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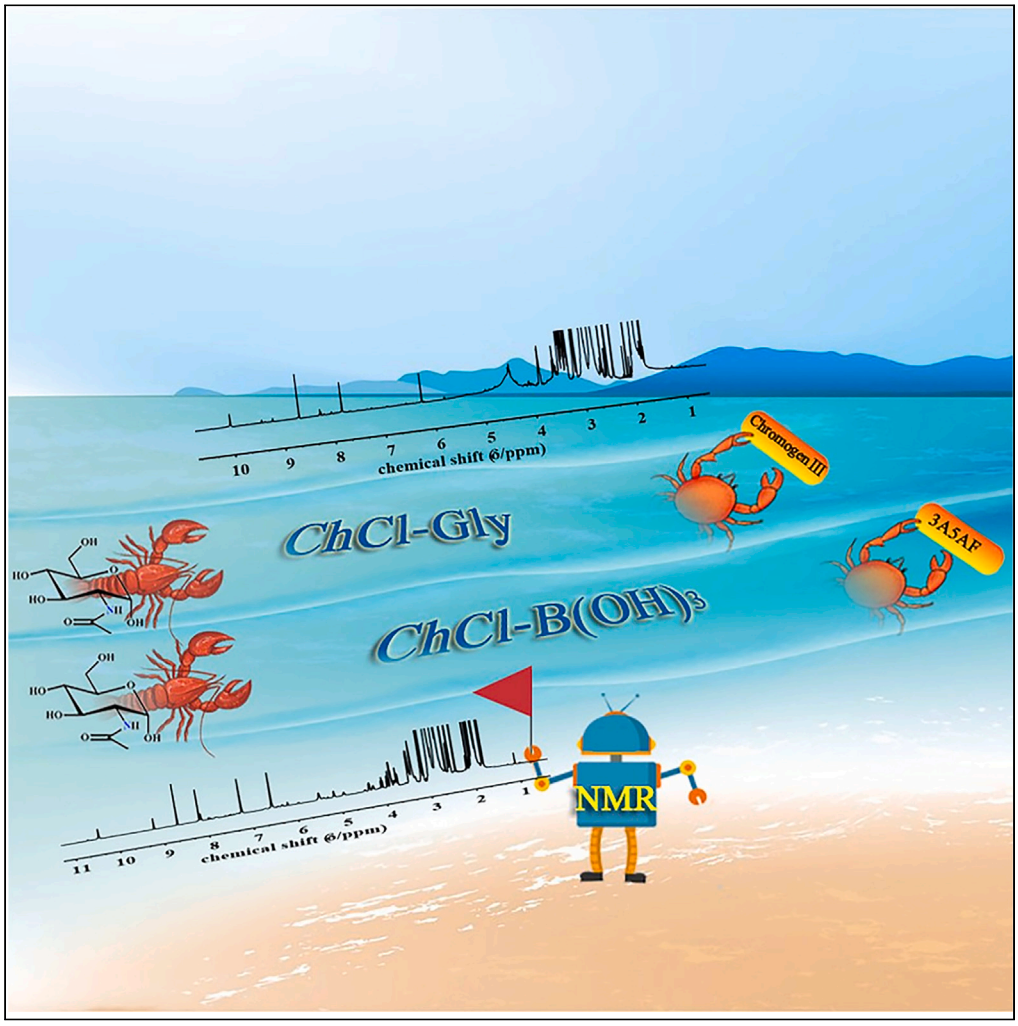

Jiancheng Zhao, Christian Marcus Pedersen, Honghong Chang, Xianglin Hou, Yingxiong Wang, Yan Qiao

wangyingxiong@tyut.edu.cn (Y.W.)
qiaoy@sxicc.ac.cn (Y.Q.)

Highlights

GlcNAc was promoted by DESs to selectively produce Chromogen III or 3A5AF

Reaction intermediates were detected by *in situ* NMR experiments

The relationship between DESs and GlcNAc was investigated by NMR technologies

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Switchable product selectivity in dehydration of *N*-acetyl-D-glucosamine promoted by choline chloride-based deep eutectic solvents

Jiancheng Zhao,[1,2] Christian Marcus Pedersen,[3] Honghong Chang,[4] Xianglin Hou,[1,2] Yingxiong Wang,[1,5,6,*] and Yan Qiao[1,2,*]

SUMMARY

Herein, we report choline chloride-based deep eutectic solvents (DESs) promoted conversion of *N*-acetyl-D-glucosamine (GlcNAc) into nitrogen-containing compounds, i.e., 3-acetamido-5-(1′,2′-dihydroxyethyl) furan (Chromogen III) and 3-acetamido-5-acetylfuran (3A5AF). The binary deep eutectic solvent choline chloride-glycerin (ChCl-Gly), was found to promote the dehydration of GlcNAc to form Chromogen III, which reaches a maximum yield of 31.1%. On the other hand, the ternary deep eutectic solvent, choline chloride-glycerol-B(OH)$_3$ (ChCl-Gly-B(OH)$_3$), promoted the further dehydration of GlcNAc into 3A5AF with a maximum yield of 39.2%. In addition, the reaction intermediate, 2-acetamido-2,3-dideoxy-D-*erythro*-hex-2-enofuranose (Chromogen I), was detected by *in situ* nuclear magnetic resonance (NMR) techniques when promoted by ChCl-Gly-B(OH)$_3$. The experimental results of the ^{1}H NMR chemical shift titration showed ChCl-Gly interactions with α-OH-3 and α-OH-4 of GlcNAc, which is responsible for promoting the dehydration reaction. Meanwhile, the strong interaction between Cl$^-$ and GlcNAc was demonstrated by ^{35}Cl NMR.

INTRODUCTION

Nitrogenous fine and platform chemicals have broad application prospects and a huge market demand. They find wide applications in the fields of advanced functional materials, pharmaceuticals, agrochemicals, nutrition, textiles, polymers and surfactants are hence economically very attractive.[1,2] The industrial synthesis of nitrogenous chemicals mostly relies on the Haber-Bosch process, which is an energy-intensive and environmentally unfriendly route.[3,4] In the context of increasingly strict environmental and resource protection policies, it is particularly critical to explore efficient and green reaction paths to prepare nitrogenous chemicals.

Chitin biomass is the largest nitrogen-containing biomass resource in the world. It mainly comes as an industrial waste product from shells and bones of marine animals such as shrimps and crabs, with abundant reserves and low cost.[5–8] Chitin biomass can be converted into a variety of value-added chemicals through several reaction pathways (shown in Scheme 1).[5,9–13] Converting chitin biomass waste into nitrogen-containing fine chemicals is an economic and green path while achieving the goals of carbon and nitrogen fixation.[6,14] This also complies with the requirements of atom economy in the principles of green chemistry.[15–17] Hence, the production of organic nitrogenous chemicals from chitin and its monomer *N*-acetyl-D-glucosamine (GlcNAc) has attracted increasing attention.[18] At present, research teams have successfully converted chitin or its monomer GlcNAc into the nitrogenous chemicals 2-acetamido-2,3-dideoxy-D-*erythro*-hex-2-enofuranose (Chromogen I), 3-acetamido-5-(1′,2′-dihydroxyethyl) furan (Chromogen III), 3-acetamido-5-acetylfuran (3A5AF), etc.[7,13,19–22] These compounds are widely used in the agriculture, food and pharmaceutical industries, as they contain valuable functional groups such as amides, ketones and other active groups.[6,23–25] However, because of the complexity of the biorefinery reactions, efficient and highly selective conversion of GlcNAc remains a challenge. Therefore, it is necessary to design improved green catalysts based on mechanistic insight. Hence, exploring the reaction mechanism for the efficient conversion of GlcNAc is key to develop new efficient methods for biomass valorization.

