriostatic effects against *Vibrio anguillarum* (*7*).

The breakdown of xylan into xylooligosaccharides can be carried out by a variety of methods, including direct enzymatic conversion of susceptible raw materials, xylan isolation (for example, by alkaline extraction and ethanol precipitation) followed by enzymatic hydrolysis, or autohydrolysis reaction. In the latter case, a suitable raw material is heated in an aqueous media, and the catalytic action of hydronium ions coming from water ionization and from in situ generated acids (such as acetic acid coming from acetyl groups) leads to the cleavage of the xylan heterocyclic ether bonds, resulting in the formation of xylooligosaccharides. Depending on the severity of the operational conditions, autohydrolysis can result in the formation of sugars and sugar degradation products (*8*−*11*).

However, the autohydrolysis reaction is not specific, and a variety of side processes take place simmultaneously to the hydrolytic xylan degradation, including extractive removal, solubilization of acid soluble lignin, ash neutralization, and reactions involving proteins. Because of this, undesired, nonsaccharide compounds appear in the reaction media, and further processing of autohydrolysis liquors is needed to improve the purity of xylooligosaccharide concentrates.

A variety of strategies have been reported in the literature for purification of hemicellulose-derived products. Solvent extraction has been employed to separate nonsaccharide fractions

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with antioxidant and antimicrobial properties from media obtained by hydrolytic processing of biomass (*12*, *13*). Adsorption (*14*) and chromatographic separation (*15*−*17*) have been used for purification of xylooligosaccharide-containing liquors, whereas ion exchange treatments are useful for desalination and removal of other undesired compounds (*18*−*24*).

This work deals with the experimental evaluation of selected experimental strategies (solvent extraction followed by other treatments, including solvent precipitation, freeze-drying solvent extraction, or ion exchange) for increasing the content of saccharide-derived products starting from liquors obtained by aqueous treatments of rice husks performed under optimized conditions.

MATERIALS AND METHODS

Raw Material. Rice husks were obtained in a local factory (Procesadora Gallega de Alimentos, Lalín, Pontevedra, Spain), air-dried, homogenized in a single lot to avoid compositional differences among aliquots, and stored until use.

Analysis of the Raw Material. Aliquots from stored samples were subjected to moisture determination (method ISO 638), to ash determination (method ISO 776), and to quantitative acid hydrolysis (method TAPPI T13m). The high-performance liquid chromatography (HPLC) analysis of liquors (*8*) allowed the determination of their contents of cellulose, hemicellulosic polysaccharide constituents, and acetyl groups. The oven-dried weight of the solid phase from quantitative hydrolysis measured the content of Klason lignin after correction for ashes. Uronic acids were determined by the method of Blumenkrantz and Asboe-Hansen (*25*), using galacturonic acid as a standard for quantification, and the content of protein was estimated by elemental analysis (protein = 6.25 N).

Autohydrolysis Processing of Rice Husks. Rice husks and water were mixed at the desired proportions and reacted in a Parr reactor under nonisothermal conditions at a liquor-to-solid ratio of 8 kg/kg oven-dried solid (following the standard heating temperature profile) to reach the maximum xylooligomer concentration (*11*). At the end of treatments, liquors were recovered by filtration, analyzed, and processed as follows.

Analysis of Autohydrolysis Liquors. For analytical purposes, samples of liquors were filtered through 0.45 μm membranes and used for direct HPLC determination of glucose, xylose, arabinose, and acetic acid using the same method employed in the analysis of the raw material. A second sample of liquors was subjected to quantitative posthydrolysis (treatment with 4% sulfuric acid at 121 °C for 30 min), and the reaction products were assayed by the same HPLC method. The increase in the concentrations of monosaccharides and acetic acid caused by posthydrolysis provided a measure of the oligomer concentration and their degree of substitution with acetyl groups (*11*). Uronic acids were assayed as reported for the raw material. The content of raw or processed liquors in nonvolatile compounds (NVCs) was measured by oven drying at 105 °C until constant weight. All determinations were made in triplicate.

Elemental Analysis. Elemental analysis of selected xylooligosaccharide concentrates was carried out using a Thermo Finnegan Flash EA 1112 Analyzer using 130 and 100 mL/min of He and O_2 and a oven temperature of 50 °C. Determinations were made in triplicate.

