internally captured catalysts, which are then able to process artificial substrates that are designed to interact with the metal catalysts (Fig. 1). This novel design offers two significant improvements over previous strategies[5–7]. First, the chemoresponsive nanoreactors can accommodate different metal complexes, which potentially enable a wider range of reactions to be catalysed. And second, intracellular modulation of different bioorthogonal tasks can be controlled by addition of man-made triggering reagents.

To demonstrate the biocompatibility of their approach, the team performed an intraendosomal synthesis of a fluorescent substance[5,6] through two successive bioorthogonal processes: supramolecular activation and abiotic catalysis (Fig. 1). In this experiment the team used their nanoparticle device containing either a Ru or Pd catalyst to cleave the allylcarbamate groups of bis-*N,N′*-allyloxycarbonyl rhodamine 110 inside HeLa cells. This cleavage reaction turns on the fluorescence of the rhodamine dye, enabling the progress of the catalytic reaction to be monitored. In another demonstration, which is of high significance for biomedical applications, Rotello and co-workers showed that a device containing a Pd catalyst is capable of intracellular activation of prodrugs masked by a Pd-labile propargyl moiety[7]. In this experiment cell death was induced by formation of the chemotherapeutic drug 5-fluorouracil from a bioorthogonal precursor, showing the capability of this approach to manufacture drugs. It is important to note that the resulting decrease in cell viability also confirms the integration of abiotic catalysis and metabolic pathways in a one-pot (or one-cell) cascade sequence because to exert its function the generated 5-fluorouracil must be converted into cytotoxic nucleotides by intracellular enzymes.

Although Rotello's catalytic system closely resembles the performance and allosteric regulation of natural enzymes, the reversibility of the regulatory process has not yet been demonstrated inside cells. To enable such reversible control might require a new and improved design — for instance one that limits the freedom of the gatekeeper molecules by connecting them to the alkanethiols units via soft spring linkers. Given the versatility of the device, additional functional improvements could also be introduced, such as photo-responsive features or the encapsulation of two or more non-biological transition metal catalysts in the same device. This would expand the range of bioorthogonal tasks that could be conducted and could open up abiotic cascade reactions. Another problem that must be overcome before these nanobots could be applied in medicine is the incorporation of cell-targeting capabilities. Such features could be simply introduced by decorating the hydrophobic chains or the gatekeeper molecules with specific targeting ligands, such as small molecules, peptides or aptamers, to facilitate, for example, binding to cell surface receptors that are overexpressed in cancer cells.

Despite these limitations the concept of integrating bioorthogonal catalysts and mechanochemical components into artificial nanostructures is an exciting one and paves the way for the exploration of a new kind of man-made nanomachine. To fulfil their therapeutic potential, such devices will need to be 'fuelled' with specifically designed substrates, such as bioorthogonal prodrugs[7]. It is therefore likely that future research will include developments to both nanomachines and their fuels. Until the time comes when remotely controlled features can be incorporated into miniaturized machines, it seems likely that the scheduling of chemical events will be required to intercede and organise the performance of multitasking nanodevices. Bioorthogonal chemistry provides one useful way of controlling these challenging events within living systems. ❐

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STEREOSELECTIVE SYNTHESIS

Molecular editing of carbohydrates

Deoxygenation reactions have been used to convert biomass-derived carbohydrates into useful platform chemicals. Now, a method has been described that can selectively excise C–O bonds to produce valuable chiral synthons.

Andrew McNally

Carbohydrates, formed from CO_2, water and the action of photosynthesis, are one of the most abundant molecular resources[1,2]. Despite their ubiquity, their 1:1 oxygen-to-carbon ratio (C_6O_6), coupled with their complex structure and reactivity, has hampered their use as chemical feedstocks. Direct catalytic methods that can selectively 'edit out' specific OH groups within carbohydrates have so far been unavailable to synthetic chemists. Writing in *Nature Chemistry*, Michel Gagné and co-workers now report[3] that a boron Lewis acid and a silane reducing agent can effect the partial reduction of carbohydrates and generate a range of valuable chiral building blocks.