[1]Shanxi Engineering Research Center of Biorefinery, Institute of Coal Chemistry, Chinese Academy of Sciences, 27 South Taoyuan Road, Taiyuan 030001, China

[2]Center of Materials Science and Optoelectronics Engineering, University of Chinese Academy of Sciences, Beijing 100049, China

[3]Department of Chemistry, University of Copenhagen, Universitetsparken 5, 2100 Copenhagen, Denmark

[4]Shanxi Tihondan Pharmaceutical Technology Co., Ltd., Jinzhong 030600, China

[5]College of Chemistry, Taiyuan University of Technology, 79 Yingze West Street, Taiyuan, Wanbailin District 030024, China

[6]Lead contact

*Correspondence: wangyingxiong@tyut.edu.cn (Y.W.), qiaoy@sxicc.ac.cn (Y.Q.)

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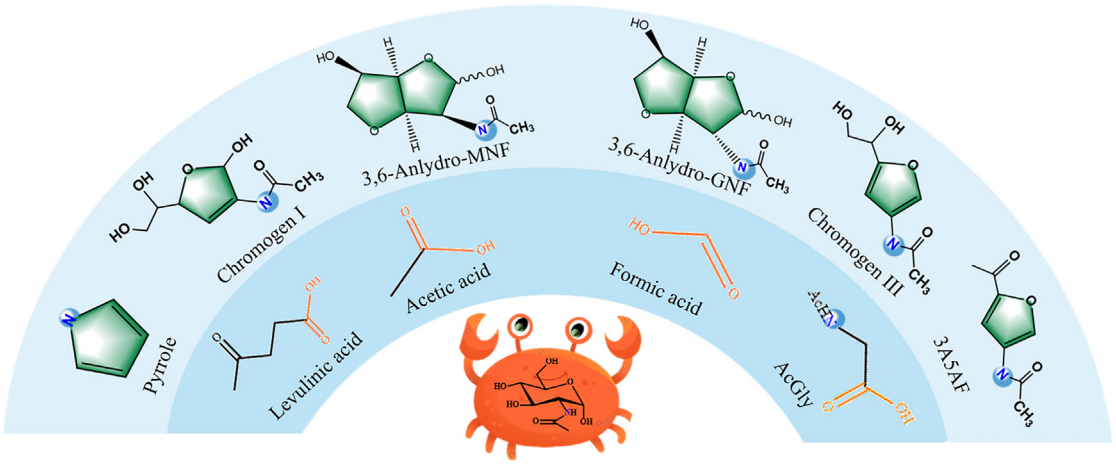


Scheme 1. The main products of the conversion of GlcNAc reported in the literature[9,13,19,33]

Ionic liquids, high-temperature water and enzyme-chemical have been applied for the conversion of GlcNAc to nitrogen containing platform molecules, among which ionic liquids have received great interest among researchers (shown in Table S1). For instance, Zang's team used chitin or GlcNAc as raw materials, using ionic liquids [PDCMPi]Cl, [CMPy]Cl, [TEA]Cl, [Pyz]Cl or [Gly]Cl to catalyze the reaction to obtain 3A5AF in a maximum yield of 69.54%.[26–30] Chen et al. converted chitin in DMA with $B(OH)_3$, LiCl and HCl into 7.5% of 3A5AF.[5] Omari et al. used $B(OH)_3$/NaCl to catalyze the reaction of GlcNAc in DMA at 200°C obtained 62% of 3A5AF.[31] Drover et al. catalyzed GlcNAc to produce 60% of 3A5AF by microwave heating at 180°C with [BMim]Cl and $B(OH)_3$.[32] Osada et al. catalyzed the reaction of GlcNAc at 220°C and 25 MPa in water into 34.5% Chromogen III and a small amount of 3A5AF.[33] Chen et al. used chitinolytic enzyme and NH_4SCN to directly catalyze the reaction of chitin with the addition of cocatalyst $CaCl_2$ in DMA obtained 56.66% of 3A5AF.[34] Although, ionic liquids have excellent performance for the GlcNAc biorefinery reactions, their tedious preparation process, high cost, and difficulty in storage limit large-scale applications. The conversion of GlcNAc by enzyme-chemical and high temperature water method is more complicated and higher requirements for equipment. In addition, the reaction product selectivity is very difficult to control, an issue that has not yet been resolved.

DESs containing hydrogen bond acceptors (HBA) and hydrogen bond donors (HBD) have been reported.[35] Compared with more expensive ionic liquids, DES is favored because of its lower cost and easier storage.[20,36] Its performance, such as catalysis, extraction, and biomass dissolution, can be tuned through the targeted design of the HBA and HBD couple.[37] These properties promote DESs as efficient and green alternatives to ionic liquids. For example, Sertuses et al. use choline chloride-based DESs to catalyze cellulose into 5-hydroxymethylfurfural (5-HMF), levulinic acid (LA), furfural and formic acid.[38] Filonenko et al. reported the transformation of glucose into deoxyfructosazine and fructosazine in ammonium formate-based solvents.[39] Our team uses ternary deep eutectic solvents catalyzed D-glucosamine self-condensation to deoxyfructosazine.[40]

In this study, we developed choline chloride-based DESs by a simple and low-cost method for the transformation of GlcNAc selectively into Chromogen III or 3A5AF. The effects of reaction conditions, such as reaction solvent, temperature, DESs dosage, substrate concentration and reaction time on the 3A5AF and Chromogen III yields were investigated. The product selectivity was achieved by simple modifying the HBD part of DESs. In addition, the interaction between GlcNAc and DESs was explored by ^{1}H NMR chemical shift titration and ^{1}H diffusion-ordered spectroscopy (DOSY) NMR. This study can take advantage of the high solubility and catalytic activity of DESs in the conversion of biomass resources, and provides ideas for the catalytic conversion of chitin-based biomass by DESs.

RESULTS AND DISCUSSION

ChCl-polyol DESs promote the dehydration of GlcNAc

A DESs system using ChCl as HBA, and ethylene glycol, 1,3-butanediol, glycerol as HBD for the dehydration of GlcNAc was prepared. Of interest, we found that such simple DESs can promote the GlcNAc dehydration to form Chromogen III (shown in Table 1 and Figure S1), which is in agreement with our previous work using the

Table 1. Yield of Chromogen III or 3A5AF by dehydration of GlcNAc promoted by different DESs

DES	HBD	ChCl:HBD (molar ratio)	Chromogen III yield (%)[a]	3A5AF yield (%)
ChCl- ethylene glycol	ethylene glycol	1:1	trace	none
ChCl-1,3-butanediol	1,3-butanediol	1:1	14.5	none
ChCl-Gly	glycerol	1:1	26.4	trace

[a]Reaction condition: 1 mmol GlcNAc, 2 mmol DESs, 5 mL DMA, 140°C, 2 h.

ionic liquid 1-allyl-3-methylimidazolium chloride as catalyst.[41] Under the same reaction conditions, the GlcNAc dehydration promoted by ChCl-ethylene glycol only produces trace amount of Chromogen III. When the reaction was mediated by ChCl-1,3-butanediol, the yield of Chromogen III was 14.5%. When using ChCl-Gly as the promoter the yield of Chromogen III reached 26.4% with a trace amount of 3A5AF. According to Zang's report, the number of hydroxyl groups on the HBD can influence the performance of DESs, i.e. HBD with more hydroxyl groups showed a higher performance in the biorefinery reaction.[28]

Owing to its excellent catalytic performance compared with other DESs composed of choline chloride-polyols, ChCl-Gly was used for the optimization of the reaction conditions. The effects of four solvents, i.e. DMSO, DMF, DMA and NMP, on the yield of Chromogen III were investigated. The results show that the main product was Chromogen III when using ChCl-Gly, and the maximum yield of Chromogen III in DMA was 26.4% at 140°C (shown in Table S2). At lower temperature GlcNAc dehydration to form Chromogen III cannot be observed. When the temperature exceeds 140°C, the decrease of Chromogen III yield results from production of humin by undesired side reactions. In addition, the experiment was carried out with the ratio of DESs:GlcNAc being 1:1, 2:1, 3:1 and 4:1 in order to explore the effect of the DESs dosage on the Chromogen III yield. The optimal molar ratio between DESs and GlcNAc for obtaining Chromogen III was found to be 2:1. Excessive DESs makes the dehydration reaction of GlcNAc more vigorous, which reduces the selectivity of the reaction, and ultimately leads to a decrease in the yield of Chromogen III. Using the optimal reaction solvent, temperature, DES dosage the effect of reaction time on the yield of Chromogen III was studied. The maximum yield of Chromogen III reached 31.1% after 140 min (shown in Figure 2).

