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A new method of solvent free *O*- and *N*-glycosylation using activated carbon fiber (ACF) as a promoter. Application to the synthesis of saponin and nucleoside analogues

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Received (in Cambridge, UK) 2nd December 2003, Accepted 15th January 2004
First published as an Advance Article on the web 3rd February 2004

By using activated carbon fiber (ACF) as solid acid promoter, we display a new system for solvent free *O*- and *N*-glycosylation, with a simple process which takes place in a drying oven; this method has been applied to the synthesis of sterol and triterpene *O*-glycosides (saponin analogues), as well as nucleoside analogues.

Interest in "Green Chemistry" has grown over the past decade.[1] Solid-state synthesis in fine chemistry appears as a worthwhile process from an ecological point of view because reactions take place in solvent free conditions and often in the presence of an environmentally harmless catalyst.[2] In this communication, we describe a novel and practical solid-state protocol suitable for *O*-glycosylation of different sterols and triterpenes, and *N*-glycosylation of 6-chloropurine in the presence of activated carbon fiber (ACF) as a solid acid promoter. ACFs constitute a new class of activated carbon which could be manufactured in different forms (cloth, pellets, felt, …) and which are obtained from natural or synthetic raw materials (cellulose, viscose, polyacrylonitrile, …). Due to a large pore volume, mainly consisting of micropores, ACFs exhibit an extremely high surface area.[3] These characteristics give them excellent adsorption properties.[4] Therefore, ACFs can be used to clean air or waste water.[5]

We focused our work on *O*-glycosylation and *N*-glycosylation reactions because the compounds obtained may exert a large range of pharmacological and biological activities;[6] some of them can be ascribed to the aglycone moiety, others to the carbohydrate residue. Their activity is often dependent on the whole structure. The carbohydrate group and the aglycone moiety constitute a hydrophilic part and a hydrophobic part respectively: this kind of conjugation can increase water solubility without affecting the therapeutic efficacy. Glycosylation reactions have previously been promoted by Lewis acids, which generally are harmful to the environment. The goal of this work is to show the ability of acidic ACF to replace Lewis acid catalysts in *O*-glycoside and *N*-glycoside synthesis, by using a solvent free procedure. Reactants used are represented in Fig. 1. Some of them possess various biological properties, like betulin (**4**) or sitosterol (**3**). In fact, betulin,[7] a compound present in the white spots of birch bark, constitutes a good precursor of betulinic acid, a selective inducer of apoptosis in melanoma cells,[8] in neuroectodermal tumors[9] and which acts as a good antiviral agent against HIV.[10] Biological properties of sitosterol have been recently underlined in relation to its hypocholesterolaemic activity.[11] Cholesterol (**1**) and cholestanol (**2**) also have biological activity by themselves but, associated with a sugar moiety, these compounds could present anticancer properties[12] or could be incorporated into cosmetics.[13] Chloropurine[14] is often used as a precursor in biologically active nucleoside analogue syntheses.

ACF, supplied by SOFRANCE (Nexon, France), was manufactured in cloth form (ACC) and was obtained from viscose rayon.[15] The surface area was measured by the BET[16] method and was about 1000 $m^2\ g^{-1}$, and the average micropore diameter was 0.6 nm. The amount of acidic sites has been evaluated by Boehm's method[17] (4.66 10^{-3} mol g^{-1}). Before use, ACC was crushed and put in a drying oven at 100 °C for 24 hours. Ground ACC was examined with SEM (Fig. 2). The grain size of ground ACC was approximately 10 μm. Reaction mixtures consisted of 50 mg (2

