Available online at www.sciencedirect.com
ScienceDirect
Carbohydrate Research 343 (2008) 2067–2074
Carbohydrate RESEARCH

'Naturalization' of textile disperse dyes through glycoconjugation: the case of a bis(2-hydroxyethyl) group containing azo dye

Roberto Bianchini,[a,*] Giorgio Catelani,[b,*] Riccardo Cecconi,[c] Felicia D'Andrea,[b] Elena Frino,[b] Jalal Isaad[a] and Massimo Rolla[a]

[a]*Dipartimento di Chimica Organica 'U. Shiff', Università di Firenze, Via della Lastruccia 13, I-50019 Sesto Fiorentino (FI), Italy*
[b]*Dipartimento di Chimica Bioorganica e Biofarmacia, Università di Pisa, Via Bonanno 33, I-56126, Pisa, Italy*
[c]*Lanartex snc Laboratory, Via 1 Maggio, Montemurlo (PO), I-59013, Italy*

Received 27 November 2007; received in revised form 7 February 2008; accepted 8 February 2008
Available online 15 February 2008

Presented at Eurocarb 14th Lübeck, Germany, September 2007

Abstract—A family of five strictly related glycoconjugated azo dyes (GADs), characterized by the presence of the same chromophore and a variable number (1–4) of deprotected hexose units, has been prepared by employing succinate bridges for connecting the azo dye and the sugar portions. The modulation of the hydrophilic portion determines the appreciable changes in the water solubility of GADs. In all the cases, however, hydrophobic fibres (polyester) were homogeneously dyed with GADs at temperatures lower than that used for original azo dyes, at atmospheric pressure, and avoiding the use of surfactants. Furthermore, GADs show an interesting multipurpose character leading to dyeing well also the natural fibres as, for instance, wool. The presence of a variable number of hexose units in the different GADs determines some changes in the colour intensity of dyed fabrics, but in all the cases an appreciable rubbing and water fastness were maintained.

Keywords: Azodyes; D-Galactose; Lactose; Succinates; Glycoconjugation; Textile dyeing

1. Introduction

Archaeological traces of dyed clothes have been found through the world, showing that ancient civilizations in America, in the Mediterranean area and in the Central Asia developed efficient dyeing techniques using natural dyestuffs prevalently obtained from plants, but also from insects and shellfish.[1] Until the second half of the 19th century, textile industry developed more and more sophisticated techniques obtaining an impressive level of quality in dyeing fabrics with natural dyestuff.[1] Many vegetable natural colours are constituted by different classes of chromophores O- or C-glycosylated with mono-, di- or oligosaccharide moieties. Examples are[1] the cartamine, a deep red glycoside from Safflower (*Cartamus tinctorius*), the blue indicotine from Woad (*Isatis tinctoria*), obtained by the hydrolysis of the colourless glycoside indacane, the ruberitric acid from Madder (*Rubia tinctorum*), a glycoside of shining red colour constituted from alizarine and the disaccharide primaverose. Starting from the second half of the 19th century, following the Industrial Revolution, natural dyes have been almost completely replaced by synthetic compounds, which are less expensive and much easily available. Nowadays, textile industries dispose off a huge number (about 10,000) of synthetic dyes,[2] produced by chemical industries throughout the world in an estimated overall production of 700,000 tons/year.[2]

Amongst the different classes of synthetic dyes currently used in the textile industry, the by far most common is constituted by the so-called 'disperse dyes', represented mainly by the azo and the anthraquinone

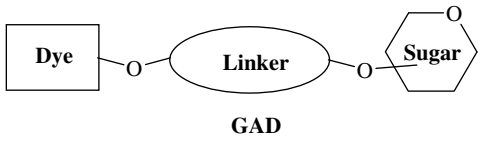
* Corresponding authors. Tel.: +39 0502219700; fax: +39 0502219660 (G.C.); tel.: +39 0554573486; fax: +39 0554573531 (R.B.); e-mail addresses: roberto.bianchini@unifi.it; giocate@farm.unipi.it

0008-6215/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.carres.2008.02.009

families.[2] Industrial dyeing processes are generally carried out in aqueous solution and require several auxiliary chemicals, as for instance surfactants.[3] The decontamination of effluents is a difficult task because the type and the extent of contamination largely vary depending either on the fabric dyed or the class of dye used. Furthermore, an approach coming back to an extensive use of natural dyes extracted from plants or animals at present is not a realistic hypothesis, because of the high quantities of dyes required by the global market, the impact of the extraction processes and the general low affinity of natural dyes with fibres requiring the use of specific chemical auxiliaries.

A recently proposed[4] innovative type of textile dyes is based on the idea of mimicking the structure of natural dyes through the modification of commercial synthetic disperse dyes by their glycoconjugation. A first family of glycoconjugated azo dyes (GADs, Chart 1) has been obtained,[5] putting a succinyl bridge from the azo chromophore and a selected, nonanomeric position of easily available mono- or disaccharides (D-glucose, D-galactose and lactose). The GADs obtained by this 'naturalization' method showed interesting dyeing properties. More recently, a second series of GADs, having a diethereal linker, have also been proposed.[6] In all cases, the appreciable water solubility of the GADs permits the development of effective dyeing processes in water employing reduced amounts of dye, without the addition of surfactants or other additives, under mild conditions of temperature and pressure.[4,5] Furthermore, GADs acquire an unexpected multipurpose nature, allowing a large kind of different fabrics (wool, silk, nylon, polyester, acrylic, polyacetate and polyurethane) to be dyed,[4,5] opening thus the way to solve the challenging problems related to the efficient dyeing of fabric blends.

A remaining problem to be solved is related to the modulation of the hydrophilic pocket, that is, the sugar portion of GADs, in view of their application in industrial processes allowing the most favourable compromise between water solubility and colour fastness.



GAD

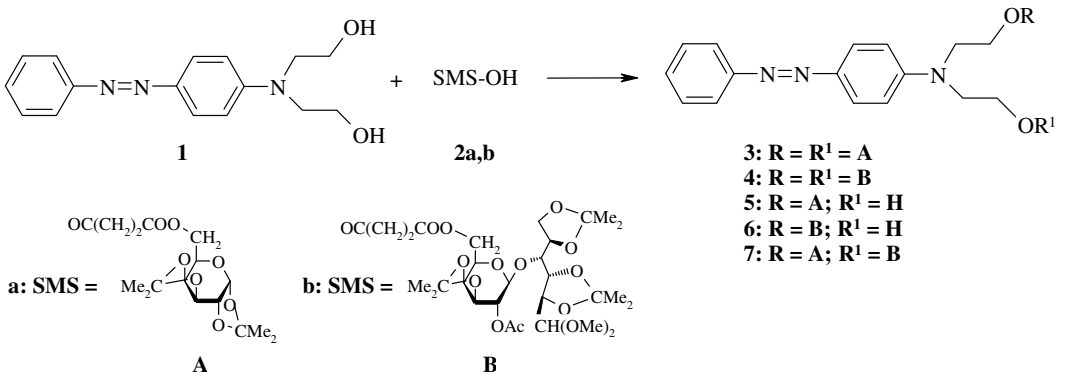
Chart 1. Schematic representation of a glycoazodye (GAD).

