Supporting information for

Hydrolysis of chitosan to yield levulinic acid and 5-hydroxymethyl-furfural in water under microwave irradiation

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Contents:

Experimental

Materials

Chitosan low, medium and high molecular weights (75-85% deacetylated), chitin (from crab shell), tin (IV) chloride pentahydrate 98% and 2,5-dihydroxybenzoic acid 98% (DHB) were purchased from Aldrich. Further samples of chitin and chitosan from crab and shrimp were provided gratis by ChitinWorks America LLC. All other catalysts tested were purchased in 98% purity or greater from Fisher Scientific, Alfa Aesar, Aldrich or Strem Chemicals. (+/-)-3-hydroxy-gamma-butyrolactone 96%, 5-hydroxymethylfurfural 98% and levulinic acid (98%) were purchased from Alfa Aesar. Ethyl acetate (HPLC

grade) 99.8% was purchased from EMD Chemicals Inc.. Deionized water was obtained *via* a Nanopure II system (manufactured by Barnstead/Thermolyne, USA) using distilled water as a source for the inlet feed.

General procedure for the hydrolysis of chitosan and product extraction

Chitosan (100 mg) was mixed with a specific volume of aqueous solution (or suspension in the case of Amberlyst) of of known concentration. The mixture was heated to the desired temperature under microwave irradiation using a Biotage initiator 2.5 instrument for a specific period of time. After this time, an aliquot of the reaction mixture (Concentrated reactions for LA analysis, V = 250 μL; Dilute reactions for 5-HMF analysis, V = 500 μL) was mixed with 3-gamma-hydroxy-butyrolactone (internal standard) and extracted with 3 × 2 mL ethyl acetate. After each addition of ethyl acetate, the mixture was vortex-mixed at high speed for 30 seconds and then centrifuged at 1500 rpm for 2 min. The combined ethyl acetate layers were transferred to a quartz tube and evaporated to dryness at 45 °C using a Radleys Greenhouse Blowdown Evaporator under a stream of nitrogen. The contents of the dried tube were reconstituted in 1 mL ethyl acetate for GC-MS analysis.

LA and 5-HMF identification

After hydrolysis of chitosan, extraction with ethyl acetate and evaporation of the solvent, the dried residue was dissolved in $CDCl_3$. ^{1}H-NMR spectra obtained using a Bruker AVANCE 500MHz spectrometer showed the presence of LA and 5-HMF. GC-MS was also used for characterization and quantification of the products. Samples were compared with authenticated samples and with the NIST database.

LA: ^{1}H-NMR δ_H (298 K, 500MHz; $CDCl_3$; Me_4Si) 2.17 (s, 3H), 2.64 (t, 2H, J 6.4 Hz) and 2.77 (t, 2H, J 6.4 Hz). MS *m/z* 116 (10%), 101 (13), 99 (9), 73 (28), 57 (7), 56 (100), 55 (37).

5-HMF: 1H-NMR δ_H (298 K, 500 MHz; $CDCl_3$; Me_4Si) 4.73 (s, 2H), 6.53 (d, 1H, J 3.5 Hz), 7.22 (d, 1H, J 3.5 Hz) and 9.62 (s, 1H). MS *m/z* 126 (61%), 109 (9), 97 (100), 81 (6), 69 (31), 53 (16).

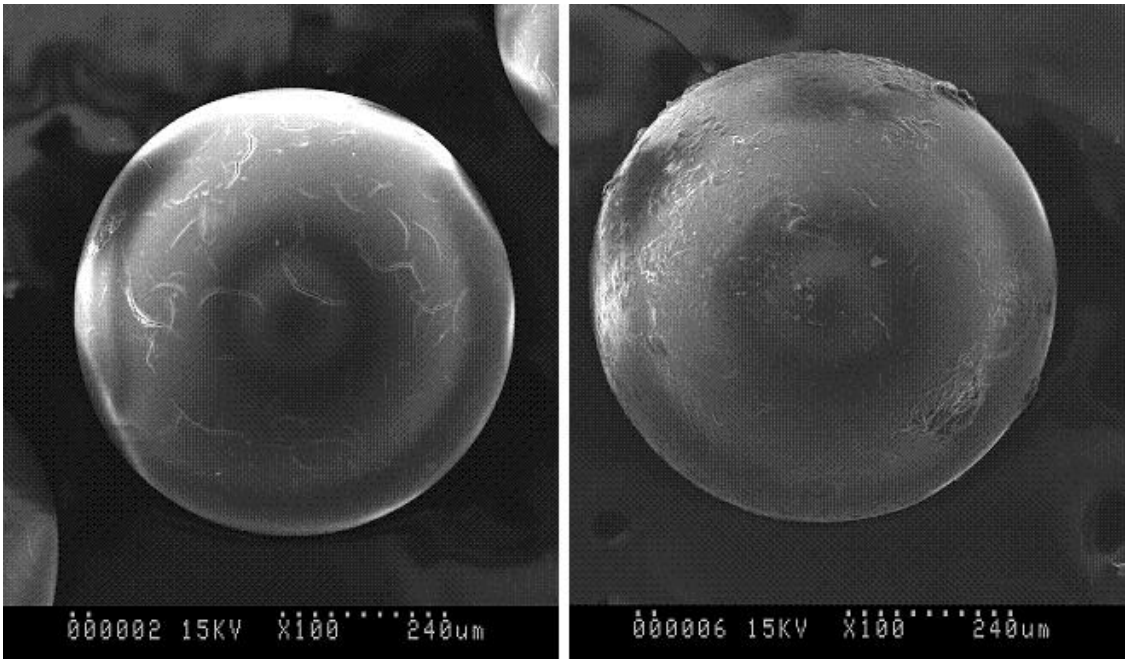
Gas chromatography-mass spectrometry (GC-MS) determination of LA and 5-HMF

