Imidazole Promoted Efficient Anomerization of β-D-Glucose Pentaacetate in Solid State and Reaction Mechanism

Meifeng Wang[♀a,b], *Liyin Zhang*[♀a], *Yiqun Li*[b]* *and Liuqun Gu*[a]*

[a] Department of Biomedical Engineering, Jinan University; [#]601, Huangpu Avenue West, Guangzhou, China

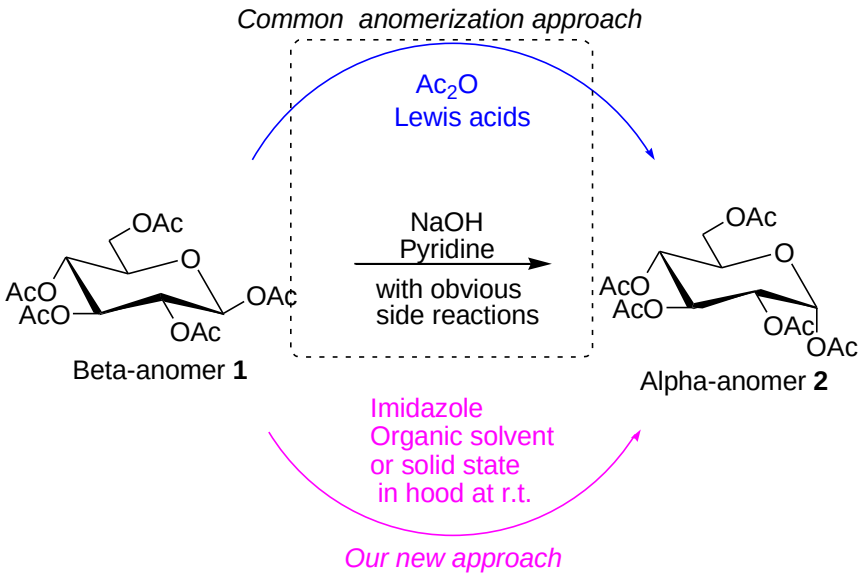
[b] Department of Chemistry, Jinan University; [#]601, Huangpu Avenue West, Guangzhou, China

KEYWORDS. Imidazole; Anomerization; Basic condition; hydrogen bonding network; solid state.

ABSTRACT. Anomerization of glycosides was rarely performed under basic condition. Here an imidazole promoted anomerization of β-D-glucose pentaacetate in solid state at room temperature was discovered. This unprecedent anomerization in solid state occurred after simple mixing and reaction proceeded continuously to full conversion without stirring or mechanomixing. Current understanding of reaction mechanism involved with inter/intramolecular acyl transfer promoted by two imidazole in concerted manner may promote discovery of more new transformations of glucosides in solid state.

Carbohydrates play important role in various biological processes relating to virology, immunology, cancer and hence sugar-based molecules attracted increasing attention of medicinal chemists.[1] Different conformational preferences can influent biological properties remarkably.[2] Even after full acetylation of all hydroxyl groups, anomeric effect still persistent. D-Glucose pentaacetate is an important intermediate for synthesis of different types of glycosides.[1,3] During glycosylation, although β-D-glucose pentaacetate was found to react faster with nucleophiles in the presence of Lewis acids,[4] its α anomer showed better performance in more applications like CO_2 absorption[5] and stimulation of insulin release[6]. α-D-Glucose pentaacetate was usually prepared from anomerization of β-anomer with acetyl anhydride catalyzed by Lewis acids.[7] Treating β-D-glucose pentaacetate with Lewis acids for α form is a classic anomerization method, based on a fact that a good stability of α-anomer towards a variety of acidic conditions which readily dissociate the β form,[8,9] including a recent $TiCl_4$ or $SnCl_4$ promoted anomerization of O-glycosides or S-glycosides at relative low concentration of glycoside substrates from P. V. Murphy group.[8]

Scheme 1. Our anomerization of β-D-glucose pentaacetate under basic condition in comparison of reported approaches.