Acid Soluble Lignin. The acid soluble lignin content of selected xylooligosaccharide concentrates was measured spectrophotometrically after hydrolysis with 4% sulfuric acid at 121 °C for 30 min using the method of Maekawa et al. (*26*). Determinations were made in triplicate.

Concentration of Liquors. Autohydrolysis liquors were concentrated at the desired volume ratio using a centrifugal evaporator (Savant SC210A SpeedVac Plus), operating at the lowest heating power in order to avoid sample degradation.

Solvent Extraction of Liquors. Aliquots of autohydrolysis liquors were extracted with ethyl acetate in three sequential stages at a liquor: solvent mass ratio of 1:1 (kg/kg) per stage. The organic phases were mixed and vacuum-evaporated to dryness in a rotary evaporator to remove both solvent and volatile dissolved compounds, and the remaining solid phase was used to calculate the yield of the autohydrolysis−extraction process. The aqueous phases were assayed for composition using the same methods described for the autohydrolysis liquors.

Solvent Precipitation. Liquors from the ethyl acetate extraction stage were mixed with ethanol, acetone, or 2-propanol at a mass ratio of 4.5 kg solvent/kg liquor and kept for 12 h at room temperature. The solid fraction was collected by membrane (0.45 μm) filtration, weighed, and redissolved in water. Separate aliquots from this solution were assayed for composition using the same methods described for the autohydrolysis liquors. The samples were processed in triplicate, and both average values and standard deviations are persented.

Freeze Drying and Solvent Extraction of Freeze-Dried Solids. Liquors from the ethyl acetate extraction stage were freeze-dried, and the resulting solids were contacted for 12 h with ethanol, acetone, or 2-propanol at room temperature at a mass ratio of 5 kg solvent/kg solid. The insoluble fraction was recovered by membrane filtration, weighed, and redissolved in water. Separate aliquots from this solution were assayed for composition using the same methods described for the autohydrolysis liquors. The samples were processed in triplicate, and both average values and standard deviations are presented.

Ion Exchange Processing of Liquors. Liquors from the ethyl acetate extraction stage were treated with Amberlite IRA 400 (a strong anion exchange, quaternary ammonium, gel type resin, supplied in Cl^- form) and Amberlite IRA 96 (a weak anion exchange, polyamine, macroreticular resin, supplied in free base form). Both resins were obtained from Rohm and Hass. Liquors and resins were contacted for 12 h with gentle agitation at room temperature using 1 kg of resin (o.d. weight)/20 kg of liquor. Samples were processed in triplicate, and both average values and standard deviations are presented.

RESULTS AND DISCUSSION

Raw Material Composition and Characterization of Liquors. Xylooligosaccharides produced by autohydrolysis present complex structures, related to the ones of the xylan fraction. In mild autohydrolysis treatments, a part of the functional groups present in xylan is preserved in oligosaccharides, leading to a variety of compounds with potential biological activity. The structure of low molecular weight oligosaccharides has been considered in recent studies (*27*, *28*) as well as their fermentability (*29*).

Table 1a shows the composition of the rice husk lot employed in this study. The most important data refer to the contents of xylan and xylan substituents (arabinose, uronic acid units, and acetyl groups), which accounted jointly by 20 wt % of the oven-dried sample. Even if hemicelluloses are made up of a xylan backbone with arabinose substituents, arabinose and xylose units show a different susceptibility to hydrolysis reactions. Because of this, and in order to enable an easier understanding of the experimental results, arabinan and xylan are considered as separate polymers. Cellulose and Klason lignin, which are expected to suffer little alteration during mild autohydrolysis processing, accounted jointly for about 57 wt % of the o.d. feedstock. The content of proteins was low (2.5 wt %, o.d. basis), whereas ashes and other fractions (which corresponded mainly to extractives and acid soluble lignin) represented jointly about 20% of the sample o.d. weight.