5-HMF and LA were analyzed using an Agilent Technologies 7890 GC with 5975 MSD. 1 μL of reconstituted sample was injected through a 7683B Series Injector using a split mode of 50%. The GC separation was done using a DB5 column at a flow rate of 1 mL/min He 99.999%. The oven temperature was programmed as follows: 50 °C (hold 1 min), 25 °C/min to 150 °C, 20 °C/min to 170 °C and 80 °C/min to 250 °C for 3 min. (The total run time was 10 min). Products were detected using a 5975C VLMSD with Triple-Axis Detector (*m/z* 50-250). Under these conditions, the retention times of LA, 3-hydroxy-gamma-butyrolactone and 5-HMF were 4.63, 5.27 and 5.71 min., respectively. R^2 of LA and 5-HMF calibration curves were 0.9978 and 0.9990, respectively. Accuracy was greater than 91% and RSD was less than 3.3% for both products. The limit of detection (LOD) and limit of quantitation (LOQ) of LA were 8 and 26 ppm, respectively. The LOD and LOQ of 5-HMF were 2 and 8 ppm, respectively.

Matrix assisted laser desorption ionization (MALDI) mass spectrometry

MALDI analysis was conducted using AB SCIEX 4800 MALDI TOF/TOF analyzer. 2,5-dihydroxybenzoic acid (DHB) was used as a matrix. A DHB solution was prepared in water (10 mg/mL). The DHB solution was mixed with a portion of the reaction mixture

in 1:1 volume ratio. Then, 1 µL of the mixed solutions was spotted on the MALDI plate. The spot was dried at room temperature. Then, the plate was loaded into the instrument for analysis.



(a) **(b)**

Fig S1. SEM analysis of Amberlyst 15 (a) before microwave reaction, there are cracks on the surface of the beads (b) after microwave reaction, biopolymer deposited on the surface of the beads and filled the cracks.

