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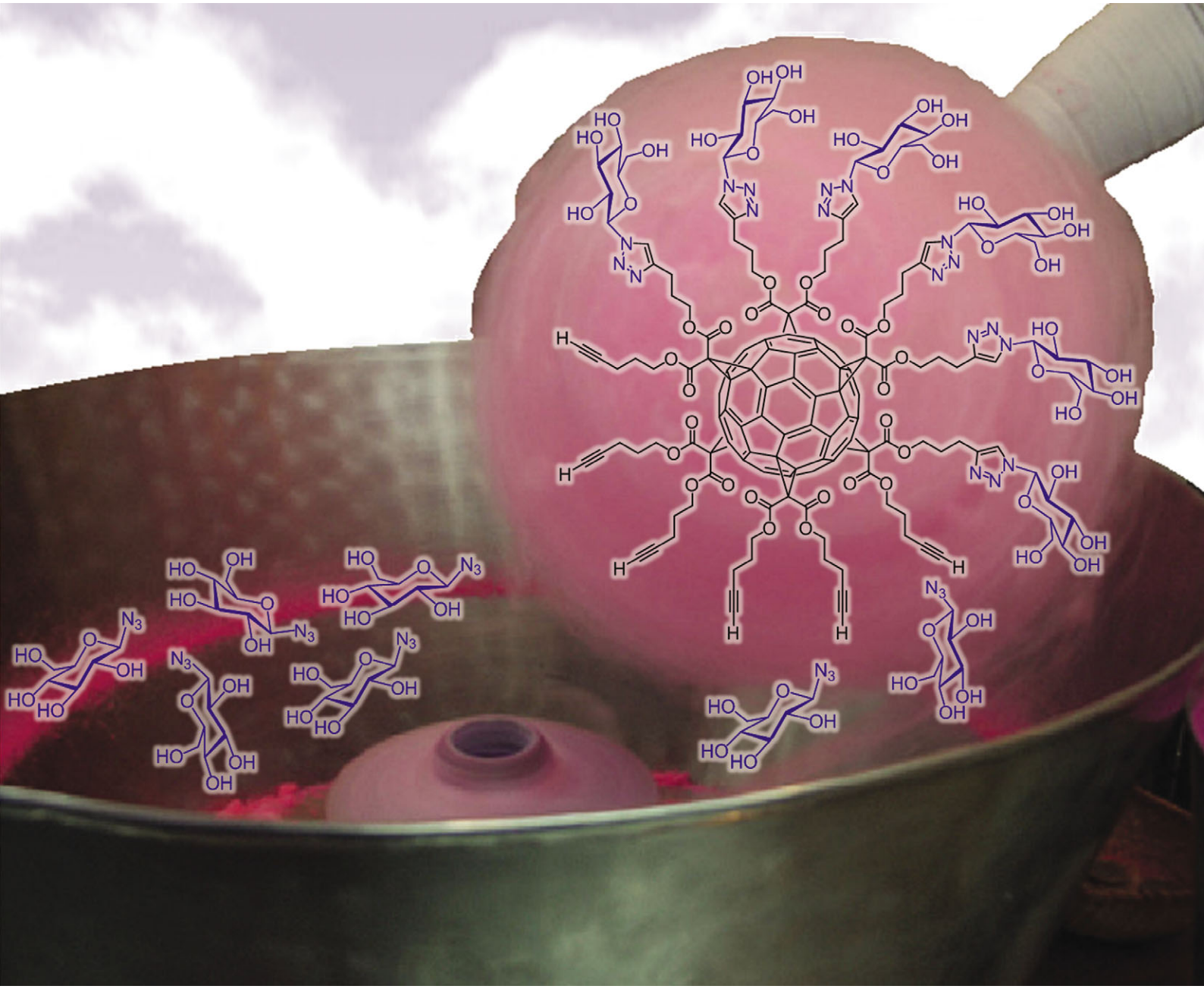