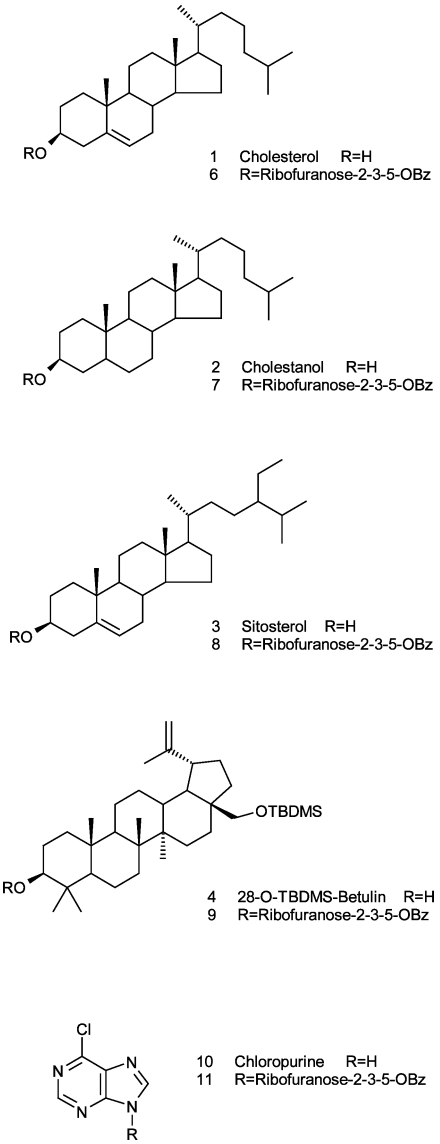


Fig. 1 Reactants and products.

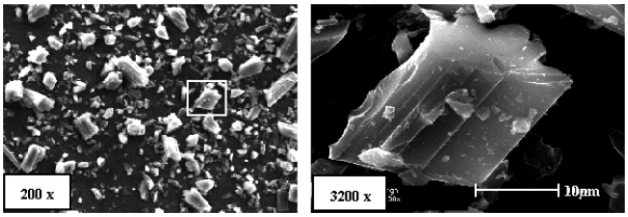
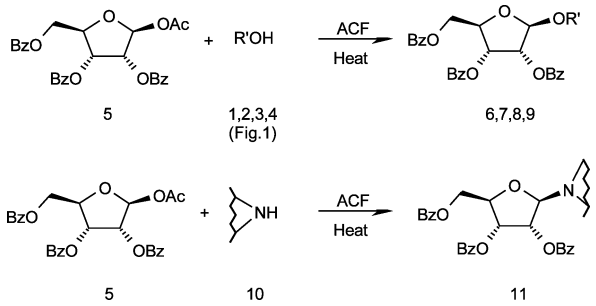


Fig. 2 Observations of ground ACC by SEM.

DOI: 10.1039/b315699k



Scheme 1 Solvent free *O*-glycosylation and *N*-glycosylation with ACF as promoter.

Table 1 Results of *O*-glycosylation and *N*-glycosylation with ACF

Reactants	Products	Reaction time/h	Temperature/ °C	Yields (%)
1	**6**	24	100	93
2	**7**	24	100	82
3	**8**	1	200	56
4	**9**	16	160	64
10	**11**	0.5	200	53

equiv.) of β-D-ribofuranose-2,3,5-tribenzoate-1-acetate ground in a mortar with sterol/triterpene or chloropurine (1 equiv.) and ACC (75 mg) until a fine powder was obtained.

Then, the mixture was put in a drying oven for the appropriate reaction time (Scheme 1). The temperature of the oven was set above the lowest melting point of solid reagents. A softening of the mixture, which then hardened at room temperature, was observed. This melted phase allowed the minimal motion of the reagents required by the reaction. Then, the medium was extracted with a minimum of dichloromethane–ethanol (90 : 10) and the end products were then isolated by thin layer chromatography (toluene–ethyl acetate–petroleum ether, 6 : 2 : 2). Results are shown in Table 1. All the structures were established on the basis of ^{1}H NMR spectroscopy.†

All reactions have been optimized by studying the effects of time and temperature on reaction yields. Only the best conditions have been reported in this note. For example, the synthesis of sitosteryl ribofuranoside-2′,3′,5′-tribenzoate has been studied for given times (0.5 to 24 hours) and different temperatures (100–210 °C). The results show a selective formation of β-ribofuranosides which, after deprotection with an appropriate reagent (sodium methylate in methanol), could be incorporated into cosmetics or drugs. This kind of synthesis is worthwhile from an environmental point of view because it takes place in the solid state, in the absence of harmful solvent. Moreover, the simplicity of the reaction procedure associated with the good yields (50–90%) permits us to forecast an application in fine chemistry, notably in glycosylation reactions.