In the autohydrolysis of xylan-containing feedstocks, xylooligosacchrides behave as reaction intermediates, reaching maximal concentrations under defined operational conditions. **Table 1b** summarizes the effects caused by nonisothermal autohydrolysis carried out under conditions reported as optimal for xylooligosaccharide production (*11*): 24.2% of the o.d. raw material was solubilized in treatments, leading to NVCs (mainly substituted xylooligosaccharides and sugars) and volatile compounds (VCs). Acetic acid (derived from acetyl groups) was the main VC identified, but material balances proved the

Table 1. Raw Material Composition and Effects Caused by Autohydrolysis

(a) Raw Material Composition

component	mass fraction (kg/kg o.d. solid)
cellulose	0.3441
xylan	0.1623
arabinan	0.0133
acetyl groups	0.0143
uronic acids	0.0114
Klason lignin	0.2300
proteins	0.0250
ashes	0.1133
others (by difference)	0.0863
moisture (water)	0.0900

(b) Effects Caused by Autohydrolysis

weight percent of o.d. solid solubilized in reaction	24.2
substrate conversion in VCs (% o.d. weight)	1.40
substrate conversion in NVCs (% o.d. weight)	22.8
cellulose conversion into glucose (%)	0.96
xylan conversion into xylose (%)	4.6
arabinan conversion into arabinose (%)	39.4
cellulose conversion into glucooligosaccharides (%)	7.8
xylan conversion into xylooligosaccharides (%)	64.3
arabinan conversion into arabinooligosaccharides (%)	34.4
acetyl groups conversion into acetic acid (%)	45.4
uronic acids present in soluble components (%)	70.6

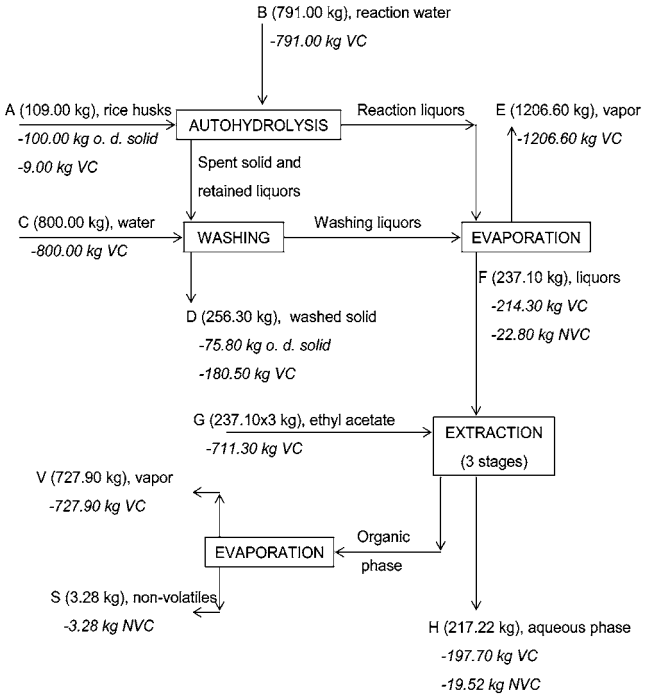

Figure 1. Material balances concerning the autohydrolysis, concentration, and ethyl acetate extraction stages (VC, volatile components; NVC, nonvolatile components).

presence of other VCs (derived from extractives and sugars), which are of minor importance for the purposes of this study. Xylan and arabinan were converted into sugars and sugar oligomers, which conserved most of the acetyl and uronic substituents of the original hemicellulosic polymer.

Figure 1 shows the processing of solid and liquid phases obtained in autohydrolysis experiments. The spent solid was recovered by filtration and washed. The washed solid had a higher cellulose content than the original rice husks (owing to the preferential removal of hemicelluloses during autohydrolysis)