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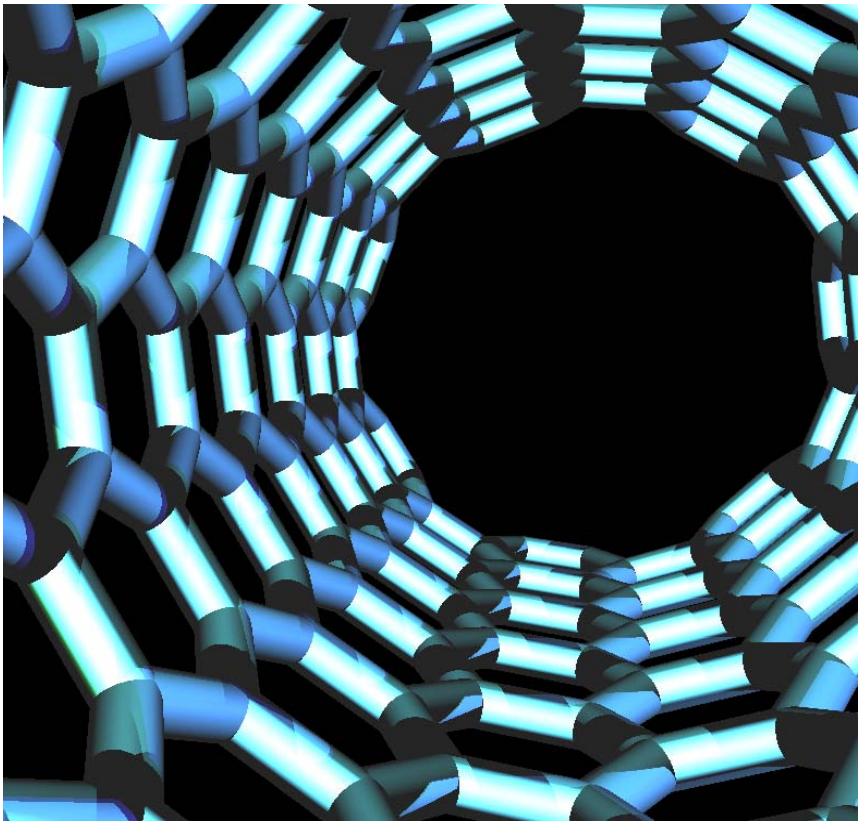

Fullerene sugar balls†‡

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Fullerene hexakis-adducts bearing 12 peripheral carbohydrate moieties have been prepared by grafting sugar derivatives onto the fullerene core through the copper mediated Huisgen 1,3-dipolar cycloaddition of azides and alkynes.

Carbohydrates play a key role in many natural and pathological processes through interaction with their corresponding receptors, mainly proteins.[1] This interaction is highly selective, calcium dependent in some cases, and multivalent.[2] The multivalent presentation of carbohydrates is a crucial point for achieving high affinity interactions and, therefore, an essential requirement for developing artificial tools with the aim to understand, unravel and interfere with those processes where carbohydrates are involved.[3] Different synthetic scaffolds allowing a multivalent presentation of ligands have already been described in the literature. Among them, the most popular are dendrimers,[4] nanoparticles,[5] polymers,[6] and liposomes.[7] Owing to their rigid spherical shape, fullerene hexakis-adducts with a T_h-symmetrical octahedral addition pattern[8] appear to be a very attractive core molecule for the synthesis of unique globular multivalent derivatives. Until now, some fullerene derivatives bearing sugar residues have been reported in which either a monoaddition pattern[9] on the fullerene sphere or a "one-side" multiaddition[10,11] led to amphiphilic structures, with a hydrophobic fullerene subunit occupying a part of the outer architecture of the molecule. The aggregation of these molecules has been documented in some cases.[11] Such amphiphilic character could be avoided by preparing fullerene hexakis-adducts, thus obtaining truly "sugar balls" and providing a spherical platform for a globular multivalent presentation of ligands. Herein, we describe the efficient preparation of fullerene sugar balls with different sugars as potential carbohydrate multivalent systems for biological applications.

As far as the synthesis of the sugar balls is concerned, the direct grafting of six malonates bearing sugar residues to the fullerene core would be quite limited. Indeed, fullerene hexakis-adducts are generally obtained in rather low yields from structurally complicated malonates.[12] Furthermore, this strategy would require the preparation of new malonates for each derivative as well as the use of protecting groups for the hydroxyl functions of the sugars. Therefore, we have decided to apply the approach based on click chemistry developed by some of us[13] for the efficient functionalization of fullerene hexakis-adducts with sugar residues. As click reactions are modular, tolerant to a wide range of functional groups, and high yielding,[14] no protecting groups are needed and a large number of sugar balls should become easily available by using this synthetic approach.

Fullerene hexakis-adduct **1** was prepared in three steps as previously described.[13b] Treatment with an excess of tetrabutylammonium fluoride (TBAF) gave the corresponding hexa-adduct **2** bearing 12 terminal alkyne units (Scheme 1).