In order to study the reaction mechanism of the DESs promoted GlcNAc conversion, we performed *in situ* NMR experiments using ChCl-Gly (shown in Figure 1). After 20 min of reaction, the signals of GlcNAc (such as α-NH at 7.6 ppm and α-OH-1 at 6.4 ppm of GlcNAc) disappeared from ^{1}H NMR spectrum. As the reaction progress, the signal of Chromogen III (6.2 ppm, 7.8 ppm) gradually increased from 60 min. The main product of the ChCl-Gly mediated GlcNAc dehydration was Chromogen III, with a small amount of 3A5AF which could be observed by NMR after 180 min.

Dehydration of GlcNAc promoted by DESs with $B(OH)_3$ as HBD

It is generally accepted that the conversion of GlcNAc to Chromogen I, Chromogen III, and 3A5AF is a stepwise process of dehydration (shown in Figure S2). The main product of the ChCl-Gly mediated GlcNAc dehydration was Chromogen III. We aim at controlling the distribution of dehydration products through the design of DESs. Boronic esters are key for regioselectivity in many chemical reactions and their catalytic activity have recently been demonstrated by Hou et al.[42] On this basis, we prepared the DESs ChCl-$B(OH)_3$ and ChCl-Gly-$B(OH)_3$ to compare their performance in GlcNAc dehydration.

As shown in Figure 3, when $B(OH)_3$ was used as the HBD in the binary DES ChCl-$B(OH)_3$, the product selectivity changed significantly. Under the same conditions, the yields of Chromogen III and 3A5AF were 8.5% and 26.8% respectively after 2 h reaction. In the literature, the yield of 3A5AF was only 10% when using boric acid alone,[31] which indicate that both components in the DESs play a role. After 20 min reaction, the GlcNAc signal had disappeared completely. Chromogen III and a small amount of Chromogen I were detected during the reaction. After 60 min, with the increase of intensity of 3A5AF signals (7.2 ppm, 8.2 ppm, 10.2 ppm), the signal of Chromogen III gradually weakened and almost disappeared.

We further investigated the GlcNAc reaction using ternary DES, ChCl-Gly-$B(OH)_3$. Using this DES, the yield of 3A5AF increased significantly to 37.7%. According to Zang et al., the alcoholic hydroxyl group has a

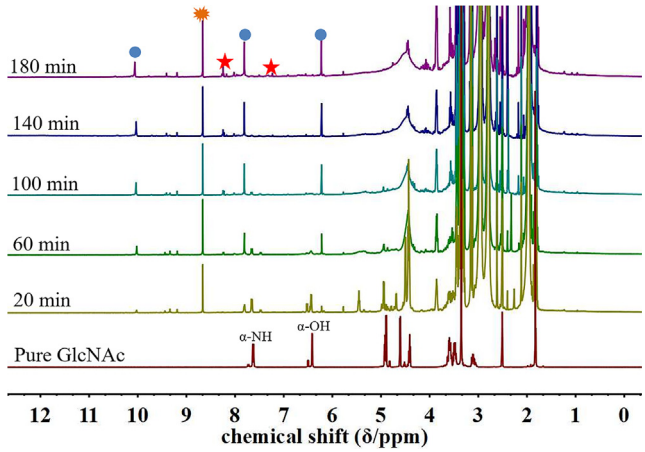


Figure 1. *In situ* ^{1}H NMR spectra of GlcNAc promoted by ChCl-Gly in DMA at 140°C for 3 h. Notes: ● Chromogen III, ★ 3A5AF, ✹ Pyrazine

promoting effect on GlcNAc to 3A5AF reaction.[28] The ability of glycerol to enhance the $B(OH)_3$ acidity may also increase the reaction yield. In the reaction, promoted by ternary DESs ChCl-Gly-$B(OH)_3$, a clear signal of the intermediate Chromogen I (6.3 ppm, 7.9 ppm) was still detected after 100 min (shown in Figure 4). Compared with the reaction mediated by binary DESs, ChCl-Gly and ChCl-$B(OH)_3$, the signal intensity of Chromogen I is enhanced when using ChCl-Gly-$B(OH)_3$. Also, the yield of 3A5AF increased significantly, whereas the yield of by-product acetic acid (9.2 ppm, 2.4 ppm) decreased. Thus, product selectivity and performance of the reagents system can be controlled by the design of DESs. The role of DESs in the reaction and their interaction with GlcNAc will be explained below.

The ternary DESs system ChCl-Gly-$B(OH)_3$ was used to optimize the reaction conditions (shown in Table S3). Similarly, DMA exhibited an excellent performance in this reaction with 37.7% yields of 3A5AF when the reaction was performed at 140°C using a molar ratio of DESs to GlcNAc of 2:1. The yields of intermediates Chromogen I and Chromogen III gradually decreased with increasing reaction time, and the yield of 3A5AF reached a maximum of 39.2% after 160 min (shown in Figure 5).

Effect of GlcNAc concentration

Meanwhile, adjusting the amount of GlcNAc to investigate the effect of substrate concentration on the yield of Chromogen III or 3A5AF when reacting with ChCl-Gly or ChCl-Gly-$B(OH)_3$ as catalyst (shown in Table S4). When ChCl-Gly catalyzes the reaction, the yield of Chromogen III decreases to 10.6% and 3.5% as the amount of GlcNAc is adjusted to 2 and 3 mmol. After increasing the amount of GlcNAc, most of it was dehydrated to Chromogen III and a small amount of 3A5AF was generated when catalyzed by ChCl-Gly-$B(OH)_3$.

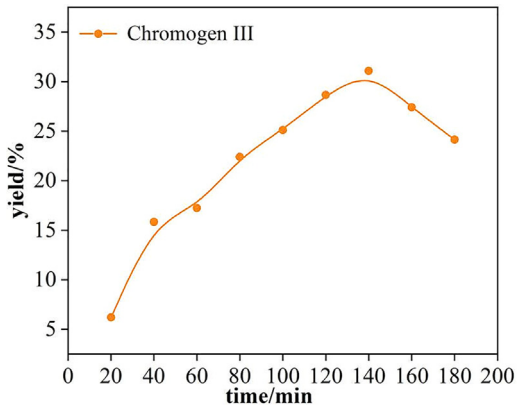


Figure 2. Effect of reaction time on Chromogen III yield under normal atmospheric reflux

Reaction conditions: 1 mmol GlcNAc, 2 mmol ChCl-Gly, 5 mL DMA, 140°C.