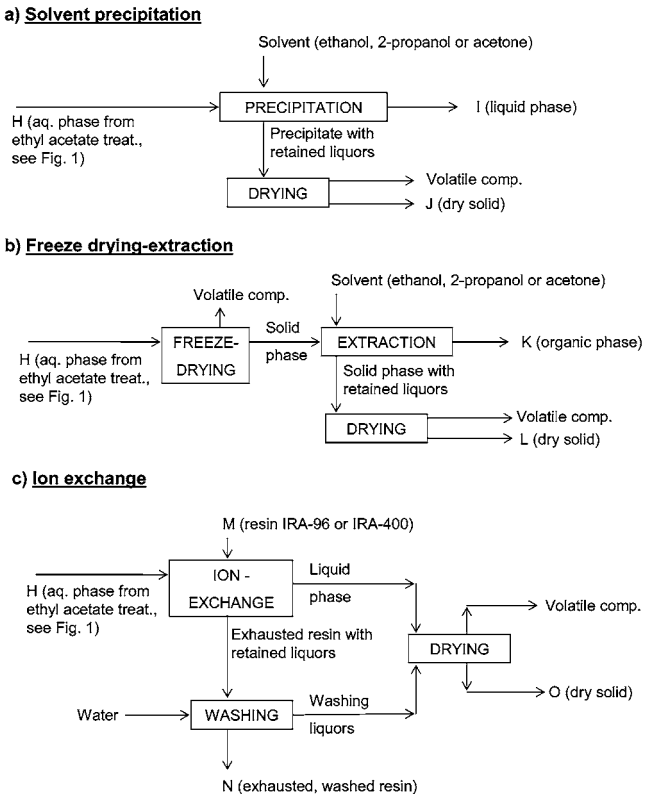

Figure 2. Processes assayed for refining concentrated, ethyl acetate-extracted, autohydrolysis liquors.

and is suitable for further processing (for example, enzymatic hydrolysis). Even though the washing liquors presented low contents of NVC, they were subjected to evaporation together with the autohydrolysis liquors, to provide a better estimate of the yields that could be reached in an integrated, industrial process. Evaporation was carried out at low temperature in a centrifugal evaporator, in which the degradation of products was negligible. **Figure 1** presents material balances to the reaction and evaporation stages in terms of NVCs and VCs (which included the solvent). According to the data in **Figure 1**, the yield of NVC after the evaporation stage was 22.80 kg/100 kg o.d. rice husks.

Ethyl Acetate Extraction of Concentrated Autohydrolysis Liquors. Concentrated autohydrolysis liquors (stream F in **Figure 1**) were made up of NVCs and VCs. As the target compounds in this study (substituted xylooligosaccharides) are nonvolatile and considering that the VC fraction can be directly removed by oven drying, our attention was focused on the processing of the NVC fraction.

The NVC fraction was made up of saccharides (glucose and glucooligosaccharides coming from cellulose, xylose, arabinose, and oligomers of both sugars coming from xylan and arabinan hydrolysis), saccharide substituents (acetyl groups and uronic acids linked to oligosaccharides), and nonsaccharides, denoted "other NVCs", ONVC. The ONVC fraction is expected to contain compounds derived from extractives, acid soluble lignin, and proteins. For food-related applications, purification of the NVC fraction and further insight on its composition are needed.

Table 2 lists data on the composition of the NVC fraction of stream F. Monosaccharides accounted for about 8 wt % of NVC and glucooligosaccharides for 11.8% of NVC. Substituted oligosaccharides, the most important fraction for the objective of this study, are given by the sum of xylooligosaccharides, arabinooligosaccharides, and acetyl groups and uronic moieties

Table 2. NVCs Present in Streams F, H, and J in **Figure 1**

(a) Amounts of NVC and NVC Content of the Considered Streams

	stream F	stream H	stream S
kg NVC	22.80	19.52	3.28
kg NVC/kg stream	0.0962	0.0898	1.0000

(b) Composition of the NVC Fractions

	stream F		stream H		stream S
component	mass fraction (kg/kg NVC)	SD	mass fraction (kg/kg NVC)	SD	mass fraction (kg/kg NVC)[a]
glucose	0.0161	0.0091	0.0137	0.0033	0.0301
xylose	0.0374	0.0012	0.0427	0.0049	0.0055
arabinose	0.0261	0.0004	0.0288	0.0022	0.0102
glucooligosaccharides	0.1180	0.0075	0.1302	0.0002	0.0452
xylooligosaccharides	0.4578	0.0072	0.5081	0.0072	0.1589
arabinooligosaccharides	0.0201	0.0001	0.0229	0.0011	0.0036
acetyl groups-oligosaccharides	0.0343	0.0023	0.0365	0.0012	0.0211
uronic acids	0.0353	0.0011	0.0332	0.0039	0.0480
ONVC	0.2550	0.0135	0.1840	0.0112	0.6774

[a] Calculated by material balances from the average values determined for the compositions of streams F and H in **Figure 1**.

