Novel Glycomimetics: Anomeric and *N*-Glycosyl Sulfonamides

Pedro A. Colinas*

Laboratorio de Estudio de Compuestos Orgánicos (LADECOR), Departamento de Química, Facultad de Ciencias Exactas, Universidad Nacional de La Plata. 47 y 115 (1900), La Plata, Argentina

Abstract: Native glycosidic bonds in carbohydrates, are sensitive to the presence of enzymes. Thus, design of small molecule mimics (glycomimetics) is an active area of research. This review will focus on the development of a new class of glycomimetics: anomeric and N-glycosyl sulfonamides. These novel compounds have been demonstrated to be enzyme inhibitors or antiproliferative agents.

This work is dedicated to the memory of my mother, who passed away in July 20, 2011.

Keywords: Glycomimetics, Anomeric sulfonamides, N-glycosyl sulfonamides, Carbohydrates, Enzyme Inhibition.

INTRODUCTION

In many cases, use of carbohydrates as drugs has an important drawback: they are sensitive to the presence of enzymes and acidic or basic media. Thus, design of mimetics that bind to enzymes but are not processed to product in the usual way is an active area of research [1]. An unusual enzyme-resistant replacement for the glycosidic linkage, is the *sulfonamide* corresponding to the union of an anomeric sulfonic acid and an amine, or a glycosylamine carrying a sulfonyl group at nitrogen. These novel glycomimetics have been demonstrated to be enzyme inhibitors or antitumor agents. This review aims to outline the progress in the synthesis of anomeric and N-glycosyl sulfonamides, study of their conformational behaviour and biological activity [2].

A) ANOMERIC SULFONAMIDES

Anomeric sulfonamides is a class of glycosides which possess a sulfonamide moiety directly attached to the anomeric center of a carbohydrate. The synthesis of sulfonamides is tipically achieved by ammonolysis of the corresponding sulfonyl chlorides, which are generally prepared by chlorination of sulfonates. Several sulfonic acids and their salts attached to a sugar skeleton were described [3, 4]. The general method for obtaining such compounds was a nucleophilic displacement by thiolacetic acid or thiourea in the corresponding peracetylated sugar. Subsequent oxidation with dimethyldioxirane provided the anomeric sulfonic salts in good yields. Unfortunately treatment of anomeric sulfonates with PCl_5, $POCl_3$ or $SOCl_2$ gave several compounds [5, 6]. Knapp isolated the corresponding α-anomeric chlorides from the chlorynation mixtures [5]. It was proposed that the anomeric sulfonyl chlorides are unstable toward loss of SO_2, rapidly converting into the chlorides. For these reasons only recently suitable synthetic strategies has been developed to prepare these chemical entities.

First synthesis of S-glycosyl sulfonamides was described by Knapp *et al*. (Schemee **1**) [5]. Glucosyl thiazoline **2** has been prepared by reaction of peracetylated 2-acetamido-2-deoxy-β-D-glucopyranose **1** with Lawesson´s reagent [7]. Subsequent treatment with acidic conditions gave thioglycopyranose **3** in quantitative yield. Reaction with bromine and dietylamine afforded the corresponding glycosyl sulfenamide **4**. The last step involved oxidation with m-chlorperbenzoic acid to give the peracetylated N,N-diethyl-D-glucosylsulfonamide **5** in 56% yield over 4 steps.

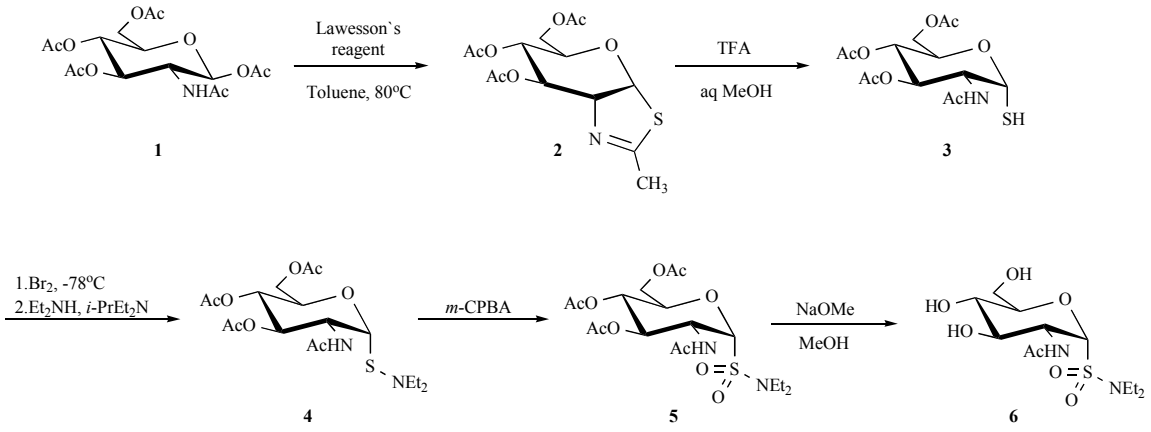
A small series of galactofuranosyl sulfonamides were prepared by oxidation of anomeric sulfenamides with the aim to develop new inhibitors of mycobacterial growth (Scheme **2**) [8]. One pot synthesis of glycosyl sulfenamides has been developed by reaction of peracetylated anomeric thiolacetates **8** with diethyl bromomalonate in the presence of a dialkyl amine. Subsequent oxidation with an excess of m-chloroperbenzoic acid afforded the protected galactofuranosyl sulfonamides **10**. Deprotection using sodium methoxide in methanol gave the target compounds in 75-92 % overall yield. Only *N*,*N*-dioctyl derivative showed antimycobacterial activity against *M. smegmatis* (ATCC 14468).

Recently Poulsen`s group has reported a related methodology for the synthesis of S-glycosyl sulfonamides throug oxidation of 2,4-dimethoxybenzyl protected sulfenamides (Scheme **3**) [9]. These compounds have been prepared by reaction of glycosyl thioacetates **12** with diethyl bromomalonate and 2,4-dimethoxybenzylamine. Subsequent oxidation with $KmnO_4/CuSO_4$ and remotion of the protecting group under acidic conditions afforded the per-O-acetylated glycosyl sulfonamides **16** (10-45% yields over three steps). Although other substituted benzylamines were used to prepared the intermediate sulfenamides, only 2,3-dimethoxybenzyl group could be easily removed to give the target compounds. The O-acetate groups of the carbohydrate moiety were removed using Zemplen`s conditions to afford the fully deprotected S-glycosyl sulfonamides **17** in good to high yields. This methodology was also applied to the synthesis of saccharides linked *via* a sulfonamide bridge [10]. Reaction of thioglycopyranoses **13** with different glycosyl amines afforded the corresponding sulfenamide disaccharides. Subsequent oxidation with *m*CPBA and deprotection with sodium methoxide gave the sulfonamide-bridged disaccharides. This was the first general methodology that allows the incorporation of the sulfonamide linker in the place of a native O-glycosidic bond.

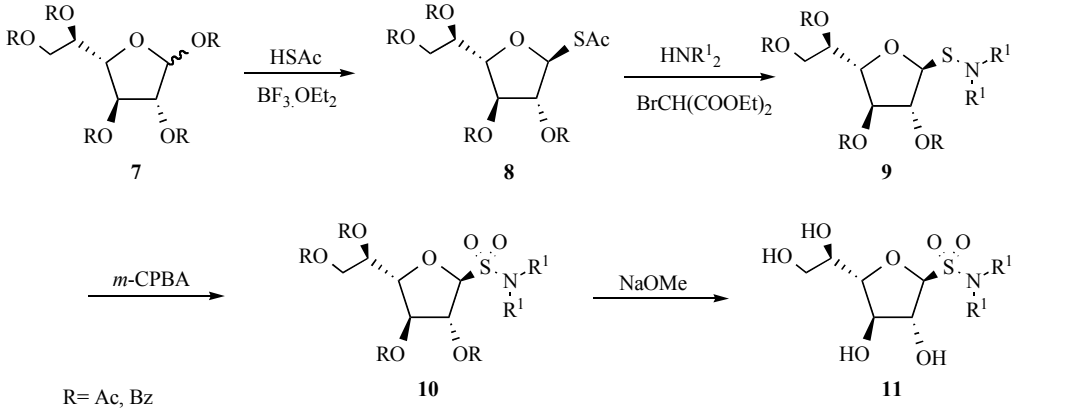
The protected **16** and deprotected anomeric sulfonamides **17** were tested as carbonic anhydrase inhibitors [11]. Recently this zinc metalloenzyme, which catalyzes the reversible hydration of cell-generated carbon dioxide into protons and bicarbonate ions, has emerged as a potential target in cancer therapy [12]. Mammalian cells express different carbonic anhydrase isozymes, which differ in