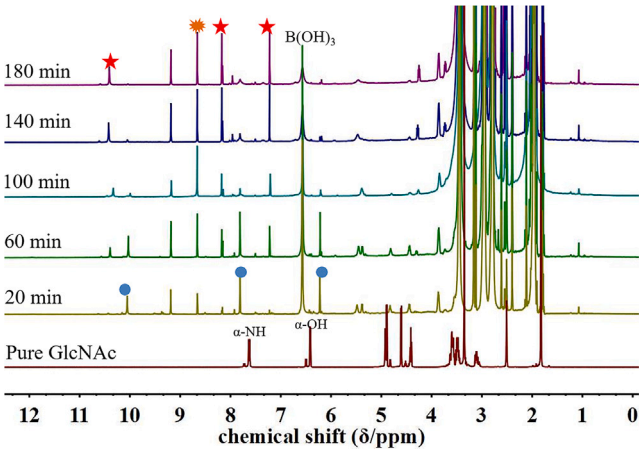


Figure 3. *In situ* ^{1}H NMR spectra of GlcNAc promoted by ChCl-B(OH)$_3$ in DMA at 140°C for 3 h
Notes: ● Chromogen III, ★ 3A5AF, ✹ Pyrazine.

Recycling experiment

Reusability is one of the evaluation criteria for green catalysts and a necessary requirement for green chemistry. The mixture of ChCl-Gly-B(OH)$_3$ catalyzed reaction was treated and the ChCl-Gly-B(OH)$_3$ obtained was cycled under the same reaction conditions. After 2 h of reaction, 31.1% Chromogen III was obtained, and only trace 3A5AF was produced. Although the ChCl-Gly-B(OH)$_3$ showed almost no activity in the third cycle reaction. We speculated that these were because of the complexation of B(OH)$_3$ and GlcNAc in the reaction process, which resulted in the loss of B(OH)$_3$, so that GlcNAc was only dehydrated to Chromogen III during the cyclic reaction. In addition, we treated the mixture after the catalytic reaction of ChCl-Gly in the same way, unfortunately, it lost activity in the second cycle reaction. This may be because of the shortcomings of DES treatment and recycling methods that require further research.

Interactions between GlcNAc and DESs

The interactions between DESs and GlcNAc were explored by ^{1}H NMR chemical shift titration in DMSO-d_6, which is a weakly coordinating reagent and suitable for the exploration of intermolecular interactions. GlcNAc has dominant two isomers, α-pyranose and β-pyranose, and the α-pyranose accounted for more than 95% in DMSO-d_6.[43] Therefore, we mainly explored the interaction between DESs and α-pyranose in the ^{1}H NMR chemical shift titration experiments. Firstly, ^{1}H NMR chemical shift titration experiments were performed with different molar ratios of ChCl-Gly to GlcNAc. ^{1}H NMR spectra and Δδ values of α-pyranose

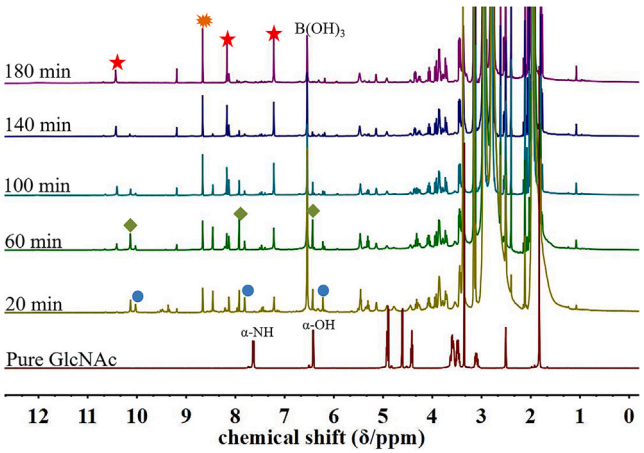


Figure 4. *In situ* ^{1}H NMR spectra of GlcNAc dehydration mediated by ChCl-Gly-B(OH)$_3$ in DMA at 140°C for 3 h
Notes: ◆ Chromogen I, ● Chromogen III, ★ 3A5AF, ✹ Pyrazine.

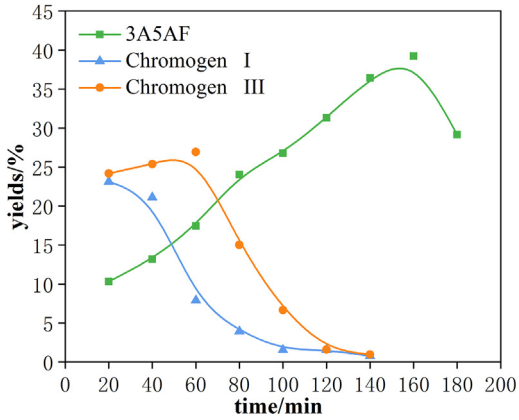


Figure 5. Effect of reaction time on intermediate and 3A5AF yield

Reaction conditions: 1 mmol GlcNAc, 2 mmol ChCl-Gly-$B(OH)_3$, 5 mL DMA, 140°C.

chemical shift are shown in Figure 6A and Figure S3.[41] The proton signal of α-pyranose moves downfield with increasing ratio of ChCl-Gly, and the changes of α-OH-3 and α-OH-4 are the most significant. This chemical shift drift suggests that ChCl-Gly primary interacts with α-OH-3 and α-OH-4 on GlcNAc. To further explore the interaction between GlcNAc and ChCl-Gly, ^{1}H NMR chemical shift titration experiments were performed with different molar ratios of GlcNAc to ChCl-Gly (shown in Figure 6B and Figure S4). With the increase of GlcNAc, the hydroxyl proton signals of ChCl-OH and Gly-OH obviously shifted to upfield (ChCl-OH from 5.51 ppm shift to 5.43 ppm; Gly-OH-1 from 4.54 ppm shift to 4.52 ppm; Gly-OH-2 from 4.47 ppm shift to 4.45 ppm). In addition, the interaction between glycerol and GlcNAc was further demonstrated by DOSY NMR experiments, which is a pesedu-2D NMR spectroscopic technique with chemical shifts on the F1 dimension and diffusion coefficients (D) on the F2 dimension (shown in Figure S5).[44] The diffusion coefficients of glycerol and GlcNAc were $2.92 \cdot 10^{-10}$ m^2/s and $1.85 \cdot 10^{-10}$ m^2/s, respectively. The diffusion coefficients for glycerol and GlcNAc decreased to $1.54 \cdot 10^{-10}$ m^2/s and $1.49 \cdot 10^{-10}$ m^2/s when mixed. The DOSY NMR results directly proved that the alcoholic hydroxyl group devote a catalytic effect on the conversion of GlcNAc. Using $B(OH)_3$ as the HBD of DES can significantly change the selectivity of the reaction, because $B(OH)_3$ not only provides acidity but more importantly can be complexed with GlcNAc to promote further dehydration.[5,28,45–47]

In addition, the changes of Cl^- during the formation of ChCl-Gly and their interactions with GlcNAc were investigated by ^{35}Cl NMR experiments. As shown in Figure 7, the peak width at half height of the ^{35}Cl NMR signals gradually increased from 277.93 Hz to 471.18 Hz for ChCl, ChCl-Gly, while peak width at half height of KCl in the inner-coaxial tube as the standard, keep constant of 18.46 Hz. This is because of the formation of DESs makes the electric field gradient larger, which leads to the broadening of the peak of ^{35}Cl. In addition, the introduction of GlcNAc increases the half-width of the chlorine signal of ChCl-Gly from 471.18 Hz to 807.22 Hz. The broadening of this peak is due to the confinement of free Cl^- ions and the decrease of its mobility, which is in line with an interaction between Cl^- ions and GlcNAc.