Table 3. NVCs Present in Streams J and I in **Figure 2a** in Operation with Ethanol, 2-Propanol, or Acetone

(a) Amounts of NVC in the Considered Streams

	kg of NVC in stream J	kg of NVC in stream I
ethanol	7.14	12.38
2-propanol	12.84	6.68
acetone	12.05	7.47

(b) Composition of the NVC Fractions

		stream J		stream I
solvent	component	mass fraction (kg/kg NVC)	SD	mass fraction (kg/kg NVC)[a]
ethanol	glucose	0.0051	0.0004	0.0186
	xylose	0.0039	0.0000	0.0650
	arabinose	0.0031	0.0013	0.0435
	glucooligosaccharides	0.2206	0.0224	0.0780
	xylooligosaccharides	0.5746	0.0034	0.4696
	arabinooligosaccharides	0.0224	0.0004	0.0231
	acetyl groups-oligosaccharides	0.0493	0.0048	0.0290
	uronic acids	0.0302	0.0146	0.0349
	ONVC	0.0907	0.0135	0.2383
2-propanol	glucose	0.0031	0.0006	0.0340
	xylose	0.0069	0.0005	0.1114
	arabinose	0.0044	0.0007	0.0756
	glucooligosaccharides	0.1797	0.0168	0.0350
	xylooligosaccharides	0.5858	0.0152	0.3586
	arabinooligosaccharides	0.0233	0.0015	0.0219
	acetyl groups-oligosaccharides	0.0500	0.0111	0.0105
	uronic acids	0.0268	0.0051	0.0454
	ONVC	0.1200	0.0135	0.3076
acetone	glucose	0.0031	0.0006	0.0307
	xylose	0.0081	0.0009	0.0984
	arabinose	0.0048	0.0003	0.0674
	glucooligosaccharides	0.1682	0.0016	0.0689
	xylooligosaccharides	0.5599	0.0256	0.4246
	arabinooligosaccharides	0.0256	0.0017	0.0185
	acetyl groups-oligosaccharides	0.0530	0.0076	0.0098
	uronic acids	0.0394	0.0042	0.0230
	ONVC	0.1379	0.0112	0.2587

[a] Calculated by material balances from the average values determined for the composition of streams H in **Figure 1** and J in **Figure 2a**.

linked to oligomers, which accounted jointly for about 66% of NVC. The content of ONVC corresponded roughly to one-fourth of NVC and could include undesired compounds for food purposes. Ethyl acetate extraction was selected for preliminary refining of the raw autohydrolysis liquors, intending to decrease their content of ONVC. With this processing, a variety of undesired extractive- and lignin-derived compounds (for example, resin and fatty acids, alcohols, esters, waxes, and low molecular phenolics) can be removed (*10*). On the other hand, the ethyl acetate soluble fraction may present valuable antioxidant properties, making it suitable for commercial developments in the food and cosmetic fields (*12*, *13*).

Table 2 lists the experimental data on the composition of the NVC fractions corresponding to the ethyl acetate-extracted

Table 4. NVCs Present in Streams L and K in **Figure 2b** in Operation with Ethanol, 2-Propanol, or Acetone

(a) Amounts of NVC in the Considered Streams

	kg of NVC in stream L	kg of NVC in stream K
ethanol	12.09	7.44
2-propanol	16.41	3.11
acetone	16.98	2.55

(b) Composition of the NVC Fractions

		stream L		stream K
solvent	component	mass fraction (kg/kg NVC)	SD	mass fraction (kg/kg NVC)[a]
ethanol	glucose	0.0049	0.0003	0.0280
	xylose	0.0585	0.0021	0.0170
	arabinose	0.0111	0.0002	0.0574
	glucooligosaccharides	0.1421	0.0042	0.1108
	xylooligosaccharides	0.5154	0.0042	0.4955
	arabinooligosaccharides	0.0186	0.0013	0.0297
	acetyl groups-oligosaccharides	0.0426	0.0014	0.0265
	uronic acids	0.0307	0.0080	0.0372
	ONVC	0.1761	0.0220	0.1979
2-propanol	glucose	0.0082	0.0001	0.0424
	xylose	0.0500	0.0021	0.0041
	arabinose	0.0247	0.0007	0.0502
	glucooligosaccharides	0.1256	0.0028	0.1545
	xylooligosaccharides	0.5180	0.0105	0.4555
	arabinooligosaccharides	0.0234	0.0054	0.0200
	acetyl groups-oligosaccharides	0.0413	0.0014	0.0110
	uronic acids	0.0348	0.0040	0.0246
	ONVC	0.1740	0.0310	0.2377
acetone	glucose	0.0087	0.0001	0.0468
	xylose	0.0476	0.0024	0.0096
	arabinose	0.0260	0.0007	0.0473
	glucooligosaccharides	0.1269	0.0020	0.1520
	xylooligosaccharides	0.5267	0.0145	0.3821
	arabinooligosaccharides	0.0193	0.0051	0.0466
	acetyl groups-oligosaccharides	0.0410	0.0019	0.0063
	uronic acids	0.0343	0.0040	0.0252
	ONVC	0.1695	0.0190	0.2841