34

35 However, only less than a hand of anomerizations of D-glucose pentaacetate conditions is
36 reported under basic reaction probably due to lack of efficiency. It was M. L. Wolfrom and D. R.
37 Husted who reported the first case, in which a good conversion of β-form sugar to α-form was
38 observed in dioxane or diethyl ether when mixing with solid sodium hydroxide and a suitable
39 drying agent.[10] In 1950, the following study by Lindberg indicated anomerization in pyridine were
40 6 – 7 times faster than that in dioxane or diethyl ether although strong side reactions were still
41 observed; a heterogeneous catalysis mechanism was also proposed[11]. Treatment of β-2,4-
42 dinitrophenyl 2,3,4,6-tetra-0-acetyl-D-glucopyranoside with potassium carbonate in DMF could
43 also lead to an excellent anomerization to the α-form.[12] Epimerization of β-D-glucose pentaacetate
44 to the α-form in dilute deuterochloroform solution was also observed by J. H. Goldstein and co-
45 authors, however experimental detail was missing in the literature.[13] Here we report an imidazole
46 promoted efficient anomerization in solid state at room temperature (**Scheme 1**); to our best
47 knowledge anomerization reaction of D-glucose pentaacetate or D-glucose in solid state under
48 basic condition was not yet reported in literature although very recently an interesting chemically
49 reversible isomerization of inorganic clusters was discovered by a team of researchers[14]. It should

be noted that this reaction in solid state is different from current popular mechanochemistry because no mechanic mixing (grindling) was used during reaction; It belongs to a new type of "mixing and stand" solid state reaction.[15]

Table 1. Optimization of loading of imidazole and reaction time.[a]

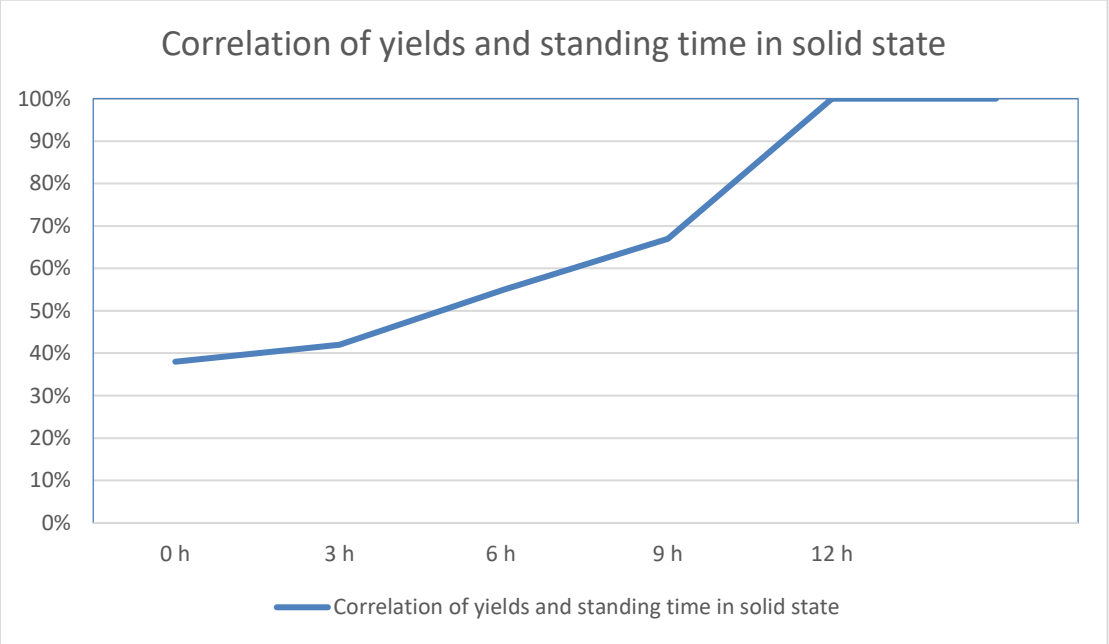
Entry	Imidazole (equiv.)[b]	Time (h)	Yield (%)[c]
1	0.5	1	22
2	1	1	40
3	2	1	92