The reaction conditions for the 1,3-dipolar cycloaddition of compound **2** with sugars bearing an azide group were first adjusted with the commercially available derivatives **3a** and **3b**. In the case of the acetylated derivative **3a**, the reaction conditions previously developed[13] for the grafting of azide groups to fullerene derivative **2** were successfully applied to give compound **4a** in excellent yield. A mixture of **2** (1 equiv.), **3a** (13 equiv.), $CuSO_4{\cdot}5H_2O$ (0.1 equiv.) and sodium ascorbate (0.3 equiv.) in CH_2Cl_2–H_2O was vigorously stirred at room temperature for 12 h. Compound **4a** was obtained in 91% yield after work-up and purification by fast filtration on SiO_2 followed by gel permeation chromatography (Biobeads SX-1, CH_2Cl_2). The reaction conditions used for the preparation of **4a** from **3a** were then adapted to unprotected azide **3b**. Actually, in CH_2Cl_2–H_2O, no reaction could be observed. The latter observation is most probably related to the difference in solubility of the two precursors, **3b** being only soluble in the aqueous phase and **2** only in the CH_2Cl_2 one. This prompted us to attempt the reaction between **2** and **3b** in different solvents. The best results were obtained in DMSO in which both **2** and **3b** are soluble. Under optimized conditions, a mixture of **2** (1 equiv.), **3b** (13 equiv.), $CuSO_4{\cdot}5H_2O$

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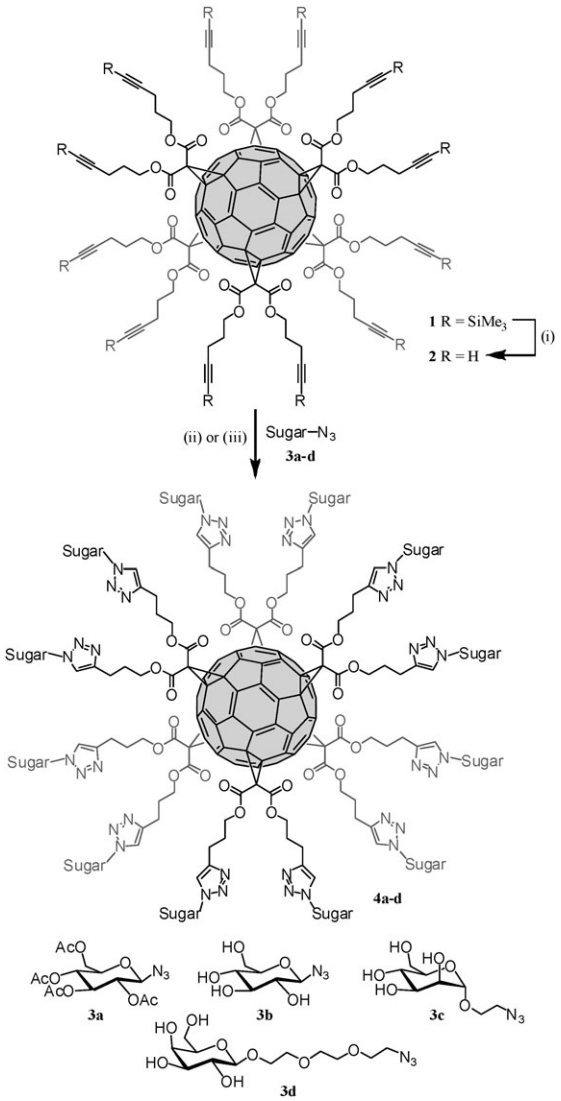
‡ Electronic supplementary information (ESI) available: Experimental details for the preparation of the new compounds and NMR data of compounds **4c–d** and **7a–d**. See DOI: 10.1039/c0cc00034e



Scheme 1 *Reagents and conditions*: (i) TBAF, CH_2Cl_2 (92%); (ii) from **2**: $CuSO_4{\cdot}5H_2O$, sodium ascorbate [**4a**: in CH_2Cl_2–H_2O (91%); **4b–d**: in DMSO (**4b**: 74%; **4c**: 90%; **4d**: 72%)]; (iii) from **1**: TBAF, $CuSO_4{\cdot}5H_2O$, sodium ascorbate [**4a**: in CH_2Cl_2–H_2O (51%); **4b**: in DMSO (**4b**: 58%)].