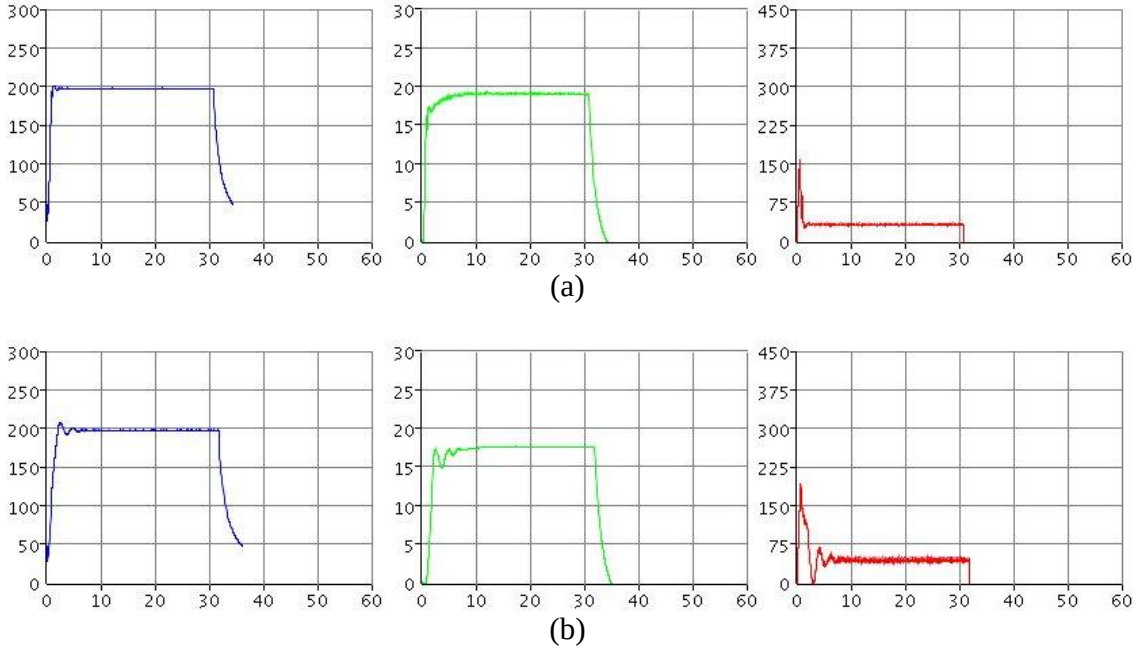


Fig S2. Microwave reactions graphs (a) LA and (b) 5-HMF optimum conditions. Each set consists of three graphs temperature (°C), pressure (bar) and power (W), respectively against time (min) on x-axes.

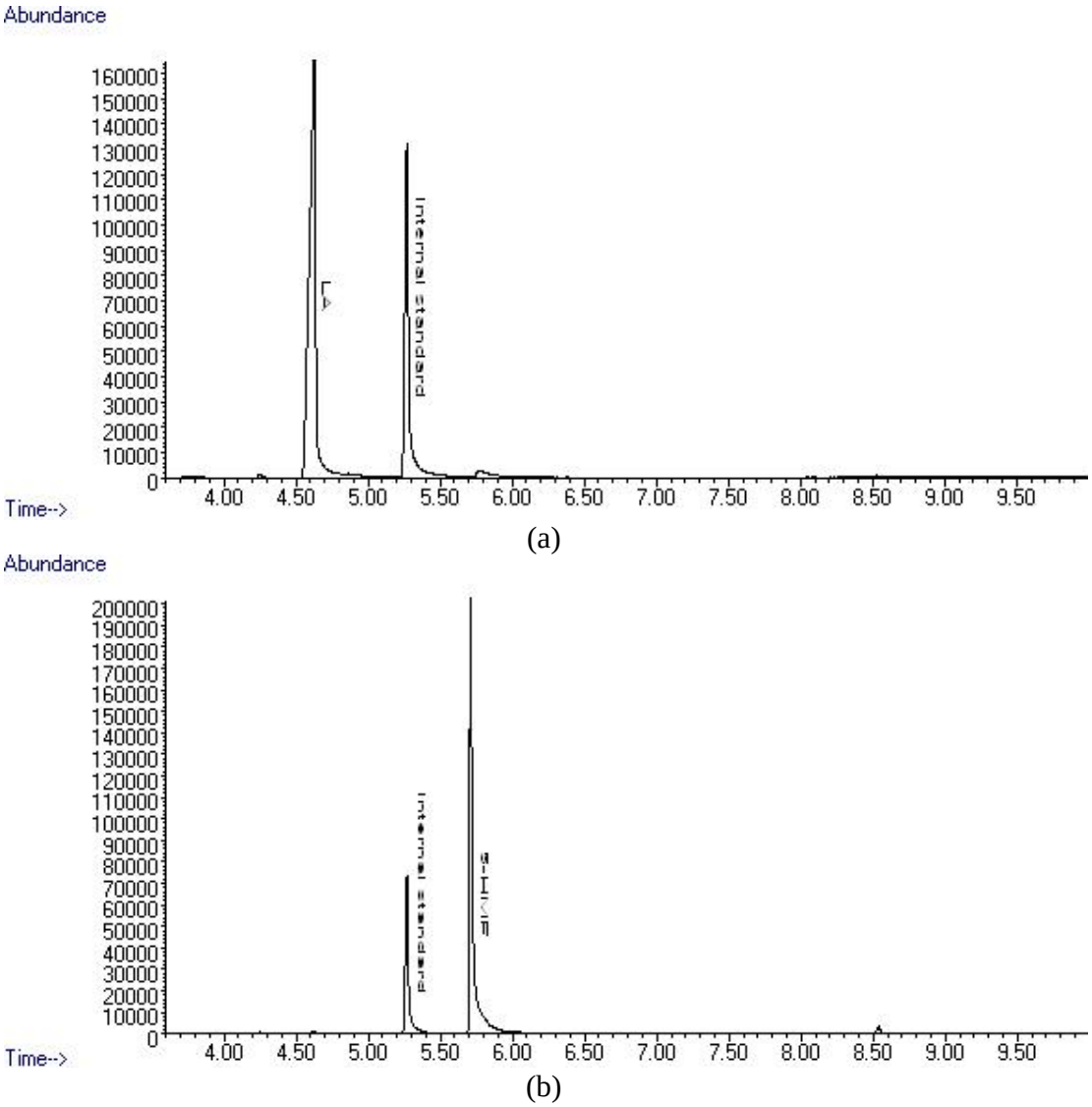
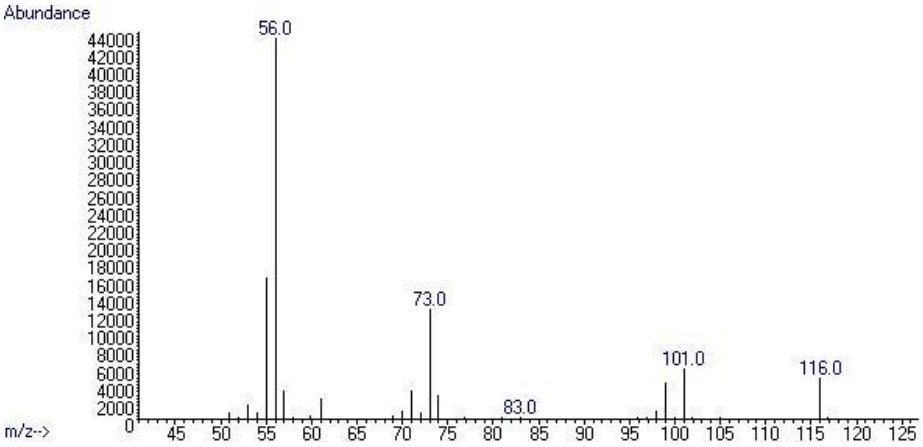
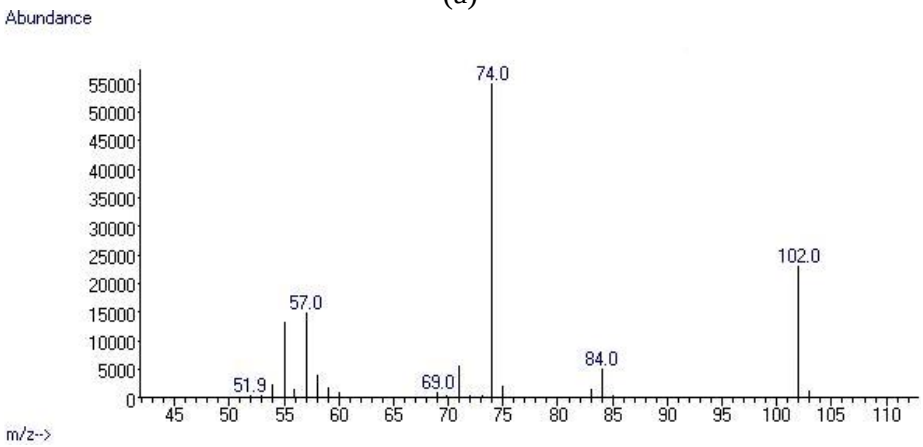