We are presenting now the synthesis of a family of five structurally related GADs obtained by the conjugation of one or two mono- or disaccharide units to the same synthetic yellow azo dye **1**,[7] characterized by the presence of a bis(2-hydroxyethyl) group exposing thus, two equivalent primary alcoholic functions (Scheme 1).

2. Results and discussion

2.1. Synthesis of glycoconjugated azo dyes (GADs)

The preparation of the protected bis-conjugated succinyl GAD **3** was easily achieved by the treatment of a THF solution of azo dye **1** and a slight excess (2.2 mol) of the known[5] protected sugar monosuccinate (SMS-OH) **2a** with *N*′-(3-dimethylaminopropyl)-*N*-ethylcarbodiimide hydrochloride (EDC, 2.2 mol) in the presence of dimethylaminopyridine (DMAP). In accordance with our previous results on the conjugation of monohydroxy azodyes, the expected bis-succinate **3** was obtained in about 88% isolated yield. A very close result was obtained in the preparation of the analogous protected lactose GAD **4**. As expected, the preparation of mono-conjugated azo dyes **5** and **6** was much more complicated giving mixtures of mono- and bis-esterificated products and unreacted diol when a 1:1 molar ratio of dye and sugar monosuccinate was allowed to react. An acceptable yield of mono-conjugated GAD **5** was obtained (49% based on the sugar and 59% based on the reacted dye) by performing a slow addition at 0 °C of the sugar monosuccinate (**2a** and **2b**) to a solution containing an excess of dye **1** (1.5 mol **2a**) in the presence of EDC and DMAP. In an analogous manner,



Scheme 1. Synthesis of protected GADs.

GAD **6** was obtained in a satisfactory 49% yield. The mixed bis-conjugated protected GAD **7** was obtained in 59% yield submitting the galactosyl monosuccinate alcoholic azo dye **5** to a second EDC promoted esterification with the protected lactose monosuccinate **2b**.

The deprotection procedure for compounds **3**–**7** was carried out in all cases through short (0.5–2 h) treatment at room temperature with 90% aq CF_3COOH, followed by concentration under diminished pressure of the acid solutions. In all cases, dark red-violet glassy residues were obtained, evidently constituted by trifluoroacetate salts of the acid form of GADs. The structure of cationic form of aminoazobenzene has been the object of investigations[8] because of the presence of a tautomerism between the anilium (**C**) and the azonium (**D**) form, as indicated in Chart 2.

NMR data taken on the trifluoroacetate of the monogalactosylated GAD **8** (Chart 3) show sensible shifts on both the proton and the carbon chemical shifts of the aminobenzene ring, but the discussion on either isomer prevails is out of the scope of the present work. The neutralization of the red-violet residues was finally carried out through dissolution in aq saturated $NaHCO_3$ and a final extensive extraction with EtOAc. In the cases of the more hydrophilic GADs **11** and **12**, this procedure was slightly modified in the neutralization step, requiring dissolution in THF and neutralization with solid $NaHCO_3$. In all cases, GADs were obtained in quite good yields as yellow-orange amorphous residues constituted by mixtures of anomers, as ascertained by NMR analysis (see Section 3).

2.2. Dyeing properties of glycoconjugated azodyes (GADs)

In order to verify the influence of the different hydrophilic pockets of GADs on their dyeing properties, preliminary tests were performed only on GADs **8**, **10** and **11** characterized by the presence of 1, 2 and 4 deprotected hexose units, respectively. These new GADs present differentiated water solubility: compound **11** is soluble in cold water just upon addition, **10** is soluble under some stirring and **8** is soluble after warming and stirring. As previously found,[4–6] these new GADs are multipurpose, since they are used to dye all fabrics tested so far. Reported here (Fig. 1) are the preliminary results obtained with two extremely different fabrics. Wool (Fig. 1, left side) is a representative of natural hydrophilic fabrics for which, to the best of our knowledge, dyeing procedures with disperse dyes have not yet been described. Ultramicrofibre polyester is a representative of hydrophobic synthetic fabrics requiring drastic conditions (high temperature under pressure) and addition of surface-active compounds for allowing a satisfactory dyeing.

Dyeing conditions used in this trial for deprotected GADs **8**, **10** and **11** are those reported in Table 1.

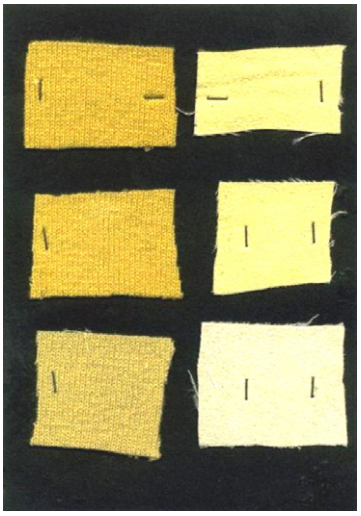
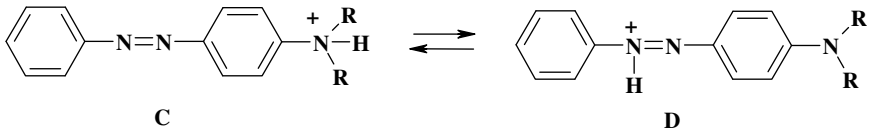
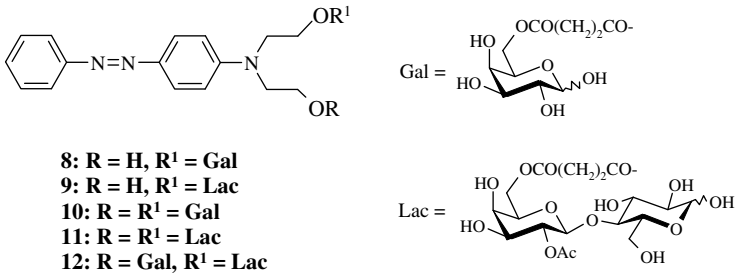


Figure 1. Tinctorial test on wool (left side) and polyester (right side) of compounds **8**, **10** and **11** (from top to bottom).