Plant material, or lignocellulosic biomass, is the most abundant source of renewable organic matter on Earth and offers a potentially sustainable alternative to petroleum for chemical production. The majority of lignocellulosic biomass consists of carbohydrate polymers (cellulose and hemicellulose) that can be further broken down into C_5- and C_6-sugars[4]. Compared with petrochemicals, which are typically simple hydrocarbons, carbohydrates are carbon frameworks with a complex stereochemical pattern of hydroxyl groups.

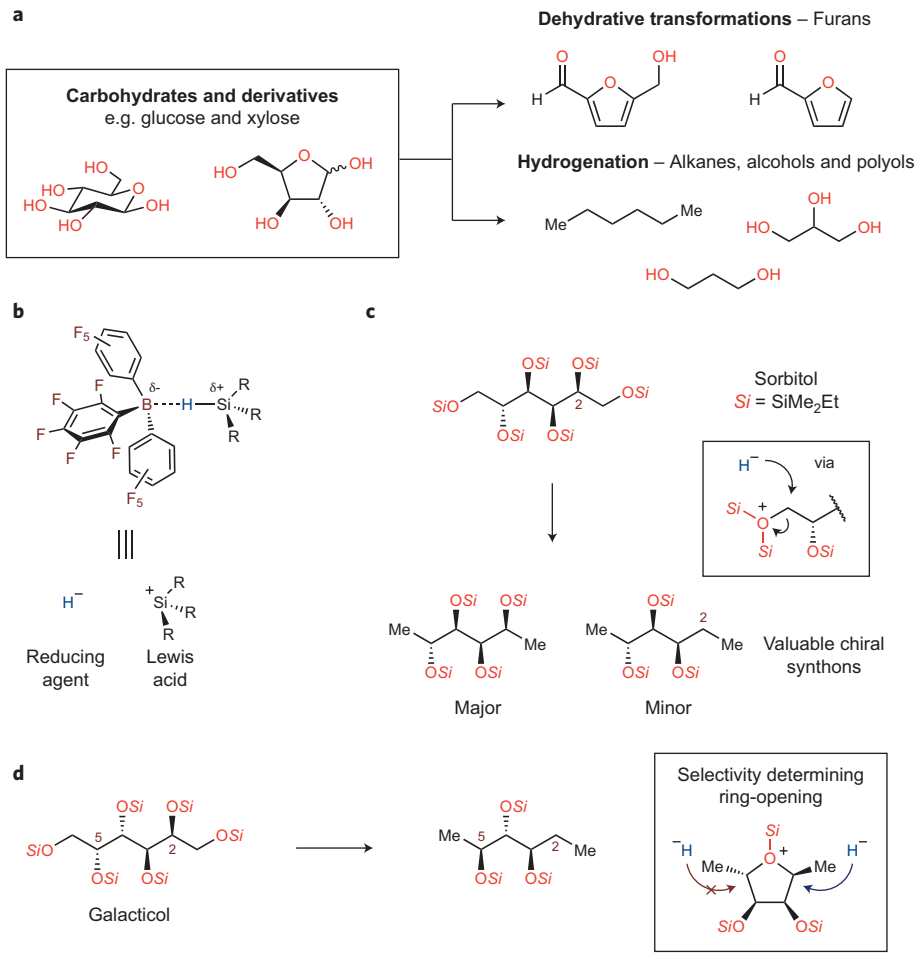

Figure 1 | Reduction of biomass-derived carbohydrates. **a**, Until now, reduction has focused on dehydration or hydrogenation reactions for producing platform chemicals on a large scale. **b**, A combination of tris(pentafluorophenyl)borane and a trialkylsilane reacts like a combination of a silylium Lewis acid and a hydride reducing agent. **c**, Sorbitol reduction results in a mixture of tetraol and triol with reduction of primary carbinols dominating. **d**, A similar reduction of galacticol leads to the development of a model to explain selectivity that invokes a cyclic intermediate.