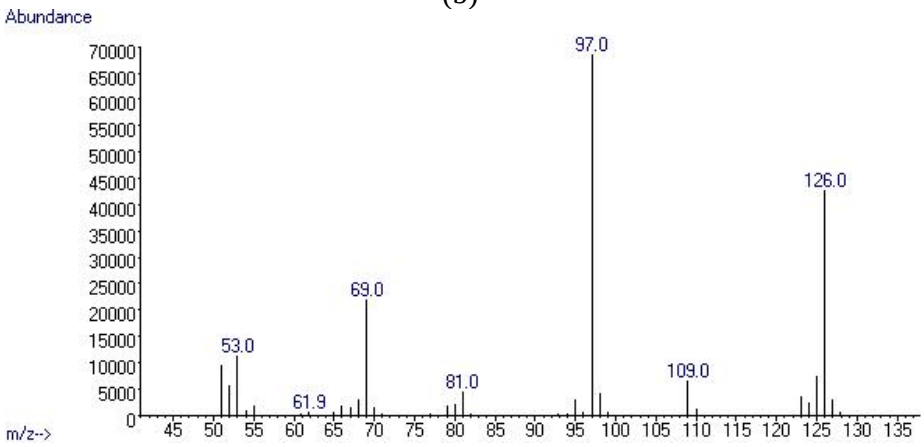
Fig S3. GC chromatograms (a) LA, t_R = 4.63 min and (b) 5-HMF, t_R = 5.71 min.



(a)



(b)



(c)

Fig S4. Mass spectra (a) LA, (b) 5-HMF and (c) Internal standard