a: Reaction conditions: β-D-Glucose pentaacetate (1 mmol), activated 4Å molecular sieves (0.2 g), and imidazole (0.5 mmol to 2 mmol) were added into a pre-dried vial (20 mL), then anhydrous dichloromethane (2 mL) was added. The vial was sealed and the mixture was stirred at room temperature for 1 h as shown in the table. Evaporation of solvent to give a crude mixture, which was kept open in hood overnight for ^{1}H NMR analysis. b: Equivalent of β-D-glucose pentaacetate. c: Determined by crude ^{1}H NMR.

We noticed than imidazole could promote anomerization of β-D-glucose pentaacetate in anhydrous dichloromethane during our new methodology exploration. Followed literature research indicated that such anomerization was ever observed in $CDCl_3$ during study of imidazole catalyzed acetyl transfer reactions in 1963 by Goldstein[13]. Because experimental part was not included in the literature and potential importance of this reaction, we revisited this rare anomerization process. After thorough study of potential affecting factors, we eventually found that the reaction was pretty sensitive to water and consistent yields (**STable 1** in *Supporting Information*.) could be achieved by carrying out reactions in the presence of activated 4Å molecular sieves.[16] With anhydrous dichloromethane as a solvent, effect of loading of imidazole was also evaluated at room temperature in the presence of activated 4Å molecular sieves. The yield became higher along with increased use of imidazole and over 93% yield could be obtained in an hour when ratio of β-D-glucose pentaacetate: imidazole = 1 : 2 (Entries 1 – 3, **Table 1**). Prolonged reaction time to 3 hours

had negligible effect on the yield (Entries 3 – 5, **Table 1**); and obvious side products were observed probably because sugar ring open reactions occurred due to more moisture inside.

Figure 1: Correlation of yields and standing time in solid state



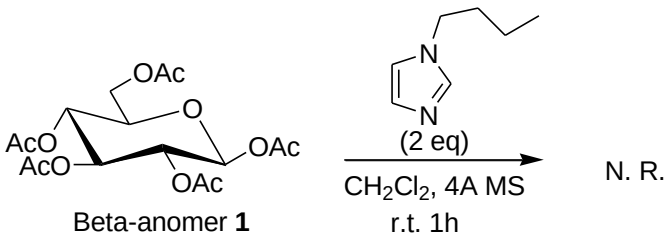
In our subsequent reproduced work when operating hands were changed, yields were however surprisingly inconsistent and sometime obvious differences were observed. Excluding possibility of existence of moisture inside of molecular sieves or anhydrous dichloromethane, the problem was eventually located at different standing times after evaporation of solvents when reactions were stopped; continued anomerization in solid state was hypothesized logically. Yields were obtained in 30% and > 99% at different standing time (0 h and 12 hrs) in solid state upon evaporation of all dichloromethane after the mixture was stirred for 1 hour; the sharp difference might well explain the previous inconsistence and encouraged us to further explore this unexpected anomerization reaction in solid state. Yields measured at 0 hr, 3 hrs, 6 hrs, 9 hrs, 12 hrs standing in solid state after 1 hr pre-mixing of β-D-glucose pentaacetate (1 mmol), imidazole (2 mmol) and

4Å molecular sieves upon evaporation of solvent. A good line correlation was observed between yield and standing time (**Figure 1**), which supported our hypothesis that anomerization indeed continues to proceed in solid state. With combination of mixing/anomerization in organic solution and continued anomerization in solid state, yields of α-form were highly reproduceable quantitively after 12 hrs standing time in hood, no matter dichloromethane was from any purification method. Under optimized conditions, solvent effect was intangible for aprotonic solvents like acetonitrile and hexane, while only moderate yield was achieved when using protonic solvent ethanol. Such observance showed that proton also inhibits anomerization in solid state, but reactions in solid state have a better moisture tolerance.