This overabundance of oxygen atoms compared with petrochemicals imparts poor volatility properties that limits their use as fuels and also makes facile transformation into a broad spectrum of high-volume commodity chemicals challenging. Current approaches for producing useful chemicals from carbohydrates include fermentation, which is an effective strategy for producing bioethanol, and chemical methods aimed at partial or complete deoxygenation (Fig. 1a). Within these, dehydrative techniques convert carbohydrates to furans — such as hydroxymethyl furfural[5] — which are emerging as renewable platform chemicals. Furthermore, hydrogenation processes enable reduction of sugars to alkanes or simple alcohols and polyols[6].

Gagné and co-workers have developed a more selective approach to producing chemicals from carbohydrates, where portions of the stereochemically pure array of C–O bonds are preserved while others are replaced with C–H bonds. The resulting derivatives are simplified chiral molecules that are valuable for pharmaceutical, fine chemical or synthesis applications[7]. This tactic is mechanistically distinct from other deoxygenation methods since eliminative deoxygenation ablates stereocentres and hydrogenation methods generally cannot distinguish between the hydroxyl-bearing stereocentres found in carbohydrates. Selective deoxygenation in polyol systems usually requires extensive protecting-group strategies to expose a particular hydroxyl group to deoxygenation.

Previous work from the group demonstrated that combining a commercial boron Lewis acid catalyst ($B(C_6F_5)_3$) and a secondary silane (R_2SiH_2) exhaustively deoxygenates sugars into C_6-alkanes[8]. In this new, tamed reduction system using tertiary silanes (R_3SiH), C–O bonds within simple sugar alcohols can undergo hydrodeoxygenation with remarkable specificity based on a fascinating interplay of input stereochemistry and reactivity.

A putative boron–silane intermediate is an effective source of 'R_3Si^+' and 'H^-' that act independently in solution (Fig. 1b)[9]. Persilylated sorbitol — a simple glucose derivative — was reduced to a tetraol, indicating a preference for reducing primary C–O bonds (Fig. 1c). Mechanistically, this outcome can be understood as a kinetic preference for primary silyl ethers to form disilyl oxonium ions and undergo S_N2 displacement by the borohydride counterion ($H–B((C_6F_5)_3^-$). The reaction also produces minor amounts of a triol — a result of further reduction at C2 — which indicates that selective deoxygenation of secondary C–O bonds is also possible. Furthermore, reduction of galacticol — the C4 epimer of sorbitol — led to the exclusive production of a previously unreported C_6-triol (Fig. 1d). This product is the result of reduction at C2 and inversion of configuration at C5. These observations led the team to formulate a hypothesis for secondary C–O reduction based on an internal displacement event to form cyclic oxonium ions. Regioselective ring opening by the borohydride is governed by steric factors that depend on the resident stereochemistry in the starting sugar derivative, and predictably accounts for the selective C–O bond reductions in these and other sugar alcohols.

Carbohydrate derivatives with lower levels of oxidation, such as deoxyglucose and deoxygalactose, are also amenable to this reduction protocol. Reaction outcomes in these cases differ from sugar alcohols in the predicted order of reactivity and selectivity, suggesting that the reduction pathway is intrinsically linked to the carbohydrate structure — it is substrate dependent. In the cases of isosorbide and isomannide reduction, however, the product distribution can be judiciously controlled via the structure and stoichiometry of the silane reductant. This facet of the study, where distinct cyclic and acyclic products arise from the same starting material, is an important future direction, especially for more complex sugars.

This report by Gagné and co-workers constitutes some key advances in the use of carbohydrate sources for chiral pool molecules. Access to these types of oxygenated chiral building block in only one or two chemical operations was simply not possible before this work. Several occurrences of previously unreported

C_6O_n synthons from a relatively small set of carbohydrate derivatives are a testament to the potential of this approach. There is clearly scope for more sophisticated catalysts that can have even greater control of the degree and selectivity of deoxygenation and enable the reaction to be conducted on larger scale. Ultimately, it is the distinct mechanism under which this process operates that enables the selective C–O reduction and is lacking in existing deoxygenation methods. It is therefore also conceivable that this method will be applied outside of carbohydrates as polyoxygenated molecules are central to many other applications, especially those with biological function. Although removing oxygen atoms from organic molecules has a complicated past, the future may just be getting brighter. ❐

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ANALYTICAL CHEMISTRY

Clamping down on cancer detection

An electrochemical clamp assay that enables the rapid and sensitive detection of nucleic acids containing single base mutations has now been developed. It has been shown to differentiate between cancer patient samples featuring a specific mutation, and controls from healthy donors or other cancer patients, all directly in unprocessed serum.