*Address correspondence to this author at the Laboratorio de Estudio de Compuestos Orgánicos (LADECOR), Departamento de Química, Facultad de Ciencias Exactas, Universidad Nacional de La Plata. 47 y 115 (1900), La Plata, Argentina; Tel: 54-221-424-3104; Fax: 54-221-422-6947; E-mail: pcolinas@quimica.unlp.edu.ar

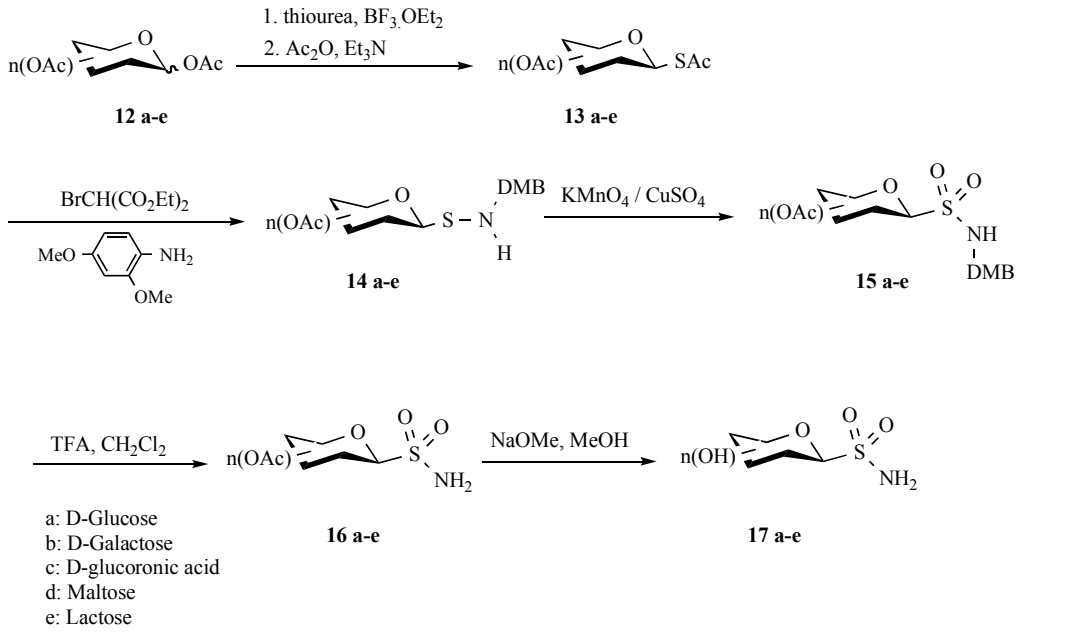
1385-2728/12 $58.00+.00



Scheme 1.



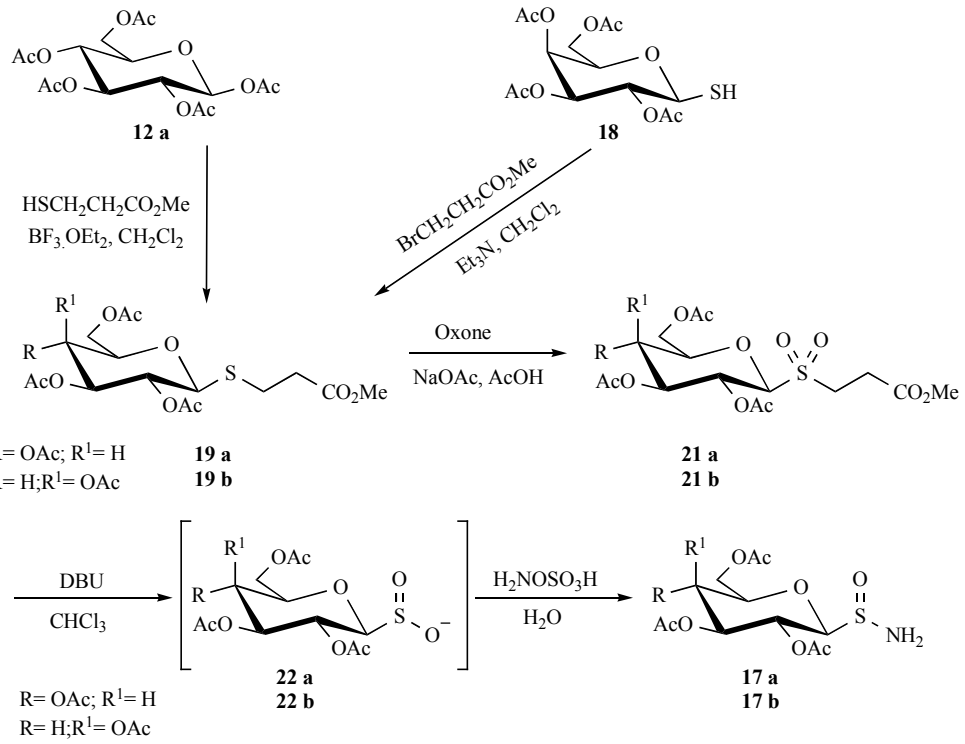
Scheme 2.



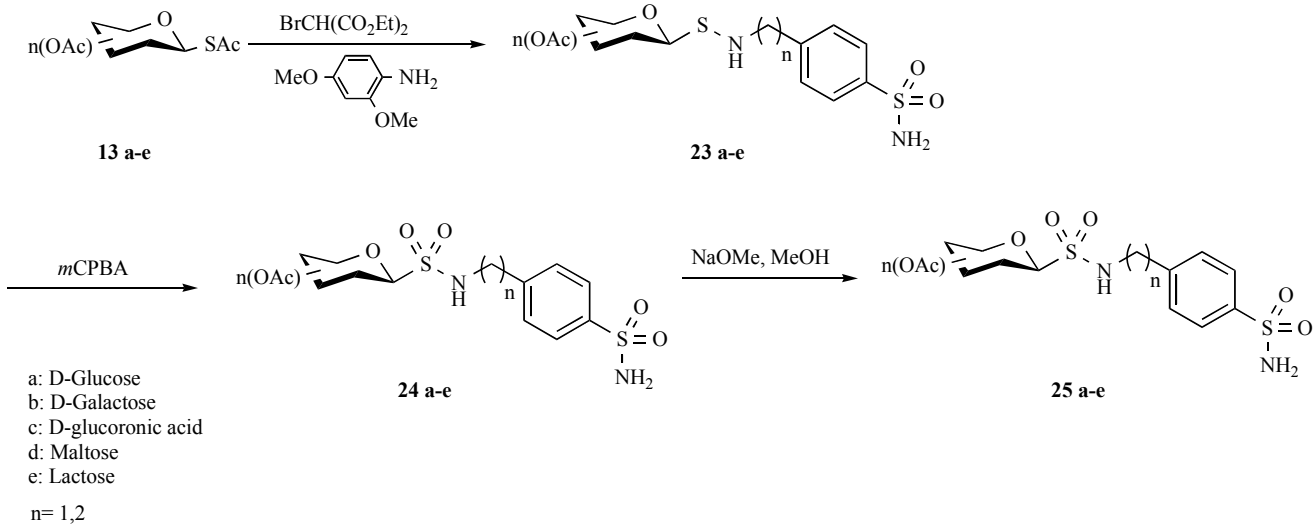
Scheme 3.

their tissue distribution and cellular localization [13]. Membrane-bound CA isozymes IX and XII are expressed at high levels and with a high prevalence in different tumor tissues, whose normal counterparts do not contain this protein [14].

The high catalytic activity of CA IX isozyme leading to formation of protons by the hydration of CO_2, was demonstrated to participate to the tumor microenvironment acidification by maintaining low extracellular acidity (pH_e) [12]. Overexpression of CA IX (or



Scheme 4.