C **D**

Chart 2. Tautomeric equilibrium between anilinium and azonium forms of aminoazobenzenes.



8: R = H, R¹ = Gal
9: R = H, R¹ = Lac
10: R = R¹ = Gal
11: R = R¹ = Lac
12: R = Gal, R¹ = Lac

Chart 3. Mono- and bis-conjugated deprotected GAD.

Table 1. Dyeing conditions with GADs **8**, **10** and **11**

	Polyester	Wool
Temperature bath (°C)	98	98
Time (min)	30	20
Bath ratio[a]	50:1	50:1
Dye concentration (g/L)	1.0	1.0
Acetic acid added	1%	1%
Other	—	$(NH_4)_2SO_4$

[a] mL of water/fabric (g).

Table 2. Fastness test for wool dyed with GADs **8**, **10** and **11**[a]

GADs	Washing fastness	Rubbing fastness
8	4:5	4:5
10	5	4:5
11	5	5

[a] Determined according to international standard fastness methods: ISO 105-C06, 40 °C (washing); ISO 105-X12 (rubbing).

Fastness tests were determined according to the standard ISO methods (see Section 3) on wool and are consistent with a really good dyeing (Table 2), excluding the possibility of surface dyeing (lacquering). Of particular relevance is the fact that the washing fastness is not dependent on the size of the hydrophilic pocket, remaining good also in the case of the higher water soluble di-lactosyl GAD **11**. Furthermore, upon boiling in pyridine small samples of dyed wool for a couple of hours, UV–Vis analysis of the dye solutions did not indicate any change in the chemical structure during the dyeing processes.

It could be outlined that a homogeneous dyeing, although with different tonalities, was obtained with either fabric, as a nice demonstration of the multipurposity of this new type of dyes. As a matter of fact, this preliminary study evidences, however, that best results were obtained with mono- and digalactosyl GADs **8** and **10**, while with the dilactosyl conjugate **11**, the affinity decreases either with wool or with polyester. If one takes into account the differences in hydrophilicity of GADs, these results are expected for the hydrophobic fibre, but are more hardly explainable in the case of hydrophilic wool, indicating that more detailed and quantitative studies are requested for correlating the dyeing properties of GADs to their molecular structure. Our next researches will be directed in this direction and will be reported in due course.

3. Experimental

3.1. General methods

Melting points were determined with a Kofler hot-stage apparatus and were uncorrected. ^{1}H NMR spectra were recorder in appropriate solvents (internal standard Me_4Si) with a Varian Gemini instruments at 200 MHz and with a Bruker Avance II operating at 250 MHz. ^{13}C NMR spectra were recorded with the spectrometers operating at 50 and 62.9 MHz. Assignments were made with the aid of DEPT, HETCOR and COSY experiments and by comparison with values for known compounds and applying the known additivity rules.[9] In the case of anomeric mixtures, the assignments were made referring to the differences in the peak intensities. All reactions were followed by TLC on Kieselgel 60 F_{254} with detection by UV light and/or with ethanolic 10% phosphomolybdic or sulfuric acid, and heating. Kieselgel 60 (E. Merck, 70–230 and 230–400 mesh, respectively) was used for column and flash chromatography. Dyeing trials were performed on two kinds of fabric, always 9 × 4 cm sized. (1) a multiple witness (wool, nylon, cotton, acrylic, polyester and acetate) and/or (2) single. In this case, microfibre and ultramicrofibre polyester were also used, without any appreciable difference in the dyeing. These materials were purchased from Italian furnisher. The ISO methods were, respectively, the UNI EN ISO 105-C06 (for washing fastness) and UNI EN ISO 105-X12 for rubbing tests. In the rubbing tests, the fabrics were kept for at least 4 h in an atmosphere characterized by 20 °C and 65% moisture. The speed of rubbing was 1 cycle/s, with a linear motion for 20 times (ahead and back) using an apparatus as described in the Technical Manual of the American Association of Textile Chemists and Colorists, methods 8 and 165, respectively. Dry and wet fabrics were tested. Protected sugar monosuccinates (SMS) 6-*O*-(3-carboxypropanoyl)-1,2:3,4-di-*O*-isopropylidene-α-D-galactopyranose (**2a**) and 4-*O*-[2-*O*-acetyl-6-*O*-(3-carboxypropanoyl)-3,4-*O*-isopropylidene-β-D-galactopyranosyl]-2,3:5,6-di-*O*-isopropylidene-*aldehydo*-D-glucose dimethyl acetal (**2b**) were available from our previous research.[5]

3.2. 4-[*N*,*N*-Bis-(2-hydroxyethyl)amino]azobenzene (1)

The starting dye **1** was prepared on 10 g scale exactly as described by Freeman et al.,[7a] except the final step, where we used a flash chromatography (EtOAc) to discard a blackish material followed by crystallization from EtOH. Compound **1** was obtained in 75% yield as a yellow-orange solid, R_f 0.24 (EtOAc); mp 144–145 °C, (lit. mp 134–135 °C[7a,b]: mp 146 °C[7c]); λ_{max} 406 nm ($CHCl_3$) [lit. λ_{max} 413 nm (Me_2CO),[7a] λ_{max} 409 nm (EtOH)[7c]]; ^{1}H NMR[†] (CD_3OD, 250 MHz): δ 7.80 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.43 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.86 (m, 2H, Ar-H-3′, Ar-H-5′), 3.81, 3.63 (2t, each 4 H, J 5.0 Hz, 4 × CH_2); ^{13}C NMR[†] (CD_3OD, 62.9 MHz): δ 154.5 (Ar-C-1), 152.2 (Ar-C-4′), 144.7 (Ar-C-1′), 130.5 (Ar-C-4), 130.1, (Ar-C-3, Ar-C-5), 126.1 (Ar-C-2′, Ar-C-6′), 123.1 (Ar-

[†] The azobenzene system was numbered in this and in the following compounds, assigning number 1 to the quaternary carbons linked to the azo group and indicating with a prime those of the amino substituted ring.

C-2, Ar-C-6), 112.7 (Ar-C-3′, Ar-C-5′), 60.3 (2 × CH_2O), 54.9 (2 × CH_2N).