Table 2. **Optimization of anomerization in solid state**.[a]

Entry	Imidazole (Equiv.)	4Å Molecular sieves (g)	Standing time in solid state (h)	Yield (%)[b]
1	0	0	0	0
2	0	0	24	0
3	0	0	48	0
4	0	0	96	0
5	2	0	0	39
6	2	0	12	70
7	2	0	24	> 99
8	2	0.2	12	> 99
9	1	0.2	24	42
10	1	0.2	48	63
11	1	0.2	96	69

Scheme 2. Anomerization with 1-butyl imidazole instead of imidazole.

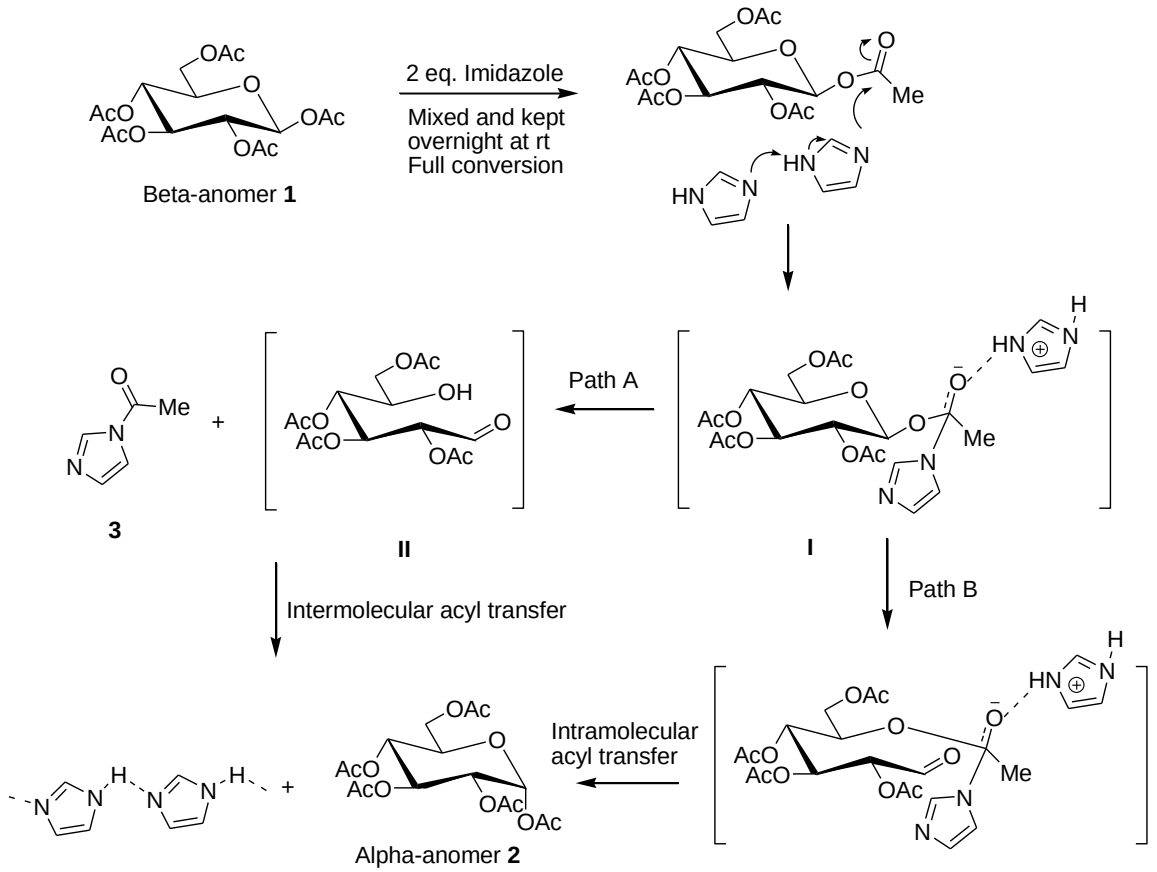


Beta-anomer **1**

Mechanistic study was also performed in order to gain more clues. Replacement of imidazole with 1-butyl imidazole led to no reaction and β-D-glucose pentaacetate was fully recovered, which indicated the necessary role of free nucleophilic amine part in imidazole (**Scheme 2**). Evidences that β-D-glucose pentaacetate kept without any change with imidazole hydrogen chloride as promoter, and moisture inside reaction mixture affected conversion yield significantly, indicated that the presence of proton inhibited anomerization. Removal of imidazole and 4Å molecular sieves prohibited anomerization and gave no α-form product at all even (β-D-glucose pentaacetate was fully recovered without any change) after 96 hrs standing in solid state (Entries 1 – 4, **Table 2**); hence it excluded a possibility of slow dissociation in solid state because of stability difference. In the absence of 4Å molecular sieves, anomerization reaction still could proceed as observed in organic solution albeit in a slower rate; almost pure α-form product could be obtained in 1 day (Entries 5 – 7, **Table 2**). Pores of 4Å molecular sieves might be benefit in absorbing of moisture and generation of more reactive amorphous state. In the presence of stoichiometric amount of imidazole, only 69% conversion was observed even after 96 hrs standing time (Entries 8 – 11, **Table 2**) and obvious impurity formed, which indicated excess amount of imidazole was pretty necessary for a clean and outstanding yield.