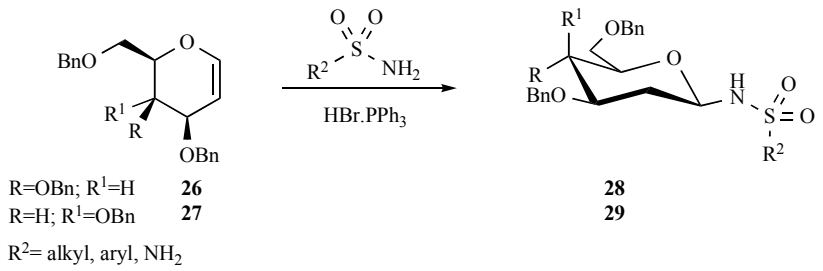


Scheme 5.

IX and XII) due to hypoxia has a strong impact on cancer progression, because maintenance of neutral intracellular pH is vital for cell proliferation and survival, whereas low pH_e contributes to aggressive tumor phenotype by promoting invasion and metastasis [12]. Supuran´s group showed that targeting of CA IX (and XII) with sulfonamide or coumarin potent and specific inhibitors, leads to effective inhibition of both primary tumor and metastases growth, and that this may provide a novel anticancer therapy [15].

The anomeric sulfonamides **16** and **17** were screened using the CO_2 hydration assay against the cytosolic hCA I and hCAII isozymes, as well as cancer-associated hCA IX and XII. Strikingly the sulfonamides showed no isozyme selectivity with inhibition constants in the micromolar range and neither the carbohydrate moiety nor the nature of the hydroxyl groups impacted to alter enzyme inhibition profile [11]. Using protein X-ray crystallography, Poulsen and co-workers demonstrated that the shape of the glycosyl sulfonamides resulted in weak interaction of the inhibitor with the enzyme active site. The sugar moieties of sulfonamides do not provide sufficient transverse bulk to span the active site cleft, which leads to a high degree flexibility of ligand conformations and to less fewer interactions of the inhibitor with the enzyme active site [11].

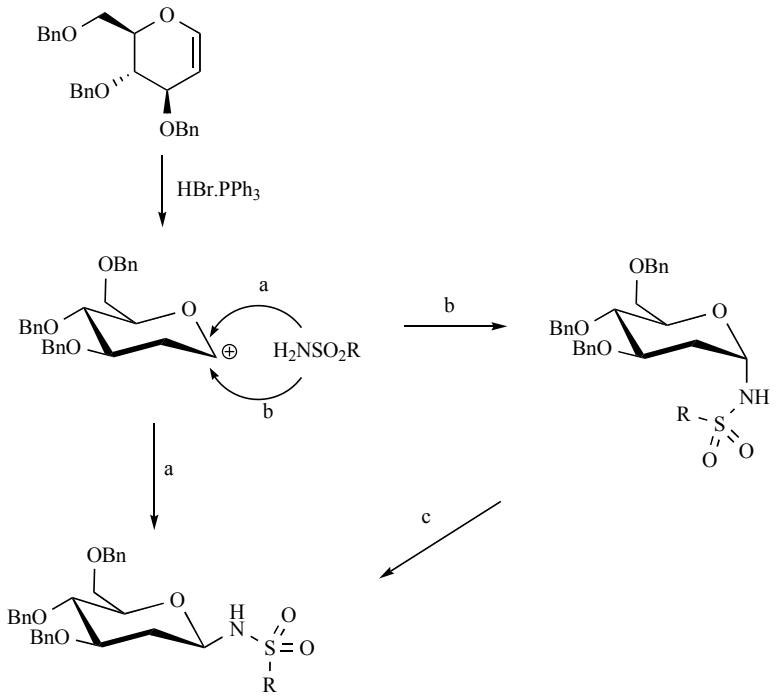
In the development of anti-cancer compounds that target selectively the membrane bound isoform CA IX versus the ubiquitous isoform CA II, the design of membrane non-permeant inhibitors is crucial. Poulsen`s group had calculated the lipophilicity and topological polar surface area for the S-glycosyl sulfonamides showing that all compounds fall within the range indicative of molecules with poor membrane permeability. The authors also measured ap-



R=OBn; R^1=H **26**
R=H; R^1=OBn **27**
R^2= alkyl, aryl, NH_2

28
29

Scheme 6.



Scheme 7.

parent *in vitro* effective permeability (Pe) using a parallel artificial membrane artificial assay (PAMPA) [16]. Although it was not possible to measure Pe of anomeric sulfonamides due to the analytical limits of the method, the results suggested that the compounds have poor passive membrane permeability. Though anomeric sulfonamides showed no selectivity for the cancer associated CAs, their pysicochemical properties would lead to preferential inhibition of the transmembrane CA IX over cytosolic CA II [11].

At the same time Somsák reported an alternate synthesis to S-glycosyl primary sulfonamides (Scheme **4**) [6]. The first step of the synthesis involved the reaction of peracetylated carbohydrate with methyl 3-sulfanylpropanoate in the presence of boron trifluoride etherate to give the thioglycosides in moderate yield. D-galacto derivative was obtained as α-anomer thus an alternative route to β-anomer was proposed. Reation of 1-thio-β-D-galactopyranose **18** with methyl 3-bromopropanate afforded the β-thiogalactoside **19 b** with excellent β-stereoselectivity and in good yield. Subsequent oxidation with Oxone and base-elimination of glycosyl sulfones afforded the S-glycosyl primary sulfinate salts **22**. The non-isolated sulfinates were treated with hydroxylamine O-sulfonic acid to afford the anomeric sulfonamides **17 a-b** in moderate overall yields (25% for D-glucosyl sulfonamide **17 a** and 67% for D-galactosyl compound **17 b**). Deprotected glucosylsulfonamide was tested against rabbit muscle GP*b*, a validated target for the treatment of type 2 diabetes mellitus, but showed to be inactive [6].

Very recently Poulsen´s methodology was employed in the synthesis of sulfonamide linked neoglycoconjugates (Scheme **5**) [17]. The first step involved the reaction of glycosyl thioacetates **13** with primary amines to give the sulfenamide glycoconjugates **23**. Oxidation of sulfenamides with *m*CPBA gave the per-O-acetylated sulfonamides **24** in good yields. Deprotection using Zemplen´s conditions afforded the target compounds **25**. The neoglycoconjugates were tested against carbonic anhydrase isozymes. Deprotected glycoconjugates **25** were shown to be slightly better CA II inhibitors than the acetylated ones. Glycoconjugates **24** and **25** were good CA IX inhibitors in the nanomolar range but the inhibition was weaker than for CA II [17].

B) N-GLYCOSYL SULFONAMIDES

i.) N-pyranosyl and N-glucosyl Sulfonamides

In 2003 we reported on the azaglycosylation of benzylated *endo*-glycals **26** and **27** using a catalytic amount of triphenylphosphine hydrobromide at room temperature (Scheme **6**) [18]. The reaction proceeded in a highly stereoselective fashion to give the β-

anomers in good to high yields. Also reaction proceeded well with sulfonamides with higher steric hindrance.

The high β-selectivity could be explained in terms of a thermodynamically controlled reaction. Petillo *et al.* reported that 2-β-iodo-1-α-sulfonamidohexoses readily epimerize at C-1 to α isomers in acidic media [19]. A similar epimerization of the kinetically formed α-sulfonamidoglycosides could be proposed for this reaction (Scheme **7**, path **c**). It was not possible to find any evidence of the α-glycosides in the crude reaction mixtures. Although it could be suggested that the proposed epimerization is very fast, another possibility is the attack of the anomeric positive charge by sulfonamide. Attack from the axial position would afford the α-isomer (Scheme **7**, path **b**) while attack from the equatorial position would give the β-isomer (Scheme **7**, path **a**). The β-anomer should be favored by steric effects in the axial isomer accentuated by an $n_N \rightarrow \sigma^*_{C\text{-}O}$ orbital interaction (*exo*-anomeric effect). An *anti* relationship between NH and anomeric hydrogen would permit an expression of the *exo*-anomeric interaction in the α-anomer. [20]. This conformation would necessarily point the hydrogen atom on nitrogen under the pyranose ring, giving an accentuated steric effect and thereby favoring the equatorial isomer (Fig. **1**).