3.3. 4-{*N*,*N*-Bis[2-(1,2:3,4-di-*O*-isopropylidene-D-galactopyranos-6-yloxy)ethyl]amino}azobenzene (protected bis-conjugated GAD 3)

A soln of monosuccinate **2a**[5] (2.40 g, 6.66 mmol) and azo dye **1**[7] (0.97 g, 3.40 mmol) in dry THF (20 mL) was treated at room temperature with 1-ethyl-3-(3-(dimethylamino)propyl)carbodiimide hydrochloride (EDC, 1.28 g, 6.68 mmol) and 4-dimethylaminopyridine (DAMP, 244 mg, 2.00 mmol) and the soln was stirred at room temp until the starting material was completely disappeared (TLC, 12 h). The reaction mixture was concentrated under diminished pressure and the crude residue was dissolved into CH_2Cl_2 (100 mL) and washed in the order with satd aq $NaHCO_3$ (3 × 40 mL) and brine (40 mL). The organic phase was dried with Na_2SO_4, concentrated and the residue was purified by flash chromatography eluting with 3:1 hexane–EtOAc leading to the title compound **3** (2.91 g, 88% yield). **3** was an orange foam solid; mp 49–50 °C (chrom); R_f 0.68 (EtOAc); λ_{max} 403 nm ($CDCl_3$); ^{1}H NMR ($CDCl_3$, 250 MHz): δ 7.92 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.52 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.88 (m, 2H, Ar-H-3′, Ar-H-5′), 5.56 (d, 2H, $J_{1,2}$ 5.0 Hz, 2 × H-1), 4.66 (dd, 2H, $J_{2,3}$ 2.5 Hz, $J_{3,4}$ 7.8 Hz, 2 × H-3), 4.38 (m, 10H, 2 × CH_2O, 2 × H-2, 2 × H-6a, 2 × H-6b), 4.27 (dd, 2H, $J_{4,5}$ 1.8 Hz, 2 × H-4), 4.06 (ddd, 2H, $J_{5,6a}$ 7.1 Hz, $J_{5,6b}$ 5.0 Hz, 2 × H-5), 3.77 (t, 4H, J 6.2 Hz, 2 × CH_2N), 2.70 [m, 8H, 2×(CH_2)$_2$], 1.50, 1.43, 1.31, 1.30 [4s, each 6H, 4 × C(CH_3)$_2$]; ^{13}C NMR ($CDCl_3$, 62.9 MHz): δ 171.8 (4 × CO), 152.7 (Ar-C-1), 149.5 (Ar-C-4′), 143.8 (Ar-C-1′), 130.6, 129.3, 124.8, 122.0, 111.3 (Ar-CH), 109.3, 108.4 [4 × C(CH$_3$)$_2$], 95.9 (2 × C-1), 70.7 (2 × C-2), 70.3 (2 × C-3), 70.1 (2 × C-4), 65.6 (2 × C-5), 63.5 (2 × C-6), 60.1 (2 × CH_2O), 49.2 (2 × CH_2N), 28.6 [2×(CH$_2$)$_2$], 25.7, 25.6, 24.6, 24.2 [4 × C(CH$_3$)$_2$]. Anal. Calcd for $C_{48}H_{63}N_3O_{18}$: C, 59.43; H, 6.55; N, 4.33. Found: C, 59.41; H, 6.53; N, 4.30.

3.4. 4-{*N*,*N*-Bis[2-[2′-*O*-acetyl-2,3:5,6:3′,4′-tri-*O*-isopropylidene-1-dimethoxy-*aldehydo*-lactos-6′-yloxy]ethyl]-amino}azobenzene (protected bis-conjugated GAD 4)

A soln of monosuccinate **2b**[5] (2.28 g, 3.50 mmol) and azo dye **1** (0.50 g, 1.75 mmol) in dry THF (20 mL) was allowed to react in the conditions described for the preparation of **3**, leading, after flash chromatography (3:1 hexane–EtOAc) to **4** (2.38 g, 88% yield). Compound **4** was an orange solid foam; mp 56–57 °C (chrom); R_f 0.20 (2:3 petroleum ether–EtOAc); λ_{max} 403 nm ($CDCl_3$); ^{1}H NMR ($CDCl_3$, 200 MHz): δ 7.91 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.45 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.87 (m, 2H, Ar-H-3′, Ar-H-5′), 5.01 (m, 2H, 2 × H-2′), 4.77 (d, 2H, $J_{1',2'}$ 8.0 Hz, 2 × H-1′), 4.45 (dd, 2H, $J_{1,2}$ 5.8 Hz, $J_{2,3}$ 6.8 Hz, 2 × H-2), 4.40–4.20 (m, 10H), 4.19–4.05 (m, 8H), 4.00–3.85 (m, 8H), 3.76 (br t, 4H, 2 × CH_2N), 3.42, 3.41 (2s, each 6H, 2 × OCH_3), 2.64 [s, 8H, 2×(CH_2)$_2$], 2.13 (s, 6H, 2 × CH_3CO), 1.55, 1.47 [2s, each 6H, 2 × C(CH_3)$_2$], 1.37, 1.31, [2s, each 12H, 4 × C(CH_3)$_2$]; ^{13}C NMR ($CDCl_3$, 50 MHz): δ 172.0, 171.9 (4 × CO), 169.3 (2 × CH_3CO), 152.6 (Ar-C-1), 149.6 (Ar-C-4′), 144.0 (Ar-C-1′), 129.7, 128.9, 126.0, 122.0, 111.8 (Ar-CH), 110.8, 110.6, 107.9 [6 × C(CH$_3$)$_2$], 104.8 (2 × C-1), 100.1 (2 × C-1′), 78.1 (2 × C-3′), 78.0 (2 × C-5), 77.2 (2 × C-3), 75.1 (2 × C-2), 73.8 (2 × C-4), 73.5 (2 × C-4′), 72.6 (2 × C-2′), 70.6 (2 × C-5′), 64.6 (2 × C-6), 63.5 (2 × C-6′), 61.5 (2 × CH_2O), 55.6, 53.3 (4 × OCH_3), 49.3 (2 × CH_2N), 28.8, 28.5 [(CH$_2$)$_2$], 27.5, 27.4, 26.2, 26.0, 25.9, 24.5 [12 × C(CH$_3$)$_2$], 21.0 (2 × CH$_3$CO). Anal. Calcd for $C_{74}H_{107}N_3O_{32}$: C, 57.32; H, 6.96; N, 2.71. Found: C, 57.31; H, 7.12; N, 2.82.