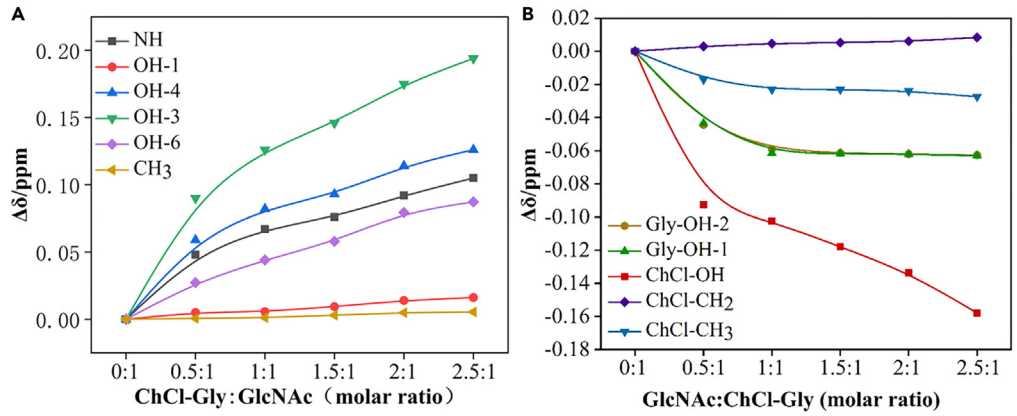


Figure 6. Trend of the chemical shift drifts of protons in ^{1}H NMR with the different molar ratios

(A) ChCl-Gly to GlcNAc and (B) GlcNAc to ChCl-Gly. Conditions: DMSO-d_6, 25 °C.

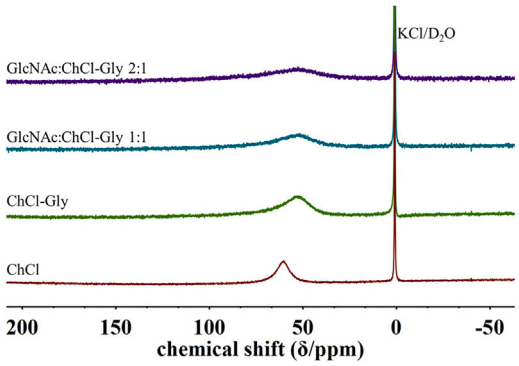


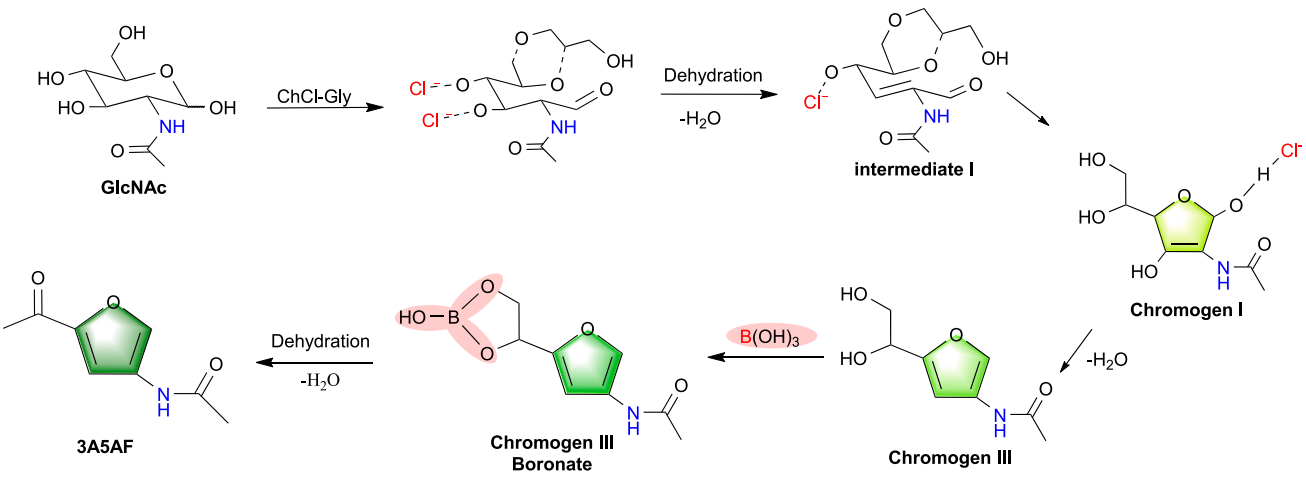
Figure 7. The ^{35}Cl NMR spectra of ChCl, ChCl-Gly, GlcNAc: ChCl-Gly (1:1) and GlcNAc: ChCl-Gly (2:1) in DMSO-d_6 at 25°C

Mass spectrometry analysis

In order to further determine the reaction intermediates and products, the reaction mixture from the reaction promoted by ChCl-Gly-B(OH)$_3$ was analyzed by negative-ion ESI mass spectrum. As shown in Figure S7, the composition of *m/z* 166 ion corresponds to the tri-dehydrated GlcNAc [M (GlcNAc)-3H$_2$O-H]$^-$, corresponds to 3A5AF. The signals at *m/z* 202.1 [M (GlcNAc)-H$_2$O-H]$^-$ and *m/z* 184.1 [M (GlcNAc)-2H$_2$O-H]$^-$ are Chromogen I and Chromogen III obtained by GlcNAc dehydrating one or two molecules of H$_2$O respectively. Besides, a signal at *m/z* 220, which is corresponding to the deprotonated molecular ion of GlcNAc [M (GlcNAc)-H]$^-$ ($C_8H_{14}O_6N^-$), was also observed because of the easily ionization of the polyol structure of GlcNAc. The intermediates and products assigned based on NMR could hence be confirmed by mass spectrometry.

Reaction pathway elucidation

It is commonly accepted that the dehydration reaction of GlcNAc first proceeds through a ring-opening to form a linear isomer, but the subsequent key steps are still under debate (shown in Figure S6). Based on the ^{1}H NMR chemical shift titration, ^{35}Cl and DOSY NMR results, we suggest a reaction mechanism of the transformation of GlcNAc under the promotion of DESs (shown in Scheme 2). Chloride interacts with the α-OH-3 and α-OH-4, and together with glycerol promote the removal of the first molecule of water from GlcNAc to form intermediate I. Intermediate I then cyclize, under the action of chloride ions, to form Chromogen I, and additional water molecule is removed forming Chromogen III. B(OH)$_3$ plays a key role in promoting the further dehydration reaction of Chromogen III to generate 3A5AF, achieving selective regulation of the reaction. Chromogen III and B(OH)$_3$ form an unstable intermediate, Chromogen III-Boronate, and finally 3A5AF was obtained by the third dehydration step. Hence, the stepwise dehydration of GlcNAc was achieved by the combined action of Cl$^-$, glycerol and B(OH)$_3$.



Scheme 2. Proposed reaction pathways for the formation of 3A5AF from GlcNAc dehydration

Conclusions

Choline chloride-based DESs were applied to promote the dehydration of GlcNAc to nitrogen-containing compounds, in high yields and selectivity under mild reaction conditions. The conversion of GlcNAc to Chromogen III or 3A5AF was achieved by altering the HBD part of DESs. ChCl-Gly promoted the dehydration of GlcNAc to produce Chromogen III, and the maximum yield was 31.1%. The maximum yield of 3A5AF was 39.2% using the ternary DES ChCl-Gly-$B(OH)_3$. The reaction intermediates were detected by *in situ* NMR experiments. ^{1}H NMR chemical shift titration experiments showed that ChCl-Gly interacted with OH-3 and OH-4 of GlcNAc. DOSY and ^{35}Cl NMR experiments confirmed the complexation between $B(OH)_3$ and GlcNAc and the interaction between Cl^- and GlcNAc. This work supports the theoretical study of the conversion of chitin monomer GlcNAc dehydration promoted by DESs. It also provides a new scheme for efficient and green synthesis of nitrogenous chemicals.