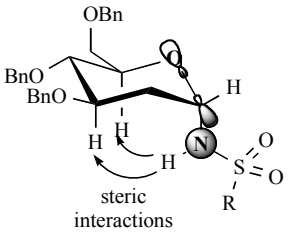


Fig. (1).

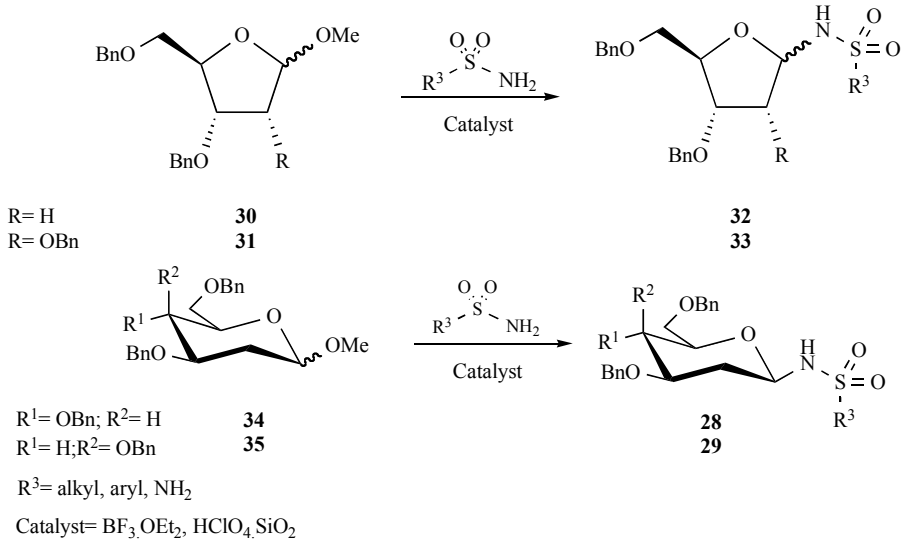
β-D-galactosyl *p*-toluenesulfonamide (**28** R^2=tolyl) and benzylsulfonamide (**28** R^2=CH_2Ph), prepared by this methodology, were tested as antiproliferative agents against human hepatocellular carcinoma cell line Hep-G2 and they showed to be potent inhibitors in the micromolar range [18].

Next we turned our attention to the preparation of sulfonamidoribofuranosides (Scheme **8**). Also several methodologies for glycals synthesis have been reported, many of these approaches involve several steps or fail in the furanoid glycal series [21]. For these reason other possibility was sought for the preparation of the target sulfonamides. The methyl glycosides are liable to generate an oxocarbenium ion under the catalyst of Lewis acids and subsequent addition of a sulfonamide to the ion, could generate the sulfonamidoglycosides. We found that sulfonamidoglycosylation of the methyl ribofuranosides **30** and **31** proceeded well in the presence of boron trifluoride etherate and molecular sieves 4 A [22]. The reaction showed no stereoselectivity as was previously found with other furanoses substrates. The methodology was also applied to methyl glycopyranoses **34** and **35**, which afforded the corresponding sulfonamidoglycosides **28** and **29** in very good yields and with excellent β-stereoselectivity.

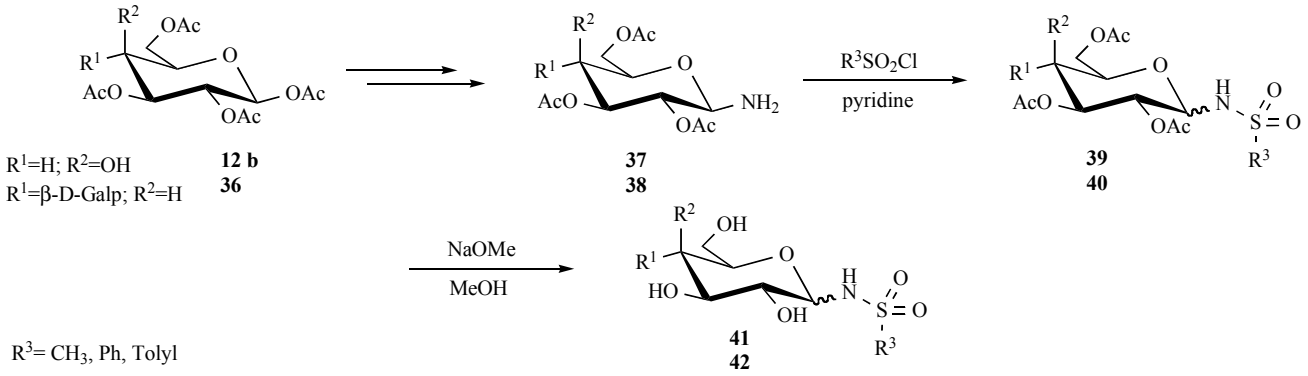
Then we decided to study the reaction of the methyl glycosides with sulfonamides in the presence of $HClO_4.SiO_2$ [23], which is an inexpensive, nontoxic, and recyclable catalyst for various organic transformations, affording the corresponding products in excellent yields with high selectivity [24]. The sulfonamidoglycosylations in the presence of this catalyst afforded the corresponding glycosyl sulfonamides in excellent yields. Although no influence of the promoter in the anomeric selectivity was found, the reaction times were reduced from 90 min with $BF_3.Et_2O$ to 5-10 min with $HClO_4.SiO_2$ and the workup merely required filtration of the catalyst [23].

Alves *et al.* reported the synthesis of *N*-D-galactosyl and *N*-lactosyl sulfonamides by treatment of the corresponding glycosyl amines with various sulfonyl chlorides (Scheme **8**) [25]. Sulfonamides **39** and **40** were obtained in poor to good yields but low stereoselectivity in almost all cases. Unfortunately anomeric ratios were not mentioned by the authors. *N*-β-glycosyl sulfonamides showed no anomerization in the same conditions employed in their synthesis. Thus it was concluded that α-anomers could arise from the anomerization of the glycosyl amines. Interaction of the sulfonamides **41** and **42** with two lectins, have been evaluated in a hemagglutination inhibitory activity assay. *N*-D-galactosyl sulfonamides **41** were less active than D-galactose. A quite similar activity was found in the lactose series [25].

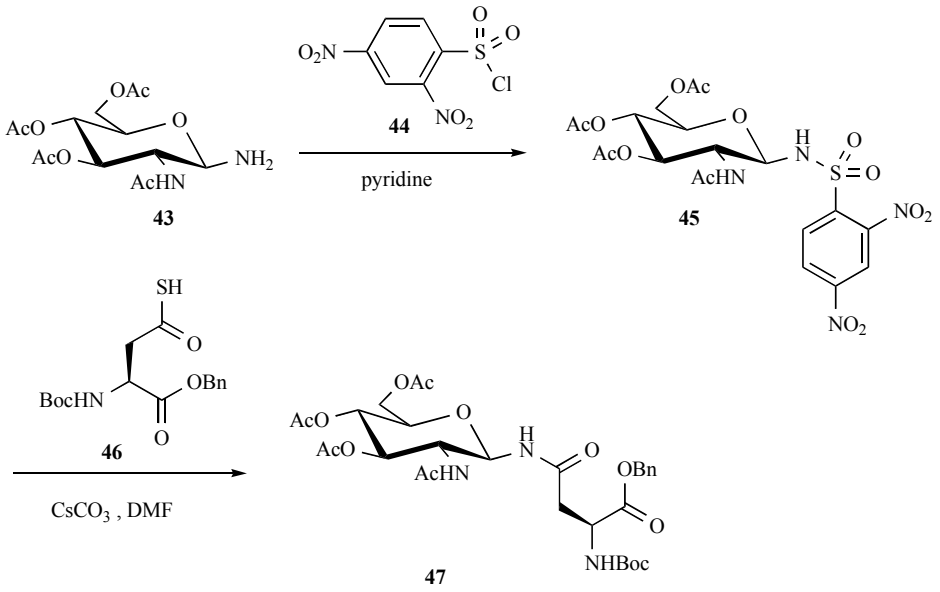
In a related methodology Sucheck´s group prepared a N-β-glucosyl sulfonamide as suitable intermediate in the synthesis of N-linked glycopeptides (Scheme **10**) [26]. N-acetyl-D-glucosamine **43** was treated with 2,4-dinitrobenzenesulfonyl chloride to give the



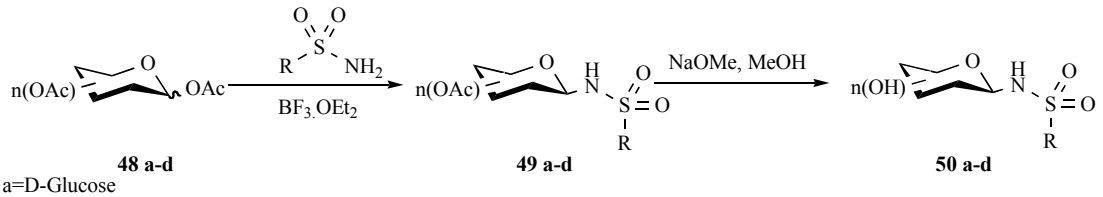
Scheme 8.