3.5. 4-{*N*-(2-hydroxyethyl)-*N*-[2-(1,2:3,4-di-*O*-isopropylidene-D-galactopyranos-6-yloxy)ethyl]amino}azobenzene (protected mono-conjugated GAD 5)

A soln of dye **1** (2.90 g, 10.2 mmol) in dry THF (30 mL) was treated with 1-ethyl-3-[3-dimethylaminopropyl]-carbodiimide hydrochloride (EDC, 1.30 g, 6.79 mmol) and 4-dimethylaminopyridine (DMAP, 249 mg, 2.04 mmol). To the soln, cooled at 0 °C, was slowly added during 1 h, under stirring, a soln of **2a** (2.44 g, 6.79 mmol) in THF (20 mL). The reaction mixture was slowly warmed at room temp, stirred overnight and the solvents removed at diminished pressure. The crude residue was partitioned between CH_2Cl_2 (50 mL) and satd aq $NaHCO_3$ (3 × 30 mL), the organic phase washed with brine, dried (Na_2SO_4) and concentrated at diminished pressure. The crude residue (5.17 g) was purified through flash chromatography (3:1 hexane–EtOAc) to give, after a first crop of the disuccinate **3** (0.94 g), the title compound **5** (2.07 g) and, finally, unreacted compound **1** (1.26 g). The reaction yield based on the starting SMS-OH **2a** was of 49% and of 59%, if calculated on the amount of azo dye **1** corrected for the recovered material. Compound **5** was a yellow-orange solid foam; mp 46–48 °C (chrom); R_f 0.53 (EtOAc); λ_{max} 407 nm ($CDCl_3$); ^{1}H NMR ($CDCl_3$, 250 MHz,): δ 7.51 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.42 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.83 (m, 2H, Ar-H-3′, Ar-H-5′), 5.53 (d, 1H, $J_{1,2}$ 5.1 Hz, H-1), 4.62 (dd, 1H, $J_{2,3}$ 2.4 Hz, $J_{3,4}$ 7.8 Hz, H-3), 4.38 (t, 2H, J 5.9 Hz, CH_2O), 4.33 (dd, 1H, H-2), 4.22 (dd, 2H, $J_{4,5}$ 1.8 Hz, H-4), 4.19 (m, 2H, H-6a, H-6b), 4.01 (ddd, 2H, $J_{5,6a}$ 6.6 Hz, $J_{5,6b}$ 4.4 Hz, H-5), 3.88 (t, 2H, J 5.6 Hz, CH_2OH), 3.77 (t, 2H, J 5.9 Hz, OCH$_2$$CH_2$N), 3.66 (t, 2H, J 5.6 Hz, HOCH$_2$$CH_2$N), 2.65 [m, 4H, ($CH_2$)$_2$], 1.46, 1.41, 1.34, 1.33 [4s, each 3H, 2 × C(CH_3)$_2$]. ^{13}C

NMR ($CDCl_3$, 62.9 MHz): δ 172.1 (2 × CO), 152.9 (Ar-C-1), 150.2 (Ar-C-4′), 143.9 (Ar-C-1′), 129.5, 128.9, 124.9, 122.1, 111.7 (Ar-CH), 109.5, 108.7 [2 × $C(CH_3)_2$], 96.1 (C-1), 70.9 (C-2), 70.5 (C-4), 70.3 (C-3), 65.8 (C-5), 63.7 (C-6), 61.8 (CH_2O), 60.3 (CH_2OH), 53.6 (OCH_2CH_2N), 50.1 ($HOCH_2CH_2N$), 29.6, 28.1 [$(CH_2)_2$], 25.8, 25.7, 24.8, 24.3 [4 × $C(CH_3)_2$]. Anal. Calcd for $C_{32}H_{41}N_3O_{10}$: C, 61.23; H, 6.58; N, 6.69. Found: C, 62.20; H, 6.54; N, 6.67.

3.6. 4-{*N*-(2-hydroxyethyl)-*N*-[2-[2′-*O*-acetyl-2,3:5,6:3′,4′-tri-*O*-isopropylidene-1-dimethoxy-*aldehydo*-lactos-6′-yloxy]ethyl]amino}azobenzene (protected mono-conjugated GAD 6)

The condensation of **2b** (1.00 g, 1.54 mmol) and **1** (657 mg, 2.31 mmol) was performed according to the procedure described above for **5**. The flash chromatography of the crude reaction product (3:1 hexane–EtOAc) lead, after a first crop of the disuccinate **4** (240 mg), to the title compound **6** (696 mg) and, finally, to unreacted compound **1** (369 mg). The reaction yield based on the starting SMS-OH **2b** was 49% or 75% when calculated on reacted azo dye **1**. Compound **6** was an orange solid foam, mp 45–46 °C (chrom); R_f 0.13 (2:3 petroleum ether–EtOAc); λ_{max} 402 ($CHCl_3$); ^{1}H NMR ($CDCl_3$, 200 MHz): δ 7.86 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.41 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.84 (m, 2H, Ar-H-3′, Ar-H-5′), 5.00 (dd, 1H, $J_{1',2'}$ 8.4 Hz, $J_{2',3'}$ 6.8 Hz, H-2′), 4.76 (d, 1H, H-1′), 4.44 (dd, 1H, $J_{1,2}$ 6.2 Hz, $J_{2,3}$ 6.8 Hz, H-2), 4.39–4.20 (m, 6H), 4.17–4.00 (m, 4H), 3.97–3.87 (m, 4H), 3.80 (m, 2H, CH_2OH), 3.72, 3.60 (2m, each 2H, 2 × CH_2N), 3.38 (s, 6H, 2 × OCH_3), 2.59 [s, 4H, $(CH_2)_2$], 2.04 (s, 3H, CH_3CO), 1.52, 1.45, [2s, each 3H, $C(CH_3)_2$], 1.35, 1.29, [2s, each 6H, 2 × $C(CH_3)_2$]; ^{13}C NMR ($CDCl_3$, 50 MHz): δ 172.0, 171.8 (2 × CO), 169.4 (CH_3CO), 152.5 (Ar-C-1), 150.1 (Ar-C-4′), 143.6 (Ar-C-1′), 129.6, 128.9, 124.9, 122.2, 111.6 (Ar-CH), 110.6, 110.4, 107.7 [3 × $C(CH_3)_2$], 105.0 (C-1), 100.1 (C-1′), 78.1 (C-3′), 78.0 (C-5), 77.2 (C-3), 75.2 (C-2), 73.9 (C-4′), 73.5 (C-4), 72.6 (C-2′), 70.6 (C-5′), 64.6 (C-6), 63.5 (C-6′), 62.0 (CH_2O), 59.4 (CH_2OH), 55.6, 52.9 (2 × OCH_3), 53.3 (OCH_2CH_2N), 50.0 ($HOCH_2CH_2N$), 28.6, 28.5 [$(CH_2)_2$], 27.4, 27.3, 26.1, 26.0, 25.8, 24.4 [3 × $C(CH_3)_2$], 21.0 (CH_3CO). Anal. Calcd for $C_{45}H_{63}N_3O_{17}$: C, 58.88; H, 6.92; N, 4.58. Found: C, 58.87; H, 7.18; N, 4.40.