Irina A. Gorodetskaya and Alon A. Gorodetsky

Electrical and electrochemical sensors have been touted as a particularly promising class of next-generation clinical diagnostics due to a number of key advantages[1–3]. Importantly, such sensors facilitate the multiplexed, simultaneous detection of various analytes at low concentrations and over a broad range of conditions. In addition, the targets of interest typically do not require labelling, greatly simplifying sample handling and processing, especially when only small amounts of material are available. Furthermore, electrical strategies in general allow for data to be collected and interpreted rapidly and in real time by minimally trained operators in virtually any setting. However, despite recent impressive progress in the field of electrochemical sensors, the detection of many clinically relevant targets from unprocessed patient samples remains quite challenging.

From a clinical cancer diagnostics perspective, cell-free nucleic acids (cfNAs), such as cfDNA and cfRNA, are highly desirable targets and represent an exciting class of analytes for detection, especially through electrochemical methods[4–6]. Tumour-derived cfNAs featuring specific mutations circulate freely in the blood of patients with cancer (albeit, at very low concentrations) and constitute excellent markers for the presence and progression of disease. Therefore, the detection of the relevant mutant cfNAs directly from a patient's serum can effectively serve as a 'liquid biopsy', providing similar information to that obtained from a more invasive traditional biopsy. However, the speed, accuracy and user friendliness of the currently available methods for cfNA analysis, such as DNA sequencing (DNAseq) or the polymerase chain reaction (PCR), are not yet ideal for use with unprocessed crude clinical samples. Consequently, there exists an opportunity for the development of alternative, electrochemical cfNA detection and analysis platforms that can surmount the disadvantages of the more traditional strategies.

Writing in this issue of *Nature Chemistry*, Shana Kelley, Edward Sargent and co-workers describe an electrochemical clamp assay for the analysis of cfNAs from both laboratory and clinical patient samples[7]. The assay leverages strategies developed for a technique known as clamp PCR, which has been previously employed for the detection of DNA mutations in the laboratory[8]. In traditional PCR, low-frequency mutations can be difficult to discriminate and selectively amplify in a sample containing both mutant and wild-type DNA[8]. In clamp PCR, the introduction of peptide nucleic acid (PNA) 'clamps' that specifically bind to matched wild-type DNA facilitates selective amplification of the mutant DNA[8]. Unfortunately, clamp PCR becomes unwieldy and less accurate when applied to clinical samples, such as patients' serum, in part due to significant background amplification and thus requires extensive sample purification[7].

The reported assay affords the selectivity provided by a clamp strategy within the context of an electrochemical sensor[7]. In a typical variant of this assay, a nanostructured microelectrode is fabricated using lithographic techniques and decorated with an appropriate PNA probe, such as one complementary to a variant of the Kirsten rat sarcoma-2 virus (*KRAS*) gene, which has been implicated in various forms of cancer (Fig. 1a). Next, the resulting sensor is electrically interrogated in the absence of any nucleic acid analytes, with an electrocatalytic redox pair consisting of $Ru(NH_3)_6^{3+}$ and $Fe(CN)_6^{3-}$ yielding a background signal (Fig. 1b). A complex cocktail containing the mutant target, the clamps that bind to the undesirable sequences, and various non-complementary mutant as well as wild-type nucleic acids is then introduced. In this mixture, the clamps sequester the non-complementary nucleic acids and leave only the target free to hybridize with the microelectrode-bound probe (Fig. 1c). Finally, the sensor is electrically interrogated again, with a change in the initial signal indicating the presence of the captured negatively charged target that is, the mutant gene (Fig. 1b).

The most striking aspect of the electrochemical clamp assay is its ability to directly and rapidly detect cancer-implicated mutant cfNA in unprocessed clinical samples[7]. In proof-of-principle experiments, the team prepared microelectrodes modified with not just one type of PNA, but with multiple probes complementary to different cancer-linked mutant *KRAS* sequences. This approach ensured that the sensors could