Scheme 9.



Scheme 10.

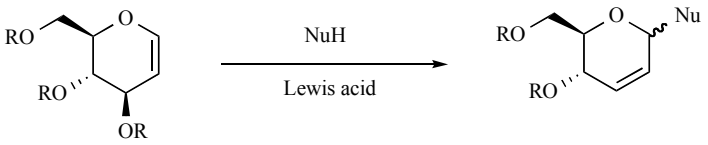


Scheme 11.

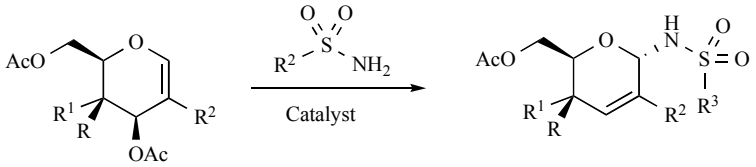
corresponding sulfonamide **45** in moderate yield. Subsequent treatment with thioacid **46** in the presence of cesium carbonate afforded the N-β-glucosylasparagine derivative **47** in good yield and excellent stereoseletivity.

As can be seen above glycosyl amines are prone to hydrolysis and anomerization thus their use in the synthesis is limited [27]. To overcome this problem we have developed the synthesis of N-glycosyl sulfonamides, by sulfonamidoglycosylation of peracetylated monosaccharides [28].

Per-O-acetylated pyranoses **48** derived from the monosaccharides D-glucose, D-galactose, D-mannose and L-rhamnose were reacted with boron trifluoride diethyl etherate and various sulfonamides to provide the β-sulfonamidoglycosides **49** in very good yields (Scheme **10**). Next Zemplen´s conditions were applied to afford the fully deprotected N-glycosyl sulfonamides **50** in nearly quantitative yields. To our surprise the sulfonamidoglycosylation of peracetylated D-mannose **48c** also afforded the corresponding β-mannosyl sulfonamides **49c** (R=CH_3, Tolyl, NH_2) [28]. The stereochemical outcome could be explained in terms of the *exo*-americ effect and steric interactions as was discussed above. The glycoysl sulfamides **49** and **50** (R=NH_2) prepared were tested as carbonic anhydrase inhibitors [29]. Per-O-acetylated compounds **49** were



Scheme 12.

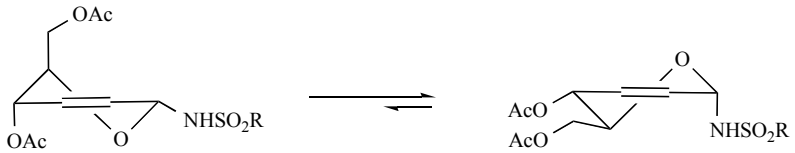


R=Ac; R^1=H	**60**	**63**
R=H; R^1=Ac	**61**	**64**
R^2= OAc, H	**62**	**65**

R^3= alkyl, aryl, NH_2

Catalyst: $(F_5C_6)_3B$; $BF_3.OEt_2$; Amberlyst 15; $ZnCl_2/Al_2O_3$

Scheme 13.



Scheme 14.

micromolar inhibitors of hCA I, while deprotected carbohydrate derivatives showed a diminished affinity. A similar pattern was found for the dominant isoform hCA II although acetylated glycosyl sulfamides **49** were quite effective inhibitors in the micromolar range. These compounds also were shown to be very good hCA IX inhibitors with inhibition constants clustered below 8 nM with very good selectivity over CA I and CA II. On the other hand the deacetylated sulfamideglycosides **50** were weaker inhibitors of CA IX in the micromolar range. Anyhow, deprotected sulfamides inhibited selectively this isoform over CA II. Calculated physicochemical properties of sulfamide glycosides (topological polar surface area and lipophilicity) showed that all compounds fall within the range indicative of molecules with poor membrane permeability and thus would lead to preferential preferential inhibition of CA IX over the ubiquitous cytosolic hCA II *in vivo* [29].

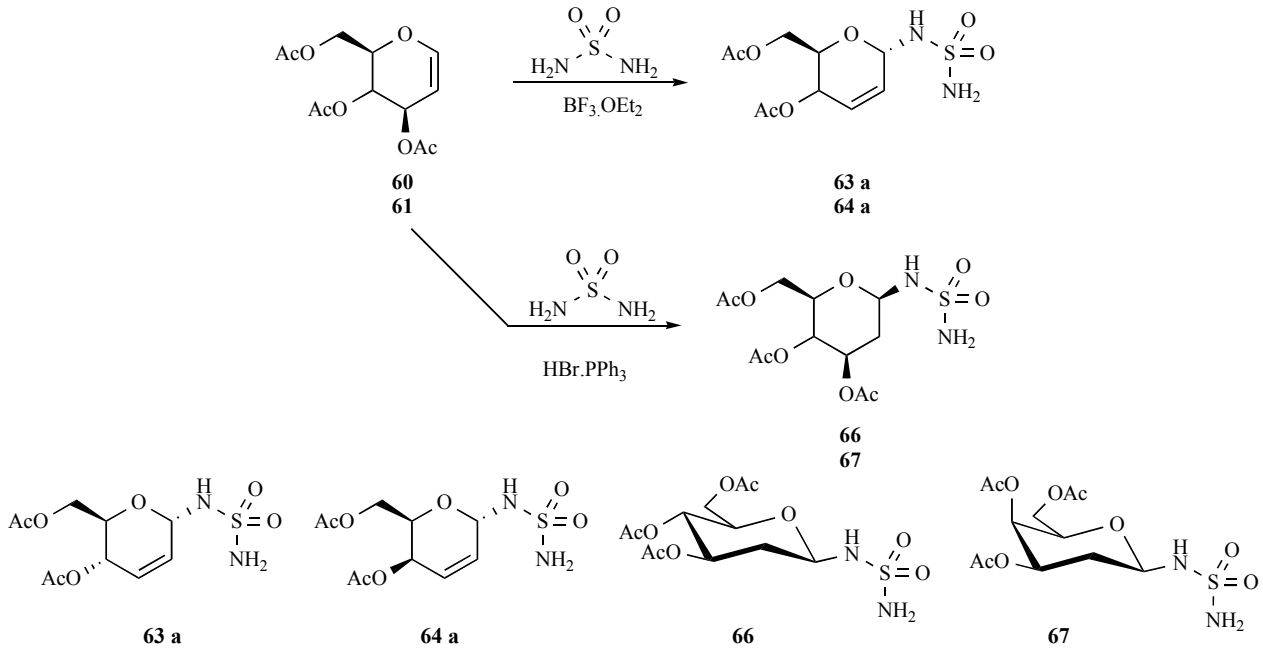
ii) N-(hex-2-enopyranosyl) Sulfonamides

In the presence of Lewis acids, D-glycals having leaving groups at the allylic site readily undergo nucleophilic displacement reaction with allylic rearrangement resulting in 2,3-unsaturated glycosides (Scheme **12**). This is commonly known as the Ferrier rearrangement [30].