In summary, this paper presents for the first time an efficient and new method for *O*- and *N*-glycosylation using ACF under solvent free conditions. The approach described here could be applied to other compounds. Additional glycosides are currently under investigation in our laboratory.

Notes and references

† ^{1}H NMR data of the synthesized products: cholesteryl-β-D-*O*-ribofuranoside-2′,3′,5′-tribenzoate (**6**) ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.5 (15H OBz, m); 5.88 (H-3′, dd, 4.9 and 6.4 Hz); 5.64 (H-2′, d, 4.8); 5.41 (H-1′, s); 5.34 (H-6, d, 5.1); 4.69 (1H-5′a, dd, 4.4 and 12.8); 4.68 (H-4′, m); 4.53 (1H-5′b, dd, 6.8 and 13); 3.56 (H-3, m); 0.66 (CH_3, s). For cholestanyl-β-D-*O*-ribofuranoside-2′,3′,5′-tribenzoate (**7**) ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.5 (15H OBz, m); 5.87 (H-3′, dd, 5 and 6.2 Hz); 5.61 (H-2′, d, 5); 5.41 (H-1′, s); 4.69 (1H-5′a, dd, 4.4 and 12.8); 4.68 (H-4′, m); 4.53 (1H-5′b, dd, 6.8 and 12.9); 3.63 (H-3, m); 0.73 (CH_3, s); 0.61 (CH_3, s). For sitosteryl-β-D-*O*-ribofuranoside-2′,3′,5′-tribenzoate (**8**) ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.5 (15H OBz, m); 5.88 (H-3′, dd, 4.9 and 6.4 Hz); 5.64 (H-2′, d, 4.8); 5.41 (H-1′, s); 5.34 (H-6, d, 5); 4.69 (H-4′, m); 4.68 (1H-5′a, dd, 6 and 12.8); 4.33 (1H-5′b, dd, 6.8 and 13); 3.56 (H-3, m); 0.67 (CH_3, s). For 28-*O*-TBDMS-betulin-β-D-*O*-ribofuranoside-2′,3′,5′-tribenzoate (**9**) ^{1}H NMR (400 MHz, CD_3OD + $CDCl_3$) δ 7.5 (15H OBz, m); 5.83 (H-3′, dd, 5 and 6); 5.65 (H-2′, d, 4.8); 5.33 (H-1′, s); 4.69 (H-4′, m); 4.69 (1H-5′a, m); 4.69 (1H-29_a, m); 4.56 (1H-29_b, s); 4.53 (1H-5′b, dd, 6.3 and 12.3); 3.25 (1H-28_a, d, 9.7); 3.18 (1H-28_b, m); 3.13 (H-3, dd, 5 and 12.1); 2.39 (H-13, m); 1.67 (CH_3, s); 1.00 (CH_3, s); 0.97 (CH_3, s); 0.95 (CH_3, s); 0.89 (Si-C(CH_3)$_3$, s); 0.88 (CH_3, s); 0.76 (CH_3, s); 0.68 (H-5, d, 9.3); 0.04 ($2CH_3$-Si, s). For 6-chloropurine ribofuranoside-2′,3′,5′-tribenzoate (**11**) ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.6 (1H-2, s); 8.3 (H-8, s); 7.5 (15H OBz, m); 6.45 (H-1′, d, 5); 6.41 (H-2′, dd, 5.2 and 5.5); 6.25 (H-3′, dd, 5.5 and 5.3); 4.94 (1-H5′$_a$, dd, 3 and 12.2); 4.85 (H-4′, m);.4.7 (1H-5′$_b$, dd, 4 and 12.2).

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