[a] Calculated by material balances from the average values determined for the composition of streams H in **Figure 1** and L in **Figure 2b**.

liquors (stream H in **Figure 1**) and organic phase from extraction (stream S in **Figure 1**). **Figure 1** provides data on the distribution of VCs and NVCs among the streams involved in the extraction step. This stage enabled remarkable refining effects: about 38% of the ONVC were removed from the aqueous phase, in comparison with 5% of xylooligosaccharides. However, the ONVC content of extracted liquors (stream H in **Figure 1**) was still too high for the desired purposes, and further purification of concentrated, ethyl acetate-extracted liquors (by solvent precipitation, freeze drying extraction, or ion exchange) was studied (see **Figure 2**).

Solvent Precipitation. Concentrated, ethyl acetate-extracted autohydrolysis liquors were subjected to precipitation tests with selected solvents (ethanol, acetone, or 2-propanol) as a possible method for decreasing their content of ONVC (**Figure 2a**). **Table 3** shows data on the distribution of NVC between streams J and I in **Figure 2a** as well as on the composition of their NVC fractions. Among the solvents tested, ethanol led to the minimal content of ONVC (9 wt % of NVC) but at a limited yield (36.6% of NVC recovery) with poor increase in xylooligosaccharide content (mass fraction of 0.5746 in comparison with 0.5081 in the feed stream). Comparatively, 2-propanol resulted in worse ONVC removal (which accounted for 12% NVC in the concentrate), but the recovery yield increased up to 65.8% for total NVC and 75.8% for xylooligosaccharides. Acetone presented a behavior related to the one observed for 2-propanol, with slightly lower recovery yields.

Solvent Extraction of Freeze-Dried Solids. To assess possible improvements in refining derived from the absence of water, concentrated, ethyl acetate-extracted autohydrolysis liquors were freeze-dried and subjected to extraction with the same solvents employed in the precipitation tests (see **Figure 2b**). **Table 4** lists data on the NVC contents and compositions of the isolates obtained after solvent extraction of freeze-dried solids (stream L in **Figure 2b**), as well as the compositions of the corresponding organic phases (stream K in **Figure 2b**). The recoveries of NVC improved significantly with respect to the precipitation tests (to reach 61.9, 84.1, and 87.0% for ethanol, 2-propanol, and acetone, respectively), as well as the recoveries of xylooligosaccharides (62.8, 85.7, and 90.2% recovery for ethanol, 2-propanol, and acetone, respectively), but the purification effects were poor, as it is shown by the content of ONVC of stream L (mass fraction about 0.17 for all the solvents, in comparison with 0.1840 for stream H) and by the mass fraction of xylooligosaccharides in the concentrate (in the range 0.5154−0.5267, close to the result determined for stream H).

Ion Exchange. As the results obtained in solvent-based processing of extracted autohydrolysis liquors achieved limited purification effects, ion exchange was employed as an alternative method for refining. For this purpose, the concentrated, ethyl acetate-extracted liquors were contacted without previous pH regulation with commercial anionic exchange resins (Amberlite IRA-96 or Amberlite IRA-400; see **Figure 2c**). Amberlite IRA 400 was used in OH^- form, whereas Amberlite IRA96 was

Table 5. NVCs Present in Streams O and N in **Figure 2c** in Operation with Resins IRA-96 or IRA-400