In 2004 Chandrasekhar´s group developed the Ferrier azaglycosylation of peracetylated D-glucal **60** in the presence of tris(pentafluorophenyl)borane as Lewis acid catalyst (Scheme **13**)[31]. The D-hex-2-enopyranosylsulfonamides were obtained in good to high yields and with low α-selectivity. Also the azaglycosylation with N-substitued sulfonamides furnished the corresponding glycosylsulfonamides in good yields. The authors reported that reaction in the presence of boron trifluoride etherate gave a complex mixture of products. Surprisingly, in our hands, the sulfomidoglycosylation of per-O-acetylated D-glucal **60**, D-galactal **61** and 2-hydroxy-D-glucal **62** afforded the 2,3-unsaturated glycosylsulfonamides **63-65** in high yield with very good α-stereoselectivity [32].

The α anomers of the 2,3-enopyranosyl systems could be present in two equilibrium conformations ($^0H_5$ and $^5H_0$) (Scheme **14**). The values of $^3J_{3,4}$ 1.5–1.9 Hz (in $CDCl_3$) found in the *erythro* compounds **63** and $^3J_{3,4}$ 5.4–5.5 Hz in the *threo* compounds **64** indicate that the equilibrium between the two half-chair forms of the sulfonamidoglycosides lies significantly toward the $^0H_5$ conformation [32]. Also the conformation in the crystal state has been studied by X-ray diffraction of 4,6-di-*O*-acetyl-2,3-dideoxy-D-*erythro*-hex-2-enopyranosyl sulfamide **63 a** (R^3=NH_2) obtained from the sulfonamidoglycosylation of **60** with sulfamide [33]. Close inspection of the X-ray atomic coordinates showed that its conformation can be described mainly as as distorted $^0H_5$ half-chair in the crystal lattice.

Sulfonamidoglycosylation of D-glycals with sulfonamides was also studied from a theoretical point of view [34, 35]. Gas-phase results showed that the relative composition at room temperature is about 61% in the β anomeric form of 4,6-di-*O*-acetyl-2,3-dideoxy-D-*erythro*-hex-2-enopyranosyl sulfamide **63 a** (R^3=NH_2), and about 39% in the α form. When solvent effects were taken into account in the theoretical calculations, the relative composition at room temperature was reversed to about 5% in the β anomeric form of the glycosyl sulfamide and about 95% in the α form, in excellent agreement with experimental results [35]. These findings could indicate that the synthesis of the glycosyl sulfamide should occur mainly under thermodynamic control. As stated above, the β-selectivity found in the sulfonamidoglycosylation of *endo*-glycals in



Scheme 15.

the presence of triphenylphosphine hydrobromide could be explained in terms of interactions present in the axial isomer of glycopyranosylsulfonamides mainly due to the conformation of the anomeric nitrogen, thereby favoring the equatorial isomer, in which that interaction is absent. In the present 2,3-enopyranosyl α-isomer, on the other hand, the situation is quite different. Examination of dihedral angles indicates an almost planar conformation of C1-C2-C3-C4, thus the unfavorable steric interaction described previously is absent. Thus, the stereochemical outcome of Ferrier sulfonamidoglycosylation could be tentatively explained as a combination of several factors including the absence of steric interactions due to the conformation of the anomeric nitrogen in the α-isomer, which enables the exo-anomeric interaction and a $n_O \rightarrow \sigma^*_{C\text{-}N}$ orbital interaction (*endo*-anomeric effect) that it is only present in the α-isomer [35].

Althoug our methodology is useful for the synthesis of 2,3-unsaturated sulfonamideglycosides, it has an important drawback: boron trifluoride etherate is a highly toxic reagent. This prompted us to initiate studies designed to provide an environmentally friendlier route for the synthesis of sulfonamidoglycosides. Thus we investigated the use of ion exchange resin Amberlyst 15 as an alternative catalyst to prepare 2,3-unsaturated sulfonamideglycosides **63-64** [36]. It is well known that ion exchange resins are the most widely used heterogeneous catalysts due to their advantages such as high activity and selectivity, reusability, ease of separation, no corrosion, or disposal of effluent problems [37]. The sulfonamidoglycosylations proceeded well with the use of 30 wt % of the resin, a much lower amount of catalyst than the reported in the O- and S-glycosylations [38]. Although the yields and anomeric ratios were comparable to the homogeneous methodology, no aqueous workup was necessary. Recycled catalyst has been reused with no changes in the yields or in the anomeric selectivity [36].

At the same time Liu´s group developed a Ferrier sulfonamidoglycosylation of tri-*O*-acetyl-D-glucal **60** promoted by $ZnCl_2/Al_2O_3$ [39]. Reactions afforded the corresponding 2,3-enopyranosyl sulfonamides **63** in excellent yields (86-96%) with good α-selectivity. Workup is very simple in this methodology and promoter could be reused up to three times.

N-glycosyl sulfamides **63 a** and **64 a** synthetized by Ferrier sulfonamidoglycosylation of per-O-acetylated D-glycals **60** and **61** with sulfamide (Scheme **15**), were tested as carbonic anhydrase inhibitors [40]. β-N-2-deoxy-glycosyl sulfamides **66** and **67**, prepared by reaction of **60** and **61** in the presence of triphenylphosphine hydrobromide, were also tested to analyze the effect of the carbohydrate moiety in the inhibition. The glycosyl sulfamides were screened using the CO_2 hydration assay against the cytosolic hCA I and hCAII isozymes, as well as cancer-associated hCA IX and XII. The sulfamideglycosides were potent inhibitors of hCA II, IX and XII in the namolar range but weaker inhibitors of hCA I [39]. Erythro derivative **63 a** was a very effective hCA IX inhibitor and showed selectivity against hCA II. On the other hand, its *threo* epimer **64 a** showed no selectivity. It was explained in terms of negative interactions within the hCA II active site [40].

In the development of new chemotherapeutic agents, several sulfonamides have emerged as useful therapeutics for the treatment of cancer. E7010 [41], E7070 [41], ABT751 [42], and T138067 [43], have been found to be inhibitors of tumor cell proliferation, and some of them are under clinical evaluation. *N*-(2-(Cyclohexyloxy)-4-nitrophenyl)-methanesulfonamide (NS398) inhibits the growth of human hepatocellular carcinoma cell line HepG2 by inducing cells cycle arrest and is a potential candidate as an effective chemopreventive tool [44]. Celecoxib sulfonamide derivatives have shown to be highly toxic to human non-small-cell lung adenocarcinoma cells line A549 and the results suggest the potential of celecoxib-derived agents as chemotherapeutic drug [45]. Also sulfonamide containing compounds, such as N-pyridinyl- and indole-sulfonamides demonstrated effective inhibition of tubulin polymerization and were found to be potent antimitotic agents [46]. In view of these reports, the 2,3-enopyranosyl sulfonamidoglycosides **63-65** were evaluated for their cytotoxicity *in vitro* towards the human hepatocellular liver carcinoma cell line (HepG2) and human lung adenocarcinoma cell line (A549) [47]. These assays demonstrated that HepG2 cells, in general, are more sensitive

to the inhibition by the synthesized compounds. The two inhibitors of carbonic anhydrase **63 a** and **64 a** were also tested. However, despite the fact that, they inhibited CA IX and XII at low nanomolar concentrations [40], effects on cell proliferation were noted only at low milimolar concentrations for both cell lines. Absence of the effect of CA IX inhibition on cell growth in culture could be explained by the fact that both CA IX and XII grant the survival advantage to hypoxic tumor cells by regulating and maintaining pH. In the cell culture models used, cells grew in monolayer and have never become hypoxic. So inhibition of the mechanism that helps survive hypoxic conditions had no effect in cell cultures [48].

Other interesting feature is the clear activity dependency on the nature of the carbohydrate moiety present in the inhibitor. The *threo* compounds **63** were more active than *erythro* ones **64** against A549 cells. It was found that 2-acetyl-D-*erythro*-hex-2-enopyranosyl sulfonamides **65** were the most potent antiproliferative agents against both cell lines [47]. Thus results demonstrate that 2-acetyl group of the glycosyl ring plays a pivotal role in affecting cytotoxicity. Also it is important to note that alkyl sulfonamide derivatives, in general, were more potent inhibitors of tumor cells than their aryl sulfonamide and sulfamide analogs [47].