Scheme 3. Proposed bisimidazole promoted anomerization mechanism.

Bisimidazole promoted anomerization pathways under basic condition.



Reconstruction of hydrogen bonding network of imdazole in solid or solution phase is potential driving force.

115

116 Goldstein proposed an intermolecular acyl transfer catalyzed by an imidazole and a subsequent
117 sugar ring-open reaction making dissociation of two anomers possible.[13] A key evidence is
118 detection of N-acetylimidazole **3** in dilute deuterochloroform solution by NMR[13]; and a further
119 acetyl transfer to aliphatic alcohol was also observed, similar with enzyme catalyzed acyl transfer
120 reaction under biological conditions[17]. A following more comprehensive mechanistic study on
121 carbonate catalyzed anomerization of β-2,4-dinitrophenyl 2,3,4,6-tetra-0-acetyl-D-
122 glucopyranoside in DMF/DMSO from Withers' group also suggested anion form of intermediate
123 II (**Scheme 3**) could be an active intermediate in the anomerization;[18] and he also suggested an

acyl pyridinium specie like N-acetylimidazole **3** likely play a key role in pyridine-catalyzed anomerization[19].

However, there are still some facts in our system that could not be well explained by Goldstein's mechanism: 1) In our observance, 2 equivalent of imidazole was quite necessary for a completed anomerization in either dichloromethane or in solid state. It means possible that more than an imidazole was involved in promotion of anomerization. 2) We observed a full and clean anomerization in optimized condition. Meanwhile quantitively transferring acetyl group from N-acetylimidazole **3** in dilute solution or under heterogeneous system is very challenging and uncommon according to basic physical organic chemistry knowledge. 3) The anomerization in solid state proceeded without mechanomixing or heating, which is not explainable with reported mechanism and we are curious what is the driving force.