(a) Amounts of NVC in the Considered Streams

	kg of NVC in stream O	kg of NVC in stream N
IRA-96	13.52	6.01
IRA-400	14.48	5.05

(b) Composition of the NVC Fractions

		stream O		stream N
resin	component	mass fraction (kg/kg NVC)	SD	mass fraction (kg/kg NVC)[a]
IRA-96	glucose	0.0051	0.0005	0.0330
	xylose	0.0488	0.0010	0.0288
	arabinose	0.0352	0.0020	0.0141
	glucooligosaccharides	0.1334	0.0011	0.1228
	xylooligosaccharides	0.5755	0.0016	0.3557
	arabinooligosaccharides	0.0317	0.0018	0.0028
	acetyl groups-oligosaccharides	0.0404	0.0030	0.0276
	uronic acids	0.0324	0.0014	0.0348
	ONVC	0.0974	0.0030	0.3804
IRA-400	glucose	0.0052	0.0007	0.0379
	xylose	0.0520	0.0016	0.0160
	arabinose	0.0342	0.0008	0.0130
	glucooligosaccharides	0.1347	0.0035	0.1171
	xylooligosaccharides	0.5825	0.0020	0.2940
	arabinooligosaccharides	0.0300	0.0001	0.0023
	acetyl groups-oligosaccharides	0.0406	0.0028	0.0247
	uronic acids	0.0333	0.0009	0.0328
	ONVC	0.0875	0.0014	0.4622

[a] Calculated by material balances from the average values determined for the composition of streams H in **Figure 1** and O in **Figure 2c**.

employed in free base form because of its own nature (the resin remains in this form at pH > 9.0) (*30*).

Table 5 provides information on the amount and composition of NVC present in streams O and N. The amount of NVC retained by IRA-96 (30.7% of the ones contained in the feed stream) was higher than the one retained by with IRA-400 (25.8%). A similar behavior was observed for the xylooligosaccharide fraction, which was retained in a higher degree by IRA-96 (21.6% of the xylooligosaccharides contained in the feed stream in comparison with 15% for IRA-400). Considering the similar degrees of ONVC removal achieved for both resins (63.3% for IRA-96 and 64.7% for IRA-400) and the ONVC mass fraction of stream O (0.097 in the case of IRA-96 and 0.088 in the case of IRA-400), it can be concluded that the latter resin was the most favorable one and that ion exchange resulted in better purification than solvent precipitation of freeze drying solvent extraction. The yield in concentrate is directly given in **Table 5** (14.48 kg/100 kg o.d. rice husks).

On the basis of the above experimental data, the isolate from treatment with IRA-400 (corresponding to stream O in **Figure 2c** with the composition listed in **Table 5**) was selected for further studies. According to compositional data, the concentrate contained 9.1% monosaccharides, 74.7% oligosaccharides (which are measured as "monosaccharide equivalents" because of the analytical methodology employed), 7.4% of oligosaccharide substituents (acetyl groups and uronic acids), and 8.8% of unknown ONVC. Even if the literature information on the refining of xylooligosaccharide-containing solutions is scarce, it can be remarked that the processing scheme selected in this work compares favorably with the results reported for technologies involving two-stage reaction followed by chromatographic separation and ion exchange (*19*).

Considering that further characterization of ONVC could assess additional properties important for food applications, the visible UV spectra of aqueous solutions of the final isolate were recorded. The results showed typical absorption bands for phenolic compounds and melanoidins.

As free, low molecular phenolics were removed during the physicochemical processing of the samples, the occurrence of phenolic compounds was ascribed to the presence of phenolic acids chemically bonded to the xylan backbone, which could be solubilized by treatment with 4% sulfuric acid and quantified as acid soluble lignin. This operational procedure led to an acid soluble lignin content of 3.6%. It can be noted that this fraction presents antioxidant properties, which could confer additional functional properties to the concentrate. In this field, fiber with antioxidant properties has been proposed for utilization in food (*31*).

Melanoidins are Maillard reaction products generated during the autohydrolysis step by reaction between amino acids and sugars. As melanoidins are complex compounds, generated by many simultaneous reactions and difficult to quantify, the sample was subjected to elemental analysis in order to assess its nitrogen content, which provides an estimate of its importance. The experimental results [average elemental nitrogen content of 0.61 wt % in o.d. basis, with a standard deviation (SD) of 0.014] can be converted into the equivalent amount of protein using the factor 6.25 g protein/g elemental nitrogen, leading to the conclusion that proteins accounting for 3.8 o.d. weight percent of the isolate (which correspond to 22% of the proteic fraction of rice husks) were converted into melanoidins. As this kind of compound also presents antioxidant activity, they could contribute to the functional properties of the concentrate.

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