(0.1 equiv.) and sodium ascorbate (0.3 equiv.) in DMSO was stirred at room temperature for 60 h. At the end of the reaction, the product was precipitated by addition of MeOH, filtered and extensively washed with MeOH and CH_2Cl_2. Sugar ball **4b** was thus obtained in 74% yield. Interestingly, we found that compounds **4a–b** can also be directly prepared from fullerene derivative **1**. Indeed, compound **1** can be desilylated *in situ* with TBAF to form **2**, to which the azide precursors **3a** or **3b** can be subsequently clicked. Compounds **4a** and **4b** were thus obtained from **1** and the corresponding azide in 51 and 58% yield, respectively. The chemical structure of compound **4a** was easily confirmed by its ^{1}H and ^{13}C NMR spectra. In particular, the ^{13}C NMR spectrum of fullerene hexakis-adduct **4a** (Fig. 1) is in full agreement with its *T*-symmetrical structure and shows the expected signals for the 6 equivalent malonate addends. Only 3 signals out of the 5 expected ones are however observed for the fullerene C atoms (δ = 69.0 for the sp^3 C atom; 141.0 and 145.8 ppm for the sp^2 C atoms). Indeed, these 3 signals are reminiscent of those of the three non-equivalent fullerene C atoms of the hexakis-adduct carrying achiral addends (overall T_h symmetry). No influence of the overall symmetry of **4a** which is *T* could be deduced and the two pairs of diastereotopic sp^2 C atoms are pseudo-equivalent. Similar observations have been reported for mixed fullerene hexa-adducts[13b] and related C_{60} derivatives.[15] The MALDI-TOF mass spectrum further confirmed the structure of **4a** with an intense signal at m/z = 6606 corresponding to the expected molecular ion peak ($[M]^+$, calcd. for $C_{306}H_{312}N_{36}O_{132}$: 6606.01). The ^{1}H and ^{13}C NMR spectra of **4b** were also in perfect agreement with the proposed formulation. As shown in Fig. 1, the two expected resonances of the C atoms of the 1,2,3-triazole unit as well as the 6 signals of the 6 different C atoms of the 12 equivalent glucose residues are clearly observed in the ^{13}C NMR spectrum of **4b**. To further confirm the structure of **4b**, mass spectra (MALDI-TOF, ESMS and FAB) were recorded under different conditions. However, in all the cases, a high level of fragmentation prevented the observation of the expected molecular ion peak. We thus decided to acetylate a sample of compound **4b** to generate **4a** for which MS analysis has been successfully achieved. An aliquot of **4b** was treated with a large excess of acetic anhydride in pyridine. The resulting product was identical (TLC and ^{1}H NMR) to the original sample of **4a** prepared from **2** and **3a**. Importantly, its MALDI-TOF mass spectrum was also identical to the one obtained from an original sample of **4a**. The latter result clearly validates the structural characterization of the unprotected fullerene sugar balls based on their ^{1}H and ^{13}C NMR data.

The reaction conditions used for the preparation of **4b** from azide **3b** were then applied to the more elaborated sugar azide

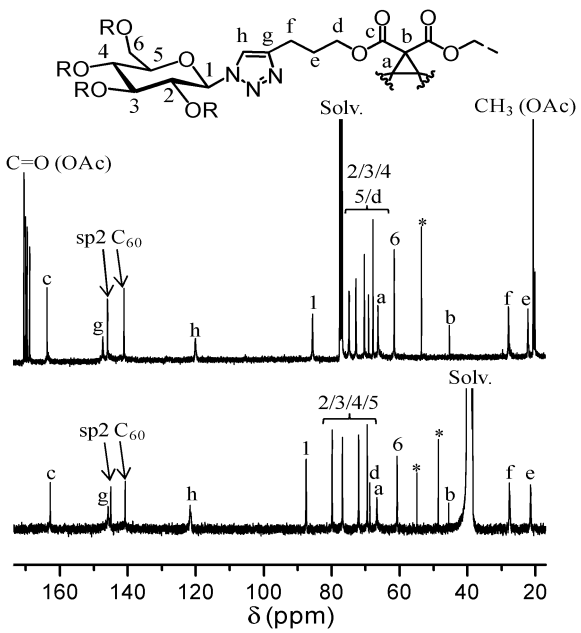
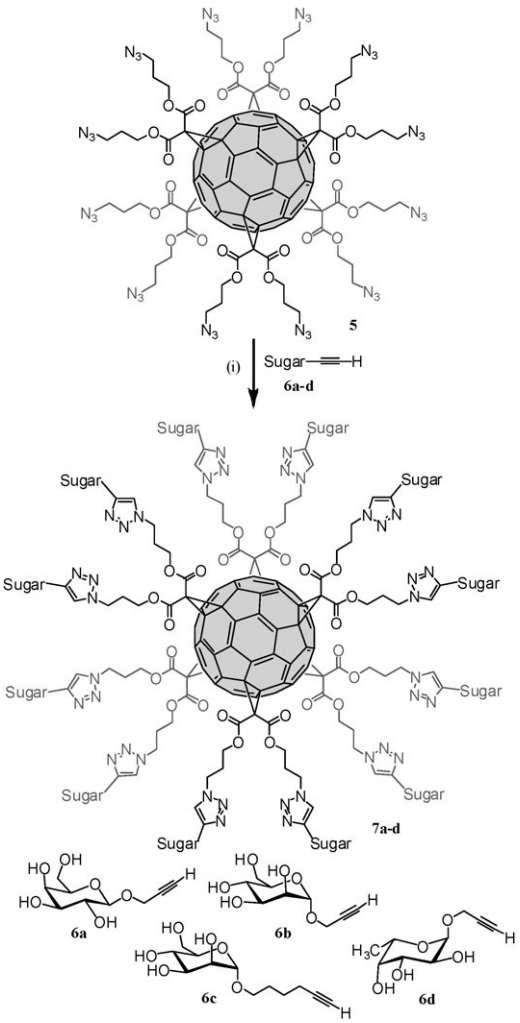