To better explain key features observed on anomerization in solid state, a different bisimidazole promoted mechanism involved two possible pathways via intermolecular or intramolecular acyl transfer is proposed here (**Scheme 3**). Bisimidazole mechanism involved concerted deprotonation step was proposed before by several groups to better explain the necessary of 2 equivalent imidazole in promotion of N-formylation via formyl transfer[20] and peptide cyclization via ester transfer[21]. We envisage that two molecules of imidazole also play concerted roles in activation of β-D-glucose pentaacetate, one acts as nucleophilic reagent and another one stabilizes oxygen atom of carbonyl group of C_1 acetyl moiety as descripted in intermediate I (**Scheme 3**). A subsequent release of N-acetylimidazole **3** lead to generation of aldehyde intermediate II, isomerization of intermediate II and a following ring closed esterification plus an intermolecular acetyl transfer back to C_1 moiety of sugar complete the anomerization forming α-form **2**. This pathway A is similar to reported mechanism proposed by Goldstein and Withers, and the proposed bisimidazole

promotion is more energetic feasible[21]. Another possible pathway (Path B) from intermediate I is a intramolecular acyl migration along with imidazole molecules from oxygen anion on C_1 to that on C_5 (after ring open reaction), forming intermediate III (**Scheme 3**). Although no experimental evidence is available yet, it is theoretically very possible and might be even kinetic favorable than path A because intramolecular functional group transfer is usually more feasible than intermolecular transfer. In fact, acetyl migrations within monosaccharides[22,23] and oligosaccharides[23] were already observed under mild basic conditions. Such double intramolecular acetyl transfer away and back to C_1 moiety could far more easily lead to a full and clean anomerization forming α-form **2**. Reconstruction of imidazole hydrogen bonding network could be a major driving force to transform intermediate II or intermediate III to final product **2**; imidazole hydrogen bonding network was known to be the major form in solution[24,25] or solid state[26] long time ago. Very recently, NMR evidence of downfield shift for imidazole N-proton[21] and observance of a peculiar behaviour of imidazole during scanning tunnelling microscopy-break junction (STM-BJ) experiments[27] also indicated the existence of in-situ generated hydrogen bonding network of imidazole. Both reaction pathways are competitive in solution or in solid state although path B is more favorable kinetically.

Above bisimidazole promoted mechanism (**Scheme 3**) could also explain the extraordinary reaction performance in solid state: 1) all active species are very less mobile in solid state, which benefit path A; 2) moisture has much lower chance to interact with intermediates I, II, III and N-acetylimidazole **3**, minimizing hydrolysis side reactions. This is could be a possible reason why reaction in solid state has a better tolerance to moisture observed in our experiments.

It should be noted that this unprecedent anomerization in solid state promoted by imidazole is very different with a reported $NaHCO_3$-catalyzed solid state anomerization of D-glucose on

mechanism, the latter one needed a mechanomixing during reactions and reactions proceeded via a protonic activating route because a stronger base Na_2CO_3 was inactive at all under the same conditions[28].

In conclusion, an imidazole promoted anomerization of β-D-glucose pentaacetate was developed; Even a relatively low yield was achieved after pre-mixing in organic solution, continued reaction in solid state (removal of organic solvent) by simple keeping the crude mixture stand for 24 hours could still led to a consistent full conversion. This is one of rare cases for anomerization of glycosides under basic conditions, and the first one in solid state (under basic condition) to our best of knowledge. A bisimidazole promoted mechanism involved both intermolecular acyl transfer and intramolecular acyl transfer was more probably based on new evidences and reported literatures. Although current study was limited to β-D-glucose pentaacetate, this unprecedent mild anomerization in solid state driven by reconstruction of imidazole hydrogen network may inspire more stereoseletive transformations of glucosides in solid state in the future.

ASSOCIATED CONTENT

Supporting Information.

The following files are available free of charge.

Additional Information

No conflict of interest was declared here.

AUTHOR INFORMATION

Corresponding Author

*(L. Gu) E-mail: guliuqun@yahoo.com;

* (Y. Li). E-mail: tlyq@jnu.edu.cn

Author Contributions

L. Gu conceived concept and designed experiments;L. Zhang and M. Wang carried out all experiments, collected data and prepared supporting information. L. Gu and Y. Li co-supervised this project; L. Gu proposed reaction mechanism and drafted this manuscript; both L. Gu and Y. Li edited it. All authors have given approval to the final version of the manuscript. ♀These authors contributed equally.