Limitations of the study

In this work, we designed and prepared several DESs to promote the conversion of GlcNAc to selectivity produce nitrogen-containing fine chemicals. This confirmed that DESs could promote the conversion of chitin biomass and achieve the regulation of the reaction. However, this is only the study of chitin monomer conversion in the laboratory, rather than the direct use of marine biomass resources such as shrimp, crab shells, etc. In addition, the understanding and study of the reaction mechanism still need to be further deepened. However, these works need to be improved and are being investigated by our group.

STAR★METHODS

Detailed methods are provided in the online version of this paper and include the following:

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  - Materials availability
  - Data and code availability
- METHOD DETAILS
  - General procedure for the DESs preparation
  - General reaction procedure
  - Recycling of the DESs
  - Characterization

SUPPLEMENTAL INFORMATION

Supplemental information can be found online at https://doi.org/10.1016/j.isci.2023.106980.

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AUTHOR CONTRIBUTIONS

J.Z. and H.C. performed the experiments, and wrote the article. C.M.P. was responsible for the revision and language polishing of the article. X.H. designed the whole experiments. Y.W. and Y.Q. conducted data analysis and provided financial support. All authors discussed the results and commented on the article.

DECLARATION OF INTERESTS

The authors declare no competing interests.

INCLUSION AND DIVERSITY

We support inclusive, diverse, and equitable conduct of research.

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REFERENCES

1. Maschmeyer, T., Luque, R., and Selva, M. (2020). Upgrading of marine (fish and crustaceans) biowaste for high added-value molecules and bio(nano)-materials. Chem. Soc. Rev. *49*, 4527–4563. https://doi.org/10.1039/c9cs00653b.

2. Wang, Y., Furukawa, S., Song, S., He, Q., Asakura, H., and Yan, N. (2020). Catalytic production of alanine from waste glycerol. Angew. Chem., Int. Ed. Engl. *59*, 2289–2293. https://doi.org/10.1002/anie.201912580.

3. Wang, Y., and Meyer, T.J. (2019). A route to renewable energy triggered by the haber-bosch process. Chem *5*, 496–497. https://doi.org/10.1016/j.chempr.2019.02.021.

4. Wang, M., Khan, M.A., Mohsin, I., Wicks, J., Ip, A.H., Sumon, K.Z., Dinh, C.-T., Sargent, E.H., Gates, I.D., and Kibria, M.G. (2021). Can sustainable ammonia synthesis pathways compete with fossil-fuel based Haber–Bosch processes? Energy Environ. Sci. *14*, 2535–2548. https://doi.org/10.1039/d0ee03808c.

5. Chen, X., Chew, S.L., Kerton, F.M., and Yan, N. (2014). Direct conversion of chitin into a N-containing furan derivative. Green Chem. *16*, 2204–2212. https://doi.org/10.1039/c3gc42436g.

6. Dai, J., Li, F., and Fu, X. (2020). Towards shell biorefinery: advances in chemical-catalytic conversion of chitin biomass to organonitrogen chemicals. ChemSusChem *13*, 6498–6508. https://doi.org/10.1002/cssc.202001955.

7. Chen, X., Liu, Y., Kerton, F.M., and Yan, N. (2015). Conversion of chitin and N-acetyl-d-glucosamine into a N-containing furan derivative in ionic liquids. RSC Adv. *5*, 20073–20080. https://doi.org/10.1039/c5ra00382b.

8. Chen, X., Song, S., Li, H., Gözaydın, G., and Yan, N. (2021). Expanding the boundary of biorefinery: organonitrogen chemicals from biomass. Acc. Chem. Res. *54*, 1711–1722. https://doi.org/10.1021/acs.accounts.0c00842.

9. Gao, X., Chen, X., Zhang, J., Guo, W., Jin, F., and Yan, N. (2016). Transformation of chitin and waste shrimp shells into acetic acid and pyrrole. ACS Sustain. Chem. Eng. *4*, 3912–3920. https://doi.org/10.1021/acssuschemeng.6b00767.

10. Zhao, Q., and Liu, L. (2021). One-step conversion of crab shells to levulinic acid catalyzed by ionic liquids: self-healing of chitin fraction. ACS Sustain. Chem. Eng. *9*, 1762–1771. https://doi.org/10.1021/acssuschemeng.0c07745.

11. Hou, W., Zhao, Q., and Liu, L. (2020). Selective conversion of chitin to levulinic acid catalyzed by ionic liquids: distinctive effect of N-acetyl groups. Green Chem. *22*, 62–70. https://doi.org/10.1039/c9gc02669j.

12. Wu, J., Qi, M., Gözaydın, G., Yan, N., Gao, Y., and Chen, X. (2021). Selectivity-switchable conversion of chitin-derived N-Acetyl-D-glucosamine into commodity organic acids at room temperature. Ind. Eng. Chem. Res. *60*, 3239–3248. https://doi.org/10.1021/acs.iecr.0c05805.

13. Osada, M., Shoji, S., Suenaga, S., and Ogata, M. (2019). Conversion of N-acetyl-d-glucosamine to nitrogen-containing chemicals in high-temperature water. Fuel Process. Technol. *195*, 106154. https://doi.org/10.1016/j.fuproc.2019.106154.

14. Xu, B., Du, Z., Dai, J., Yang, R., Yang, D., Gu, X., Li, N., and Li, F. (2022). Progress in catalytic conversion of renewable chitin biomass to furan-derived platform compounds. Catalysts *12*, 653. https://doi.org/10.3390/catal12060653.

15. Santos, L.B., Assis, R.S., Barreto, J.A., Bezerra, M.A., Novaes, C.G., and Lemos, V.A. (2022). Deep eutectic solvents in liquid-phase microextraction: contribution to green chemistry. TrAC, Trends Anal. Chem. *146*, 116478. https://doi.org/10.1016/j.trac.2021.116478.

16. Cao, S., Liu, Y., Shi, L., Zhu, W., and Wang, H. (2022). N-Acetylglucosamine as a platform chemical produced from renewable resources: opportunity, challenge, and future prospects. Green Chem. *24*, 493–509. https://doi.org/10.1039/d1gc03725k.

17. Deng, W., Feng, Y., Fu, J., Guo, H., Guo, Y., Han, B., Jiang, Z., Kong, L., Li, C., Liu, H., et al. (2023). Catalytic conversion of lignocellulosic biomass into chemicals and fuels. Green Energy & Environment *8*, 10–114. https://doi.org/10.1016/j.gee.2022.07.003.

18. Stoykov, Y.M., Pavlov, A.I., and Krastanov, A.I. (2015). Chitinase biotechnology: production, purification, and application. Eng. Life Sci. *15*, 30–38. https://doi.org/10.1002/elsc.201400173.

19. Bobbink, F.D., Zhang, J., Pierson, Y., Chen, X., and Yan, N. (2015). Conversion of chitin derived N-acetyl-d-glucosamine (NAG) into polyols over transition metal catalysts and hydrogen in water. Green Chem. *17*, 1024–1031. https://doi.org/10.1039/c4gc01631a.

20. Chen, B., Peng, Z., Li, C., Feng, Y., Sun, Y., Tang, X., Zeng, X., and Lin, L. (2021). Catalytic conversion of biomass to furanic derivatives with deep eutectic solvents. ChemSusChem *14*, 1496–1506. https://doi.org/10.1002/cssc.202100001.