CONCLUSIONS

Several methods for the preparation of anomeric and N-glycosyl sulfonamides have been developed in the last ten years. This enzyme-resistant linkage replacement for the glycosidic linkage, proved to be very helpful in the design of compounds with biological activity, such as carbonic anhydrse inhibitors, antitumor and antibacterial agents. Also carbohydrate moieties were shown to impart unique properties to the known sulfonamido functionality. Some studies have been performed on the conformational behaviour of these compounds. It is of course desirable that our understanding of the interaction of glycosyl and anomeric sulfonamides with enzymes and other drug targets would also be improved, thereby leading to the development of more effective enzyme inhibitors and antiproliferative agents. We hope that this review will stimulate further advances in the synthesis of S- and N-glycosyl sulfonamides and promote further studies of their biological activity.

CONFLICT OF INTEREST

Declared none.

ACKNOWLEDGMENTS

The author thanks CONICET and UNLP for financial support. P.A.C is member of the Scientific Research Career of CONICET.

REFERENCES

[1] Ernst, B.; Magnani, J. L., From carbohydrate leads to glycomimetic drugs. *Nature Reviews Drug Discovery* **2009,** *8* (8), 661-677.

[2] This review will not discuss the Danishefsky´s work. His group reported on the reaction of glycals with iodonium disym-collidine perchlorate and benzenesulfonamide to afford stereoselectively 2-β-iodo-1-α-sulfonamidohexoses. This class of glycosylsulfonamides were used for the preparation of oligosaccharides with the 2-aminohexosesubunit.(For a review see: Danishefsky, S. J.; Bilodeau, M. T., Glycals in organic synthesis: The evolution of comprehensive strategies for the assembly of oligosaccharides and glycoconjugates of biological consequence. *Angewandte Chemie (International Edition in English)* **1996,** *35* (13-14), 1381-1419.). Also we will not consider the synthesis of glycosidic compounds in which the sulfonamide functionality is not directly linked to the carbohydrate moiety (For an excellent review see: Winum, J. Y.; Poulsen, S. A.; Supuran, C. T., Therapeutic applications of glycosidic carbonic anhydrase inhibitors. *Medicinal Research Reviews* **2009,** *29* (3), 419-435)

[3] Jakab, Z.; Fekete, A.; Borbás, A.; Lipták, A.; Antus, S., Synthesis of new sulfonic acid-containing oligosaccharide mimetics of sialyl Lewis A. *Tetrahedron* **2010,** *66* (13), 2404-2414.

[4] Lipták, A.; Balla, E.; Jánossy, L.; Sajtos, F.; Szilágyi, L., The first synthesis of secondary sugar sulfonic acids by nucleophilic displacement reactions. *Tetrahedron Letters* **2004,** *45* (4), 839-842.

[5] Knapp, S.; Darout, E.; Amorelli, B., New glycomimetics: Anomeric sulfonates, sulfenamides, and sulfonamides. *Journal of Organic Chemistry* **2006,** *71* (4), 1380-1389.

[6] Czifrák, K.; Somsák, L., Synthesis of anomeric sulfonamides and their behaviour under radical-mediated bromination conditions. *Carbohydrate Research* **2009,** *344* (3), 269-277.

[7] Knapp, S.; Vocadlo, D.; Gao, Z.; Kirk, B.; Lou, J.; Withers, S. G., NAG-thiazoline, an N-acetyl-β-hexosaminidase inhibitor that implicates acetamido participation. *Journal of the American Chemical Society* **1996,** *118* (28), 6804-6805.

[8] Owen, D. J.; Davis, C. B.; Hartnell, R. D.; Madge, P. D.; Thomson, R. J.; Chong, A. K. J.; Coppel, R. L.; Von Itzstein, M., Synthesis and evaluation of galactofuranosyl N,N-dialkyl sulfenamides and sulfonamides as antimycobacterial agents. *Bioorganic and Medicinal Chemistry Letters* **2007,** *17* (8), 2274-2277.

[9] a) Lopez, M.; Drillaud, N.; Bornaghi, L. F.; Poulsen, S. A., Synthesis of S-glycosyl primary sulfonamides. *Journal of Organic Chemistry* **2009,** *74* (7), 2811-2816.

[10] Lopez, M.; Bornaghi, L. F.; Innocenti, A.; Vullo, D.; Charman, S. A.; Supuran, C. T.; Poulsen, S. A., Sulfonamide linked neoglycoconjugates-A new class of inhibitors for cancer-associated carbonic anhydrases. *Journal of Medicinal Chemistry* **2010,** *53* (7), 2913-2926.

[11] Lopez, M.; Paul, B.; Hofmann, A.; Morizzi, J.; Wu, Q. K.; Charman, S. A.; Innocenti, A.; Vullo, D.; Supuran, C. T.; Poulsen, S. A., S-glycosyl primary sulfonamides - A new structural class for selective inhibition of cancer-associated carbonic anhydrases. *Journal of Medicinal Chemistry* **2009,** *52* (20), 6421-6432.

[12] a) Neri, D.; Supuran, C. T., Interfering with pH regulation in tumours as a therapeutic strategy. *Nature Reviews Drug Discovery* **2011,** *10* (10), 767-777. b) Winum, J. Y.; Scozzafava, A.; Montero, J. L.; Supuran, C. T., Inhibition of carbonic anhydrase IX: A new strategy against cancer. *Anti-Cancer Agents in Medicinal Chemistry* **2009,** *9* (6), 693-702.

[13] Supuran, C. T., Carbonic anhydrases: Novel therapeutic applications for inhibitors and activators. *Nature Reviews Drug Discovery* **2008,** *7* (2), 168-181.

[14] Pastoreková, S.; Barathova, M.; Kopacek, J.; Pastorek, J., Carbonic anhydrase inhibitors targeting cancer: Therapeutic, immunologic, and diagnostic tools targeting isoforms IX and XI. *Drug Design of Zinc-enzyme Inhibitors*; Supuran, C. T.; Winum, J.-Y., Ed.; Wiley: Hoboken **2009**; pp. 193-222.

[15] Lou, Y.; McDonald, P. C.; Oloumi, A.; Chia, S.; Ostlund, C.; Ahmadi, A.; Kyle, A.; Auf Dem Keller, U.; Leung, S.; Huntsman, D.; Clarke, B.; Sutherland, B. W.; Waterhouse, D.; Bally, M.; Roskelley, C.; Overall, C. M.; Minchinton, A.; Pacchiano, F.; Carta, F.; Scozzafava, A.; Touisni, N.; Winum, J. Y.; Supuran, C. T.; Dedhar, S., Targeting tumor hypoxia: Suppression of breast tumor growth and metastasis by novel carbonic anhydrase IX inhibitors. *Cancer Research* **2011,** *71* (9), 3364-3376.

[16] Kansy, M.; Senner, F.; Gubernator, K., Physicochemical high throughput screening: Parallel artificial membrane permeation assay in the description of passive absorption processes. *Journal of Medicinal Chemistry* **1998,** *41* (7), 1007-1010.

[17] Lopez, M.; Bornaghi, L. F.; Driguez, H.; Poulsen, S. A., Synthesis of sulfonamide-bridged glycomimetics. *Journal of Organic Chemistry* **2011,** *76* (9), 2965-2975.

[18] Colinas, P. A.; Bravo, R. D., A Novel Sulfonamidoglycosylation of Glycals. *Organic Letters* **2003,** *5* (23), 4509-4511.

[19] Owens, J. M.; Yeung, B. K. S.; Hill, D. C.; Petillo, P. A., Facile C1 epimerization of α-1-sulfonamidyl-2-deoxy-2-iodoglycopyranosides. *Journal of Organic Chemistry* **2001,** *66* (4), 1484-1486.

[20] Batchelor, R. J.; Green, D. F.; Johnston, B. D.; Patrick, B. O.; Pinto, B. M., Conformational preferences in glycosylamines. Implications for the exo-anomeric effect. *Carbohydrate Research* **2001,** *330* (3), 421-426.

[21] Bravo, F.; Kassou, M.; Díaz, Y.; Castillón, S., Synthesis of erythro and threo furanoid glycals from 1- and 2-phenylselenenyl-carbohydrate derivatives. *Carbohydrate Research* **2001,** *336* (2), 83-97.