3.7. 4-{*N*-[2-[2′-*O*-acetyl-2,3:5,6:3′,4′-tri-*O*-isopropylidene-1-dimethoxy-*aldehydo*-lactos-6′-yloxy]ethyl]-*N*-[2-(1,2:3,4-di-*O*-isopropylidene-D-galactopyranos-6-yloxy)ethyl]amino}azobenzene (protected mixed bis-conjugated GAD 7)

The condensation of mono-conjugated GAD **5** (1.55 g, 2.47 mmol) with monosuccinate **2b** (1.93 g, 2.97 mmol) was carried out according to the procedure described above for the preparation of **3**. The flash chromatographic purification of the crude reaction product (7:13 hexane–EtOAc) gave **7** (1.83 g, 59% yield). **7** was an orange foam; R_f 0.66 (EtOAc); ^{1}H NMR[‡] ($CDCl_3$, 250 MHz): δ 7.86 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.41 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.85 (m, 2H, Ar-H-3′, Ar-H-5′), 5.51 (d, 1H, $J_{1'',2''}$ 5.0 Hz, H-1″), 5.00 (dd, 1H, $J_{1',2'}$ 8.3 Hz, $J_{2',3'}$ 7.6 Hz, H-2′), 4.77 (d, 1H, H-1′), 4.62 (dd, 1H, $J_{2'',3''}$ 2.3 Hz, $J_{3'',4''}$ 7.7 Hz, H-3″), 4.46 (dd, 1H, $J_{1,2}$ 6.2 Hz, $J_{2,3}$ 7.0 Hz, H-2), 4.41–4.18 (m, 12H), 4.17–3.92 (m, 8H), 3.72 (br t 4H, 2 × CH_2N), 3.42, 3.41 (2s, each 3H, 2 × OCH_3), 2.64 [m, 4H, $(CH_2)_2$], 2.15 (s, 3H, CH_3CO), 1.55–1.33 [m, 30H, 5 × $C(CH_3)_2$]; ^{13}C NMR ($CDCl_3$, 62.9 MHz): δ 172.0, 171.9 (4 × CO), 169.5 (CH_3CO), 152.9 (Ar-C-1), 149.6 (Ar-C-4′), 144.1 (Ar-C-1′), 129.66, 128.9, 125.0, 122.2, 111.5 (Ar-CH), 110.7, 110.6, 109.5, 108.7, 107.8 [5 × $C(CH_3)_2$], 104.9 (C-1), 100.0 (C-1′), 96.1 (C-1″), 78.0, 77.9, 77.1 (C-3, C-5, C-3′), 75.0 (C-2), 73.7, 73.4 (C-4, C-4′), 72.5 (C-2′), 70.9, 70.3 (C-2″, C-4″), 70.5 (C-3″, C-5′), 65.8 (C-5″), 64.5 (C-6), 63.7, 63.3 (C-6′, C-6″), 61.4, 61.3 (2 × CH_2O), 55.5, 53.0 (2 × OCH_3), 49.4 (2 × CH_2N), 28.8, 28.7, 28.6, 28.5 [2 × $(CH_2)_2$], 27.6, 27.5, 26.3, 26.1, 26.0, 25.9, 25.8, 24.9, 24.5, 24.4 [5 × $C(CH_3)_2$], 20.8 (CH_3CO). Anal. Calcd for $C_{61}H_{85}N_3O_{25}$: C, 58.13; H, 6.80; N, 3.33. Found: C, 58.10; H, 6.77; N, 3.30.

3.8. 4-{*N*-(2-hydroxyethyl)-*N*-[2-(D-galactopyranos-6-yloxy)ethyl]amino}azobenzene (deprotected GAD 8)

A soln of **5** (1.20 g, 1.90 mmol) in 90% aq CF_3COOH (18 mL) was stirred at room temp for 2 h and the red-violet reaction mixture was concentrated under diminished pressure and repeatedly co-evaporated with toluene (4 × 30 mL). NMR data for the aromatic portion of the acid residue are as follows: ^{1}H (CD_3OD, 250 MHz): δ 7.83 (m, 2H, Ar-H-2′, Ar-H-6′), 7.73 (m, 2H, Ar-H-2, Ar-H-6), 7.45 (m, 2H, Ar-H-3, Ar-H-5), 7.38 (m, 1H, H-4), 6.98 (m, 2H, Ar-H-3′, Ar-H-5′); ^{13}C (CD_3OD, 62.9 MHz): δ 155.7 (Ar-C-1), 148.9 (Ar-C-4′), 141.7 (Ar-C-1′), 130.4 (Ar-C-3, Ar-C-5), 130.2 (Ar-C-4), 129.1 (Ar-C-2′, Ar-C-6′), 121.5 (Ar-C-2, Ar-C-6), 115.0 (Ar-C-3′, Ar-C-5′). The crude residue was dissolved in EtOAc (80 mL) and neutralized with satd aq $NaHCO_3$ until the red-violet colour turned to yellow-orange, the aq phase was extracted with CH_2Cl_2 (3 × 30 mL) and the collected organic phases were dried (Na_2SO_4) and concentrated to give **8** (0.98 g, 94% yield) as an orange-yellow powder, R_f 0.16 (9:1 EtOAc–

[‡] For this compound and its deprotected derivative **12** (see below), the signals of the two different D-galactose units are indicated as follows: with a single prime the nonreducing unit of lactose, and with a double prime the monosaccharide one.

MeOH), constituted (NMR) by an anomeric mixture of α-pyranose, β-pyranose and β-furanose forms in ratio of 31:40:29. Selected ^{1}H NMR (Me_2SO, 250 MHz) signals for all anomers: δ 7.87 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.46 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.98 (m, 2H, Ar-H-3′, Ar-H-5′), 4.21 (m, 2H, CH_2O), 3.90–3.25 (m, 6H, CH_2OH, 2 × CH_2N), 2.59 [m, 4H, $(CH_2)_2$]; ^{13}C NMR (Me_2SO, 62.9 MHz): δ α-pyranose: 92.6 (C-1), 69.1 (C-3), 68.6 (C-2, C-4), 67.5 (C-5), 64.3 (C-6); β-pyranose: 97.4 (C-1), 73.2 (C-5), 71.8 (C-2, C-3), 68.6 (C-4), 64.4 (C-6); β-furanose: 101.8 (C-1), 82.5 (C-4), 81.4 (C-2), 75.9 (C-3), 67.5 (C-5), 65.8 (C-6); signals for all anomers: δ 171.9 (2 × CO), 152.4 (Ar-C-1), 150.3 (Ar-C-4′) 142.6 (Ar-C-1′), 129.6, 129.2, 124.9, 121.8, 111.4 (Ar-CH), 61.3 (CH_2O), 60.8 (CH_2OH), 52.9, 49.2 (2 × CH_2N), 28.6 [$(CH_2)_2$]. Anal. Calcd for $C_{26}H_{33}N_3O_{10}$: C, 57.03 H, 6.07; N, 7.67. Found: C, 56.98; H, 6.01; N, 7.64.