Fig. 1 ^{13}C NMR spectra of **4a** (top) recorded in $CDCl_3$ (100 MHz, *: residual CH_2Cl_2) and of **4b** (bottom) recorded in DMSO-d_6 (100 MHz, *: residual CH_2Cl_2 and MeOH); unambiguous assignment was achieved with the help of the corresponding DEPT spectra (see ESI‡).

derivatives **3c–d**. The clicked derivatives **4c–d** were thus obtained in excellent yields (72–90%) and their structure confirmed by their ^{1}H and ^{13}C NMR spectra as well as by IR spectroscopy. Furthermore, the UV/vis spectra of compounds **4a–d** show the characteristic features of fullerene hexa-adducts.[8,12] Having developed an efficient procedure for the preparation of sugar balls from a fullerene alkyne building block and sugar azide derivatives, we also decided to attempt the reverse approach starting from fullerene derivative **5** bearing 12 azide functional groups[13a] (Scheme 2).

Treatment of **5** with alkyne **6a** under the reaction conditions developed for the preparation of **4b** ($CuSO_4{\cdot}5H_2O$, sodium ascorbate, DMSO) gave only partially clicked derivatives. Indeed, these intermediates precipitate during the course of the reaction. Longer reaction time and/or higher temperatures were not efficient strategies to complete the reaction and another solvent had to be selected to allow the preparation of sugar balls from **5**. The best results were obtained when the reaction was carried out in a CH_2Cl_2–H_2O–DMSO (1 : 1 : 1) mixture. Hence, when the reaction between **5** and **6a** was performed in this ternary solvent system, compound **7a** was obtained in 73% yield. The same conditions were then successfully used for the preparation of **7b–d** from **5** and the corresponding sugar-alkyne derivatives **6b–d**. In the case of **7c**, the moderate isolated yield is explained by the difficulties in separating the product from unreacted **6c** (used in excess) during the successive dissolution/precipitation. The structure of compounds **7a–d** was confirmed by their ^{1}H and ^{13}C NMR spectra. Inspection of the ^{1}H NMR spectra clearly indicates the disappearance of the CH_2-azide signal at δ 3.33 ppm. IR data also confirmed that no azide (2092 cm^{-1}) residues remain in the final products. Importantly, the ^{1}H NMR spectra of **7a–d** show the typical signal of the 1,2,3-triazole unit at *ca.* δ 7.81–8.04 ppm.



Scheme 2 *Reagents and conditions*: (i) $CuSO_4{\cdot}5H_2O$, sodium ascorbate, CH_2Cl_2–H_2O–DMSO 1 : 1 : 1 (**7a**: 73%; **7b**: 81%; **7c**: 40%; **7d**: 54%).

In conclusion, we have reported for the first time on the synthesis of fullerene glycoconjugates in which the C_{60} core is completely surrounded by sugar residues. The methodology based on the Huisgen-type click chemistry allows for the easy preparation of a large variety of sugar balls. The peculiar spherical distribution of the sugar residues gives rise to unprecedented globular polytopic ligands and their biological activities are under investigation.

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