[22] Colinas, P. A.; Bravo, R. D., Sulfonamidoglycosylation of methyl ribofuranosides: A novel approach to furanosylsulfonamides. *Tetrahedron Letters* **2005,** *46* (10), 1687-1689.

[23] Colinas, P. A.; Nuñez, N. A.; Bravo, R. D., Sulfonamidoglycosylation of methyl glycosides employing perchloric acid supported on silica gel. *Journal of Carbohydrate Chemistry* **2008,** *27* (3), 141-147.

[24] Dalpozzo, R.; Bartoli, G.; Sambri, L.; Melchiorre, P., Perchloric acid and its salts: Very powerful catalysts in organic chemistry. *Chemical Reviews* **2010,** *110* (6), 3501-3551.

[25] Butera, A. P.; De Souza Filho, J. D.; Carvalho, D. T.; Figueiredo, R. C.; De Faria, L. C. A.; Nunes, M. A.; Prado, M. A. F.; Alves, R. J.; De Andrade, M. H. G.; Silva, K. T. S., Synthesis of amides and sulfonamides of β-D-galactopyranosylamine and β-lactosylamine and evaluation of their interactions with the lectins from Erythrina cristagalli and Ricinus communis. *Quim. Nova* **2007,** *30* (5), 1267-1274.

[26] Talan, R. S.; Sanki, A. K.; Sucheck, S. J., Facile synthesis of N-glycosyl amides using a N-glycosyl-2,4-dinitrobenzenesulfonamide and thioacids. *Carbohydrate Research* **2009,** *344* (15), 2048-2050.

[27] Monsigny, M.; Quétard, C.; Bourgerie, S.; Delay, D.; Pichon, C.; Midoux, P.; Mayer, R.; Roche, A. C., Glycotargeting: The preparation of glyco-amino acids and derivatives from unprotected reducing sugars. *Biochimie* **1998,** *80* (2), 99-108.

[28] Colinas, P. A.; Témpera, C. A.; Rodriguez, O. M.; Bravo, R. D., Stereoselective synthesis of novel N-β-glycosyl sulfonamides by sulfonamidoglycosylation of per-O-acetylated sugars. *Synthesis* **2009,** (24), 4143-4148.

[29] Rodríguez, O. M.; Maresca, A.; Témpera, C. A.; Bravo, R. D.; Colinas, P. A.; Supuran, C. T., N-β-Glycosyl sulfamides are selective inhibitors of the cancer associated carbonic anhydrase isoforms IX and XII. *Bioorganic and Medicinal Chemistry Letters* **2011,** *21* (15), 4447-4450.

[30] Ferrier, R. J.; Zubkov, O. A. *Organic Reactions* **2003,** *62*, 569-736.

[31] Chandrasekhar, S.; Reddy, C. R.; Chandrashekar, G., Tris(pentafluorophenyl)borane catalyzed Ferrier azaglycosylation with sulfonamides and carbamates. *Tetrahedron Letters* **2004,** *45* (34), 6481-6484.

[32] Colinas, P. A.; Bravo, R. D., Ferrier sulfonamidoglycosylation of D-glycals. *Carbohydrate Research* **2007,** *342* (15), 2297-2302.

[33] Colinas, P. A.; Bravo, R. D.; Echeverría, G. A., X-ray crystallographic and high-resolution NMR spectroscopy characterization of 4,6-di-O-acetyl-2,3-dideoxy-α-d-erythro-hex-2-enopyranosyl sulfamide. *Carbohydrate Research* **2008,** *343* (17), 3005-3008.

[34] Alegre, M. L..; Diez, R. P.; Colinas, P. A., Experimental and theoretical study of the conformational, vibrational and magnetic properties of 4,6-di-O-acetyl-2,3-dideoxy-D-threo-hex-2-enopyranosyl ethanesulfonamide. *Journal of Molecular Structure* **2009,** *919* (1-3), 223-226.

[35] Lavecchia, M. J.; Diez, R. P.; Colinas, P. A., A combined theoretical and spectroscopic study of 4,6-di-O-acetyl-2,3-dideoxy-d-erythro-hex-2-enopyranosyl sulfamide: A novel glycosyl carbonic anhydrase IX inhibitor. *Carbohydrate Research* **2011,** *346* (3), 442-448.

[36] Témpera, C. A.; Colinas, P. A.; Bravo, R. D., Efficient synthesis of 2,3-unsaturated sulfonamidoglycosides by Amberlyst 15. *Tetrahedron Letters* **2010,** *51* (41), 5372-5374.

[37] De Angelis, A.; Ingallina, P.; Perego, C., Solid Acid Catalysts for Industrial Condensations of Ketones and Aldehydes with Aromatics. *Industrial and Engineering Chemistry Research* **2004,** *43* (5), 1169-1178.

[38] Tian, Q.; Zhu, X. M.; Yang, J. S., Amberlyst 15-catalyzed efficient synthesis of 2,3-unsaturated glycosides *via* Ferrier rearrangement for glycal. *Synthetic Communications* **2007,** *37* (5), 691-701.

[39] Ding, F.; William, R.; Gorityala, B. K.; Ma, J.; Wang, S.; Liu, X. W., A mild and efficient synthetic protocol for Ferrier azaglycosylation promoted by $ZnCl_2/Al_2O_3$. *Tetrahedron Letters* **2010,** *51* (23), 3146-3148.

[40] Colinas, P. A.; Bravo, R. D.; Vullo, D.; Scozzafava, A.; Supuran, C. T., Carbonic anhydrase inhibitors. Inhibition of cytosolic isoforms I and II, and extracellular isoforms IV, IX, and XII with sulfamides incorporating sugar moieties. *Bioorganic and Medicinal Chemistry Letters* **2007,** *17* (18), 5086-5090.

[41] McDonald, P. C.; Winum, J.-Y.; Supuran, C. T.; Dedhar, S. Recent developments in targeting carbonic anhydrase IX for cancer therapeutics. *Oncotarget* **2012**, 3(1), 84-97.

[42] Jordan, M. A.; Wilson, L., Microtubules as a target for anticancer drugs. *Nature Reviews Cancer* **2004,** *4* (4), 253-265.

[43] Shan, B.; Medina, J. C.; Santha, E.; Frankmoelle, W. P.; Chou, T. C.; Learned, R. M.; Narbut, M. R.; Stott, D.; Wu, P.; Jaen, J. C.; Rosen, T.; Timmermans, P. B. M. W. M.; Beckmann, H., Selective, covalent modification of β-tubulin residue Cys-239 by T138067, an antitumor agent with *in vivo* efficacy against multidrug-resistant tumors. *Proceedings of the National Academy of Sciences of the United States of America* **1999,** *96* (10), 5686-5691.

[44] Baek, J. Y.; Hur, W.; Wang, J. S.; Bae, S. H.; Yoon, S. K., Selective COX-2 inhibitor, NS-398, suppresses cellular proliferation in human hepatocellular carcinoma cell lines *via* cell cycle arrest. *World Journal of Gastroenterology* **2007,** *13* (8), 1175-1181.

[45] Tong, Z.; Wu, X.; Chen, C. S.; Kehrer, J. P., Cytotoxicity of a non-cyclooxygenase-2 inhibitory derivative of celecoxib in non-small-cell lung cancer A549 cells. *Lung Cancer* **2006,** *52* (1), 117-124.

[46] Chen, J.; Liu, T.; Dong, X.; Hu, Y., Recent development and SAR analysis of colchicine binding site inhibitors. *Mini-Reviews in Medicinal Chemistry* **2009,** *9* (10), 1174-1190.

[47] Crespo, R.; De Bravo, M. G.; Colinas, P. A.; Bravo, R. D., *In vitro* antitumor activity of N-glycosyl sulfonamides. *Bioorganic and Medicinal Chemistry Letters* **2010,** *20* (22), 6469-6471.

[48] Marques, S. M.; Enyedy, E. A.; Supuran, C. T.; Krupenko, N. I.; Krupenko, S. A.; Amélia Santos, M., Pteridine-sulfonamide conjugates as dual inhibitors of carbonic anhydrases and dihydrofolate reductase with potential antitumor activity. *Bioorganic and Medicinal Chemistry* **2010,** *18* (14), 5081-5089.

Received: March 01, 2012 Revised: April 26, 2012 Accepted: April 27, 2012