3.9. 4-{*N*-(2-hydroxyethyl)-*N*-[2-(2′-*O*-acetyl-1-dimethoxy-*aldehydo*-lactos-6′-yloxy)ethyl]amino} azobenzene (deprotected GAD 9)

A soln of **6** (981 mg, 1.07 mmol) was deprotected as reported for the preparation of **8**. After 45 min stirring, the red-violet soln was concentrated under diminished pressure the crude residue was dissolved in EtOAc (100 mL) and neutralized with satd aq $NaHCO_3$ until the red-violet colour turned to yellow-orange, the aq phase was extracted with EtOAc (3 × 100 mL) and the collected organic phases were dried (Na_2SO_4) and concentrated to give product **9** (780 mg, 97% yield) as a orange-yellow foam. R_f 0.21 (9:1 EtOAc–MeOH) constituted (NMR) by an anomeric mixture of α- and β-pyranose forms in ratio of 55:45. Selected ^{1}H NMR (CD_3OD, 250 MHz) signals for both anomers: δ 7.82 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′ α- and β-pyranose), 7.46 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5 α- and β-pyranose), 6.90 (m, 2H, Ar-H-3′, Ar-H-5′ α- and β-pyranose), 5.12 (d, 1H, $J_{1,2}$ 3.7 Hz, H-1 α-pyranose), 5.06 (m, 1H, H-2′ α- and β-pyranose), 4.50 (d, 1H, $J_{1',2'}$ 8.0 Hz, H-1 β-pyranose), 2.64 [m, 4H, $(CH_2)_2$], 2.09 (s, 3H, CH_3CO); ^{13}C NMR (CD_3OD, 62.9 MHz,): δ α-pyranose 102.7 (C-1′), 93.5 (C-1), 81.8 (C-4), 76.2 (C-5′), 73.5 (C-3′), 73.3 (C-3), 72.7 (C-2), 72.6 (C-2′), 71.1 (C-5), 70.0 (C-4′), 64.7 (C-6′), 63.0 (C-6), β-pyranose 102.7 (C-1′), 97.8 (C-1), 81.5 (C-4), 76.2 (C-5′), 75.8 (C-5), 74.2 (C-2, C-3), 72.6 (C-2′), 73.5 (C-3′), 70.0 (C-4′), 64.7 (C-6′), 63.0 (C-6), cluster of signals for both anomers: δ 174.2, 173.9 (2 × CO), 172.0 (CH_3CO), 154.3 (Ar-C-1), 152.0 (Ar-C-4′), 144.7 (Ar-C-1′), 130.8, 130.0, 126.1, 123.1, 112.7 (Ar-CH), 61.5 (CH_2O), 60.0 (CH_2OH), 54.1, 50.8 (2 × CH_2N), 29.9, 29.7 [$(CH_2)_2$], 21.1 (CH_3CO). Anal. Calcd for $C_{34}H_{45}N_3O_{16}$: C, 54.32 H, 6.03; N, 5.59. Found: C, 54.28; H, 6.06; N, 5.63.

3.10. 4-{*N*,*N*-Bis[2-(D-galactopyranos-6-yloxy)ethyl]-amino}azobenzene (deprotected GAD 10)

The hydrolysis of **3** (0.99 g, 1.00 mmol) was performed as described above for the preparation of **8**. After 2 h of stirring, the red-violet soln was concentrated at reduced pressure, neutralized and extracted as above described to give **10** (0.78 g, 95% yield) as an orange lacquer, R_f 0.18 (4:1 EtOAc–MeOH), constituted (NMR) by an anomeric mixture of α-pyranose, β-pyranose and β-furanose forms in ratio of 35:45:20. Selected ^{1}H NMR (Me_2SO, 250 MHz) signals for all anomers: δ 7.80 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.48 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.92 (m, 2H, Ar-H-3′, Ar-H-5′), 2.53 [m, 4H, $(CH_2)_2$]; ^{13}C NMR (Me_2SO, 62.9 MHz): δ α-pyranose δ 92.7 (2 × C-1), 69.3 (2 × C-4), 69.1 (2 × C-3), 68.5 (2 × C-2), 67.7 (2 × C-5), 63.4 (2 × C-6); β-pyranose 97.4 (2 × C-1), 73.1 (2 × C-5), 71.9 (2 × C-2, 2 × C-3), 68.7 (2 × C-4), 64.5 (2 × C-6); β-furanose 101.9 (2 × C-1), 82.6 (2 × C-4), 81.5 (2 × C-2), 76.0 (2 × C-3), 67.1 (2 × C-5), 65.9 (2 × C-6), cluster of signals for all anomers: δ 172.1 (4 × CO), 152.5 (Ar-C-1), 149.8 (Ar-C-4′) 143.0 (Ar-C-1′), 129.9, 129.3, 124.9. 121.9, 111.8 (Ar-CH), 61.5 (2 × CH_2O), 49.0 (2 × CH_2N), 28.6 [2 × $(CH_2)_2$]. Anal. Calcd for $C_{36}H_{47}N_3O_{18}$: C, 53.40; H, 5.85; N, 5.19. Found: C, 52.97; H, 5.81; N, 5.24.

3.11. 4-{*N*,*N*-Bis-[2-[2′-*O*-acetyl-1-dimethoxy-*aldehydo*-lactos-6′-yloxy]ethyl]amino}azobenzene (deprotected GAD 11)