21. Hao, Y.-C., Zong, M.-H., Wang, Z.-L., and Li, N. (2021). Chemoenzymatic access to enantiopure N-containing furfuryl alcohol from chitin-derived N-acetyl-D-glucosamine. Bioresour. Bioprocess. *8*, 80. https://doi.org/10.1186/s40643-021-00435-w.

22. Pham, T.T., Lindsay, A.C., Chen, X., Gözaydin, G., Yan, N., and Sperry, J. (2019). Transferring the biorenewable nitrogen present in chitin to several N-functional groups. Sustainable Chemistry and Pharmacy *13*, 100143. https://doi.org/10.1016/j.scp.2019.100143.

23. Pham, T.T., Gözaydın, G., Söhnel, T., Yan, N., and Sperry, J. (2019). Oxidative ring-expansion of a chitin-derived platform enables access to unexplored 2-amino sugar chemical space. Eur. J. Org Chem. *2019*, 1355–1360. https://doi.org/10.1002/ejoc.201801683.

24. Liu, Y., Stähler, C., Murphy, J.N., Furlong, B.J., and Kerton, F.M. (2017). formation of a renewable amine and an alcohol via transformations of 3-Acetamido-5-acetylfuran. ACS Sustain. Chem. Eng. *5*, 4916–4922. https://doi.org/10.1021/acssuschemeng.7b00323.

25. Sadiq, A.D., Chen, X., Yan, N., and Sperry, J. (2018). Towards the shell biorefinery: sustainable synthesis of the anticancer alkaloid proximicin A from chitin. ChemSusChem *11*, 532–535. https://doi.org/10.1002/cssc.201702356.

26. Zang, H., Feng, Y., Lou, J., Wang, K., Wu, C., Liu, Z., and Zhu, X. (2022). Synthesis and performance of piperidinium-based ionic liquids as catalyst for biomass conversion into 3-acetamido-5-acetylfuran. J. Mol. Liq. *366*, 120281. https://doi.org/10.1016/j.molliq.2022.120281.

27. Zang, H., Lou, J., Jiao, S., Li, H., Du, Y., and Wang, J. (2021). Valorization of chitin derived N-acetyl-D-glucosamine into high valuable N-containing 3-acetamido-5-acetylfuran using pyridinium-based ionic liquids. J. Mol. Liq. *330*, 115667. https://doi.org/10.1016/j.molliq.2021.115667.

28. Zang, H., Li, H., Jiao, S., Lou, J., Du, Y., and Huang, N. (2021). Green conversion of N-acetylglucosamine into valuable platform compound 3-Acetamido-5-acetylfuran using ethanolamine ionic liquids as recyclable catalyst. ChemistrySelect *6*, 3848–3857. https://doi.org/10.1002/slct.202100231.

29. Du, Y., Zang, H., Feng, Y., Wang, K., Lv, Y., and Liu, Z. (2022). Efficient catalytic system for converting N-acetyl-d-glucosamine into valuable chemical 3-acetylamino-5-acetylfuran. J. Mol. Liq. *347*, 117970. https://doi.org/10.1016/j.molliq.2021.117970.

30. Wang, J., Zang, H., Jiao, S., Wang, K., Shang, Z., Li, H., and Lou, J. (2020). Efficient conversion of N-acetyl-D-glucosamine into

nitrogen-containing compound 3-acetamido-5-acetylfuran using amino acid ionic liquid as the recyclable catalyst. Sci. Total Environ. *710*, 136293. https://doi.org/10.1016/j.scitotenv.2019.136293.

31. Omari, K.W., Dodot, L., and Kerton, F.M. (2012). A simple one-pot dehydration process to convert N-acetyl-D-glucosamine into a nitrogen-containing compound, 3-acetamido-5-acetylfuran. ChemSusChem *5*, 1767–1772. https://doi.org/10.1002/cssc.201200113.

32. Drover, M.W., Omari, K.W., Murphy, J.N., and Kerton, F.M. (2012). Formation of a renewable amide, 3-acetamido-5-acetylfuran, via direct conversion of N-acetyl-d-glucosamine. RSC Adv. *2*, 4642. https://doi.org/10.1039/c2ra20578e.

33. Osada, M., Kikuta, K., Yoshida, K., Totani, K., Ogata, M., and Usui, T. (2013). Non-catalytic synthesis of Chromogen I and III from N-acetyl-d-glucosamine in high-temperature water. Green Chem. *15*, 2960. https://doi.org/10.1039/c3gc41161c.

34. Chen, K., Wu, C., Wang, C., Zhang, A., Cao, F., and Ouyang, P. (2021). Chemo-enzymatic protocol converts chitin into a nitrogen-containing furan derivative, 3-acetamido-5-acetylfuran. Mol. Catal. *516*, 112001. https://doi.org/10.1016/j.mcat.2021.112001.

35. Qin, H., Hu, X., Wang, J., Cheng, H., Chen, L., and Qi, Z. (2020). Overview of acidic deep eutectic solvents on synthesis, properties and applications. Green Energy & Environment *5*, 8–21. https://doi.org/10.1016/j.gee.2019.03.002.

36. Chen, Y., and Mu, T. (2019). Application of deep eutectic solvents in biomass pretreatment and conversion. Green Energy & Environment *4*, 95–115. https://doi.org/10.1016/j.gee.2019.01.012.

37. Wang, J., Wang, Y., Ma, Z., and Yan, L. (2020). Dissolution of highly molecular weight cellulose isolated from wheat straw in deep eutectic solvent of Choline/l-Lysine hydrochloride. Green Energy & Environment *5*, 232–239. https://doi.org/10.1016/j.gee.2020.03.010.

38. Sert, M., Arslanoğlu, A., and Ballice, L. (2018). Conversion of sunflower stalk based cellulose to the valuable products using choline chloride based deep eutectic solvents. Renew. Energy *118*, 993–1000. https://doi.org/10.1016/j.renene.2017.10.083.

39. Filonenko, S., Voelkel, A., and Antonietti, M. (2019). Valorization of monosaccharides towards fructopyrazines in a new sustainable and efficient eutectic medium. Green Chem. *21*, 5256–5266. https://doi.org/10.1039/c9gc02176k.

40. Liu, P., Pedersen, C.M., Zhang, J., Liu, R., Zhang, Z., Hou, X., and Wang, Y. (2021). Ternary deep eutectic solvents catalyzed d-glucosamine self-condensation to deoxyfructosazine: NMR study. Green Energy & Environment *6*, 261–270. https://doi.org/10.1016/j.gee.2020.04.010.

41. Guo, Z., Chen, C., Zhao, J., Guo, X., Jia, L., Liu, P., Marcus Pedersen, C., Hou, X., Qiao, Y., and Wang, Y. (2022). Mechanism of the dehydration of N-acetyl-d-glucosamine into N-containing platform molecule 3-acetamido-5-acetylfuran: NMR study. J. Mol. Liq. *365*, 120219. https://doi.org/10.1016/j.molliq.2022.120219.

42. Jia, L., Zhang, Z., Qiao, Y., Pedersen, C.M., Ge, H., Wei, Z., Deng, T., Ren, J., Liu, X., Wang, Y., and Hou, X. (2017). Product distribution control for glucosamine condensation: nuclear magnetic resonance (NMR) investigation substantiated by density functional calculations. Ind. Eng. Chem. Res. *56*, 2925–2934. https://doi.org/10.1021/acs.iecr.6b05057.

43. Zhao, J., Liu, R., Zhang, Z., Xing, Q., Chang, H., Hou, X., and Wang, Y. (2022). Tautomer distributions of N-acetyl-D-glucosamine in the condition of commonly utilized solvents and catalysts for biorefinery: NMR study. J. Mol. Struct. *1251*, 131995. https://doi.org/10.1016/j.molstruc.2021.131995.