ACKNOWLEDGMENT

We acknowledge a startup funding from Jinan University to L. Gu (No: 88015155 and 88016607).

REFERENCES

(1) Hanessian, S.; Lou, B. Stereocontrolled Glycosyl Transfer Reactions with Unprotected Glycosyl Donors. *Chem. Rev.* **2000**, *100* (12), 4443-4463. b) Das, R.; and Mukhopadhyay, B. Chemical O-Glycosylations: An Overview. *ChemistryOpen* **2016**, *5* (5), 401-433.

(2) Montero, E.; García-Herrero, A.; Asensio, J. L.; Hirai, K.; Ogawa, S.; Santoyo-Gonza´lez, F.; Canada, F. J.; Jime´nez-Barbero, J. The Conformational Behavior of Non-hydrolyzable Lactose Analogues: the Thioglycoside, Carbaglycoside, and Carba-iminoglycoside Cases. *Eur. J. Org. Chem.* **2000**, (10), 1945-1952.

(3) Lawandi, J.; Rocheleau, S.; Moitessier, N. Regioselective Acylation, Alkylation, Silylation and Glycosylation of Monosaccharides. *Tetrahedron* **2016**, *72* (41), 6283-6319.

(4) Jensen, K. J. O-Glycosylations under Neutral or Basic Conditions. *J. Chem. Soc., Perkin Trans. 1* **2002**, *20*, 2219-2233.

(5) Ma, S.-L.; Wu, Y.-T.; Hurrey, M. L.; Wallen, S. L.; and Grant, C. S. Sugar Acetates as CO2-philes: Molecular Interactions and Structure Aspects from Absorption Measurement Using Quartz Crystal Microbalance. *J. Phys. Chem. B* **2010**, *114* (11), 3809-3817.

(6) Malaisse, W. J.; Jijakli, H.; Kadiata, M. M.; Sener, A.; and Kirk, O. Stimulation of insulin release by α-D-glucose pentaacetate. *Biochem. Biophys. Res. Commun.* **1997**, *231* (2), 435-436.

(7) Huang, G.; Tang, Q.; Li, D.; Huang, Y. and Zhang, D. Synthetic Methods of α-D-Glucose Pentaacetate. *Current Organic Synthesis* **2016**, *13* (1), 82-85.