The hydrolysis of **4** (1.09 g, 0.70 mmol) was performed as described above for the preparation of **8**. After 30 min of stirring, the red-violet soln was concentrated under diminished pressure and repeatedly co-evaporated with toluene (5 × 30 mL). The crude residue was dissolved in THF (30 mL), neutralized by addition of solid $NaHCO_3$ and stirred at room temp until the red-violet colour turned to yellow-orange (10 min). The suspensions were filtered, the residue washed with THF and the organic phase was concentrated under diminished pressure to give **10** (840 mg, 98% yield) as an orange solid constituted (NMR) by an anomeric mixture of α- and β-pyranose forms in ratio of 1:1. Selected ^{1}H NMR (CD_3OD, 250 MHz) signals for both anomers: δ 7.85 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′ α- and β-pyranose), 7.45 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5 α- and β-pyranose), 6.95 (m, 2H, Ar-H-3′, Ar-H-5′ α- and β-pyranose), 5.10 (d, 2H, $J_{1,2}$ 3.7 Hz, 2 × H-1 α-pyranose), 5.08 (m, 2H, 2 × H-2′ α- and β-pyranose), 4.48 (d, 2H, $J_{1',2'}$ 8.0 Hz, 2 × H-1 β-pyranose), 2.64 [m, 8H, 2 × $(CH_2)_2$], 2.11 (s, 6H, 2 × CH_3CO); ^{13}C NMR (CD_3OD, 62.9 MHz,): δ α-pyranose 102.5 (2 × C-1′), 93.3 (2 × C-1), 81.6 (2 × C-4), 76.0 (2 × C-5′), 73.7, 73.5 (2 × C-3′, 2 × C-3), 72.2, 72.5 (2 × C-2, 2 × C-2′),

70.8 (2 × C-5), 70.0 (2 × C-4′), 64.8 (2 × C-6′), 62.8 (2 × C-6), β-pyranose 102.5 (2 × C-1′), 97.8 (2 × C-1), 81.1 (2 × C-4), 75.9, 75.5 (2 × C-5′, 2 × C-5), 74.0 (2 × C-2, 2 × C-3), 73.1, 72.6 (2 × C-3′, 2 × C-2′), 70.0 (2 × C-4′), 64.8 (2 × C-6′), 62.8 (2 × C-6), cluster of signals for both anomers: δ 174.2, 174.0 (4 × CO), 172.1 (2 × CH_3CO), 154.3 (Ar-C-1), 151.8 (Ar-C-4′), 144.5 (Ar-C-1′), 130.7, 130.3, 126.2, 122.9, 112.5 (ArCH), 61.5 (2 × CH_2O), 51.0 (2 × CH_2N), 29.8, 29.7 [$(CH_2)_2$], 21.1 (2 × CH_3CO). Anal. Calcd for $C_{52}H_{71}N_3O_{30}$: C, 51.27; H, 5.87; N, 3.45. Found: C, 51.23; H, 5.82; N, 3.42.

3.12. 4-{*N*-[2-(2′-*O*-acetyl-1-dimethoxy-*aldehydo*-lactos-6′-yloxy)ethyl]-*N*-[2-(D-galactos-6-yloxy)ethyl]amino} azobenzene (deprotected GAD 12)

The deprotection of **7** (0.93 g, 0.74 mmol) was performed as described above for the preparation of **11**. After 2 h of stirring, the red-violet soln was concentrated under diminished pressure and repeatedly co-evaporated with toluene (7 × 70 mL) to give **10** as a reddish semisolid syrup that was dissolved in THF (30 mL) and neutralized by stirring with solid $NaHCO_3$ to give **12**, (0.76 g, 98% yield), R_f 0.18 (4:1 EtOAc–MeOH) constituted (NMR) by **12** as a complex mixture of anomeric forms of either the lactose reducing moiety or the galactose one. Selected ^{1}H NMR data (Me_2SO, 250 MHz) for all anomers: δ 7.76 (m, 4H, Ar-H-2, Ar-H-6, Ar-H-2′, Ar-H-6′), 7.45 (m, 3H, Ar-H-3, Ar-H-4, Ar-H-5), 6.98. (m, 2H, Ar-H-3′, Ar-H-5′), 2.53 [m, 8H, 2×$(CH_2)_2$], 2.10 (m, 3H, CH_3CO). Selected ^{13}C NMR data (Me_2SO, 62.9 MHz): δ 172.1 (4 × CO), 169.6 (CH_3CO), 152.5 (Ar-C-1), 150.5 (Ar-C-4′) 143.1 (Ar-C-1′), 129.9, 129.3, 125.0. 122.0, 111.8 (Ar-CH), 101.5, 101.9 (C-1′, α- and β-pyranose), 101.0 (C-1″, β-furanose), 97.4 (C-1″, β-pyranose), 96.7 (C-1, β-pyranose), 92.8 (C-1″, α-pyranose), 92.1 (C-1, α-pyranose), 82.7 (C-4″, β-furanose), 81.5 (C-2″, β-furanose), 80.9 (C-4, α- and β-pyranose), 61.5 (2 × CH_2O), 49.0 (2 × CH_2N), 28.6 [2 × $(CH_2)_2$], 20.9 (CH_3CO). Anal. Calcd for $C_{44}H_{59}N_3O_{24}$: C, 52.12; H, 5.87; N, 4.14. Found: C, 51.98; H, 5.78; N, 4.17.

Acknowledgements

This work was supported by Regione Toscana, Thecnological Innovation branch and Italvelluti SpA Company, Montemurlo (Prato) Italy.

References

1. Hofenk de Graaf, J. H. *The Colourful Past. Origins, Chemistry and Identification of Natural Dyestuffs*; Abegg-Stiftung (Riggisberg) and Archetype Publications: London, 2004.
2. Zollinger, H. *Color Chemistry*, 2nd ed.; VCH: Weinheim, 1991.
3. Poon, C. S.; Huang, Q.; Fung, P. C. *Chemosphere* **1999**, *38*, 1005–1014.
4. Bianchini, R.; Catelani, G.; Seu, G.; Bartalucci, G. Eur Patent 1462484 A1, 2004; *Chem. Abstr.* **2004**, *141*, 297362.
5. Bartalucci, G.; Bianchini, R.; Catelani, G.; D'Andrea, F.; Guazzelli, L. *Eur. J. Org. Chem.* **2007**, 588–595.
6. Bianchini, R.; Catelani, G.; Cecconi, R.; D'Andrea, F.; Guazzelli, L.; Isaad, J.; Rolla, M. *Eur. J. Org. Chem.* **2008**, 444–454.
7. (a) Freeman, H. S.; McIntosh, S. A. *Textile Res. J.* **1989**, *59*, 343–349; (b) Freeman, H. S.; Hao, Z.; McIntosh, S. A.; Posey, J. C. Jr.; Hsu, W.-N. *Dyes Pigments* **1990**, *12*, 233–242; (c) Podsiadły, R.; Sokołowska, J.; Marcinek, A.; Zielonka, J.; Chrześcijańska, E.; Socha, A. *Color. Thechnol* **2003**, *119*, 269–274.
8. Stoyanova, T.; Stoyanov, S.; Antonov, L.; Petrova, V. *Dyes Pigments* **1996**, *31*, 1–12.
9. Bock, K.; Pedersen, C. *Adv. Carbohydr. Chem. Biochem.* **1983**, *41*, 27–66.