44. Yan, K., Bai, Z., and Huang, S. (2021). NMR signal separation of ionic liquids by poly(sodium-p-styrenesulfonate)-assisted chromatographic NMR spectroscopy. Magnetic Resonance Letters *1*, 153–159. https://doi.org/10.1016/j.mrl.2021.100007.

45. Ci, Y.-h., Yu, F., Zhou, C.-x., Mo, H.-e., Li, Z.-y., Ma, Y.-q., and Zang, L.-h. (2020). New ternary deep eutectic solvents for effective wheat straw deconstruction into its high-value utilization under near-neutral conditions. Green Chem. *22*, 8713–8720. https://doi.org/10.1039/d0gc03240a.

46. Jiang, W., Zhu, K., Jia, H., Zhu, L., Wang, C., Xu, L., Li, H., Zhu, W., and Li, H. (2022). Synthesis of task-specific ternary deep eutectic solvents for deep desulfurization via reactive extraction. Chemical Engineering and Processing - Process Intensification *171*, 108754. https://doi.org/10.1016/j.cep.2021.108754.

47. Zhang, H., Liu, X., Han, M., and Zhang, R. (2022). Conversion of bio-carbohydrates to 5-hydroxymethylfurfural in three-component deep eutectic solvent. RSC Adv. *12*, 14957–14963. https://doi.org/10.1039/d2ra01688e.

48. Wu, C., Wang, C., Zhang, A., Chen, K., Cao, F., and Ouyang, P. (2022). Preparation of 3-aceta mido-5-acetylfuran from N-acetylglucosamine and chitin using biobased deep eutectic solvents as catalysts. React. Chem. Eng. *7*, 1742–1749. https://doi.org/10.1039/d2re00118g.

STAR★METHODS

KEY RESOURCES TABLE

REAGENT or RESOURCE	SOURCE	IDENTIFIER
Chemicals, peptides, and recombinant proteins		
N-acetyl-D-glucosamine	Aladdin (China)	Cat#7512-17-6
Ethylene glycol	Aladdin (China)	Cat#107-21-1
1,3-Butanediol	Aladdin (China)	Cat#107-88-0
DMSO-d_6	QingdaoTengl (China)	N/A
boric acid	Sinopharm Chemical Reagent (China)	Cat#10043-35-3
pyrazine	Sinopharm Chemical Reagent (China)	Cat#290-37-9
glycerol	Sinopharm Chemical Reagent (China)	Cat#56-81-5
N-methyl-2-pyrrolidone	Macklin Chemical Reagent (China)	Cat#872-50-4
N, N-dimethylacetamide	Macklin Chemical Reagent (China)	Cat#127-19-5
Dimethyl sulfoxide	Macklin Chemical Reagent (China)	Cat#67-68-5
Choline chloride	Aladdin (China)	Cat#67-48-1
N, N-dimethylformamide	Macklin Chemical Reagent (China)	Cat#68-12-2

RESOURCE AVAILABILITY

Lead contact

Further information and requests for resources should be directed to and will be fulfilled by the lead contact, Yingxiong Wang (wangyingxiong@tyut.edu.cn).

Materials availability

This study did not generate new unique reagents.

GlcNAc, choline chloride (ChCl), ethylene glycol and 1,3-butanediol were purchased from Aladdin Co., Ltd. Dimethyl sulfoxide-d6 (DMSO-d6, 99.9% atom D) was provided by Qingdao Teng Long Microwave Technology Co. Ltd. B(OH)3, pyrazine and glycerine (Gly) were purchased from Sinopharm Chemical Reagent Co., Ltd. N-methyl-2-pyrrolidone (NMP), N, N-dimethylacetamide (DMA), dimethyl sulfoxide (DMSO) and N, N-dimethylformamide (DMF) were purchased from Macklin Chemical Reagent Co., Ltd. All the chemicals were used without further purification.

Data and code availability

- This study does not generate new unique reagent.
- This paper does not report original code.
- Any additional information required to reanalyze the data reported in this paper can be obtained from the lead contact upon request.

METHOD DETAILS

General procedure for the DESs preparation

To prepare the DESs, HBA ChCl and HBDs (ethylene glycol, 1,3-butanediol, glycerin, $B(OH)_3$) with a molar ratio of 1:1 was added in a round bottom flask and heated to 80°C in an oil bath under stirring until a homogeneous liquid was formed. Ternary DESs, were prepared by choline chloride, $B(OH)_3$ and glycerol in the ratio of 1:1:0.5 according to the above steps. The prepared DESs were dried in a vacuum oven at 80°C for 12 h. These DESs are abbreviated as ChCl-ethylene glycol, ChCl-1,3-butanediol, ChCl-Gly, ChCl-$B(OH)_3$, ChCl-Gly-$B(OH)_3$.

General reaction procedure

The preparation of nitrogenous chemicals by the dehydration reaction of GlcNAc was carried out in a round bottom flask heated in an oil bath. In a typical procedure, 1 mmol GlcNAc and 2 mmol DESs were added to 5 mL solvent. Then the mixture was heated and stirred using a magnetic stirrer at a defined temperature. After a certain reaction time, 0.1 mL of reaction mixture was taken out and immediately cooled in an ice bath to quench the reaction. This sample was used for analysis.

Recycling of the DESs

We refer to the method of DES treatment and recovery in the literature to conduct a recycle experiment, and the specific process is as follows[48]: an equal amount of water was added to the reaction mixture and extracted three times with ethyl acetate. The water in the mixture was completely removed on a rotary evaporator at 100°C, and then add the reaction solvent for the cyclic reaction.

Characterization

The one-dimensional (1D) and two-dimensional (2D) NMR spectra were obtained on a Bruker AV-III 400 MHz NMR spectrometer (9.39 T). The NMR parameters for ^{1}H measurements were: pulse program for acquisition, zg30; AQ, 4.09 s; D1, 1.0 s; DS, 2; NS, 16. The product yields were evaluated by quantitative ^{1}H NMR spectra (^{1}H qNMR) (pulse sequence: zg, relaxation delay: 15 s, number of scans: 16). Pyrazine (0.3 mg mL^{-1}) was prepared in DMSO-d_6 and used as a standard solution for the ^{1}H qNMR measurement. The ^{1}H qNMR sample was prepared by mixing 0.10 mL of reaction mixture and 0.4 mL of standard solution. Based on the ^{1}H qNMR spectra, the yields of products were calculated as:

$$\text{yield} = \frac{\text{moles of product}}{\text{moles of GlcNAc}} * 100\%$$

The ^{35}Cl NMR spectra were obtained at frequency of 39.20 MHz, with the following pulse program: zg; acquisition time, 0.42 s; relaxation delay time, 0.1 s; scans, 2048. DOSY experiments were carried out with the Bruker standard bipolar pulse longitudinal eddy current delay (BPPLED) pulse sequence at 25°C and at a gas flow rate of 400 lph without sample spinning. In order to acquire 2–5% residual signal with the maximum gradient strength, the duration of the pulse field gradient (δ/2) was adjusted between 600 and 2000 ms. The eddy current delay was 5 ms and the delay for gradient recovery was 0.2 ms. The gradient strength was incremented in 16 steps from 2% to 95% of its maximum value in a linear ramp. The data were processed by Bruker Topspin 3.1 and Dynamics Center 2.2.4 software. The reaction mixtures were further qualitatively identified by the negative-ion ESI mass spectrum on a Waters Xevo G2-XS QTOF high resolution mass spectrometer.