(8) Pilgrim, W.; and Murphy, P. V. $SnCl_4$- and $TiCl_4$-Catalyzed Anomerization of Acylated O- and S-Glycosides: Analysis of Factors That Lead to Higher α:β Anomer Ratios and Reaction Rates. *J. Org. Chem.* **2010**, *75* (20), 6747-6755.
(9) Doyle, L. M.; O'Sullivan, S.; Salvo, C. D.; McKinnney, M.; McArdle, P.; and Murphy, P. V. Stereoselective Epimerization of Glycosyl Thiols. *Org. Lett.* **2017**, *19* (21), 5802-5805.
(10) Wolfrom, M. L.; Husted, R. A Beta to Alpha Conversion of Fully Acetylated Sugars by Alkali. *J. Am. Chem. Soc.* **1937**, *59*, 364–365.
(11) Lindberg, B. The Beta to Alpha Transformation of Fully Acetylated Glycosides by Alkali. *Acta Chem. Scand.* **1950**, *4*, 49-51.
(12) Koeners, H. J.; de Kok, A. J.; Romers, C. The Use of the 2,4-Dinitrophenyl Group in Sugar Chemistry Re-examined. *Red. Trau Chim. Pays-Bas* **1980**, *99*, 355-362.
(13) Mandell, L.; Moncrief, J. W.; Goldstein, J. H. Novel Process for Anomerization of D-Glucopyranose Pentaacetate Involving Imidazole-catalyzed Acyl Transfer. *Tetrahedron Lett.* **1963**, *4*, 209-210.
(14) Williamson, C. B.; Nevers, D. R.; Nelson, A.; Hadar, I.; Banin, U.; Hanrath, T.; Robinson, R. D. Chemically reversible isomerization of inorganic clusters. *Science*, **2019**, *363*, 731.
(15) During our submission (our first version preprint in 2018 see: DOI: 10.26434/chemrxiv.7257182.v1), a similar "milling and equilibration" type article on acetylation of wet cellulose was reported, see: Beaumont, M.; Jusner, P.; Gierlinger, N.; King, A. W. T.; Potthast, A.; Rojas, O. J. & Rosenau, T. Unique Reactivity of Nanoporous Cellulosic Materials Mediated by Surface-confined Water. *Nat. Commun.* **2021**, *12*:2513.
(16) Recrystallization of both imidazole and β-D-glucose pentaacetate for the anomerization was also investigated and result indicated impurities showed little effect on yield.
(17) Mandell, L.; Moncrief, J. W.; Goldstein, J. H. Imidazole as a Catalyst in Acyl Transfer. A Model Enzyme System for Physiological Transacetylation. *Tetrahedron* **1963**, *19*, 2025-2030.
(18) Bervend, L. A.; Olphin, A.; Withers, S. G. A Novel Mechanism of Glycoside Anomerization. *J. Am. Chem. Soc.* **1988**, *110*, 4864–4866.
(19) Bervend, L. A.; Olphin, A.; Withers, S. G. The Base-catalysed Anomerization of Dinitrophenyl Glycosides: Evidence for A Novel Reaction Mechanism. *Can. J. Chem.* **1990**, *68*, 1859–1866.
(20) Suchý, M.; Elmehriki, A. A. H.; Hudson, R. H. E. A Remarkably Simple Protocol for the N-Formylation of Amino Acid Esters and Primary Amines. *Org. Lett.* **2011**, *12*, 3952–3955.
(21) Bylera, K. G.; Li, Y.; Houghtena, R. A. and Martinez-Mayorga, K. The Role of Imidazole in Peptide Cyclization by Transesterification: Parallels to the Catalytic Triads of Serine Proteases. *Org. Biomol. Chem.* **2013**, *11*, 2979–2987.
(22) Chevallier, O. and Migaud, M. Investigation of Acetyl Migrations in Furanosides. *Beilstein J. Org. Chem.* **2006**, *2*:14.
(23) Lassfolk, R.; Rahkila, J.; Johansson, M. P.; Ekholm, F. S.; Wärnå, J. and Leino R. Acetyl Group Migration Across the Saccharide Units in Oligomannoside Model Compound. *J. Am. Chem. Soc.* **2019**, *141*, 1646–1654.
(24) Aftergut, S. & Brown, G. P. Electronic Properties of Imidazole. *Nature* **1961**, *191*, 379–380.

(25) Craven, B. M.; McMullan, R. K.; Bell, J. D. and Freeman, H. C. The Crystal Structure of Imidazole by Neutron Diffraction at 20°C and -150°C. *Acta Cryst.* **1977**, *B33*, 2585-2589.
(26) Daycock, J. T.; Jones, G. P.; Evans, J. R. N. & Thomas, J. M. Rotation of Imidazole in the Solid State and its Significance in deciding the Nature of Charge Migration in Biological Materials. *Nature*, **1968**, *218*, 672–673.
(27) Wu, C.; Alqahtani, A.; Sangtarash, S.; Vezzoli, A.; Sadeghi, H.; Robertson, C. M.; Cai, C.; Lambert, C. J.; Higgins, S. J. and Nichols, R. J. In situ Formation of H-bonding Imidazole Chains in Break-junction Experiments. *Nanoscale*, **2020**, *12*, 7914-7920.
(28) Korolev, K. G.; Lomovsky, O. I.; Uvarov, N. F. and Salenko, V. L. Mechanochemical Transformations of the Crystalline Anomers of D-Glucose. *Chemistry for Sustainable Development*, **2004**, *12*, 339.