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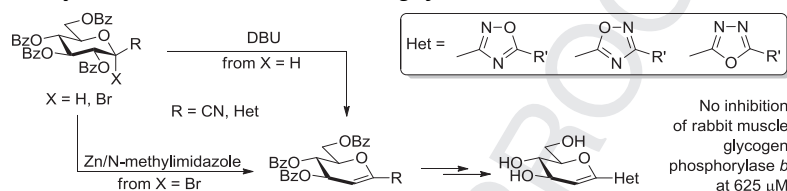
Carbohydrate Research

Graphical Abstract

C-(2-Deoxy-D-*arabino*-hex-1-enopyranosyl)-oxadiazoles: synthesis of possible isomers and their evaluation as glycogen phosphorylase inhibitors   pp. 1–9

Éva Bokor, Eszter Szennyes, Tibor Csupász, Nóra Tóth, Tibor Docsa, Pál Gergely, László Somsák*

Highlights

- Synthesis of D-glucals attached by a C-1—C bond to each isomeric oxadiazole.
- Formation of 1,2-double bonds by DBU induced elimination.
- Formation of 1,2-double bonds by Zn/N-methylimidazole mediated reductive elimination.
- No inhibition of rabbit muscle glycogen phosphorylase *b*.

C-(2-Deoxy-D-arabino-hex-1-enopyranosyl)-oxadiazoles: synthesis of possible isomers and their evaluation as glycogen phosphorylase inhibitors

Éva Bokor [a], Eszter Szennyes [a], Tibor Csupász [a], Nóra Tóth [a], Tibor Docsa [b], Pál Gergely [b], László Somsák [a,*]

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A B S T R A C T

Synthetic methods were elaborated for D-glucals attached to oxadiazoles by a C–C bond. Introduction of the double bond was effected by either DBU induced elimination of PhCOOH from the O-perbenzoylated glucopyranosyl precursors or Zn/N-methylimidazole mediated reductive elimination from the 1-bromoglucopyranosyl starting compounds. Alternatively, heterocyclizations of 2-deoxy-D-arabino-hex-1-enopyranosyl cyanide were also carried out. Test compounds were obtained by Zemplén debenzoylation, however, none of them showed significant inhibition of rabbit muscle glycogen phosphorylase b.

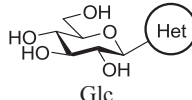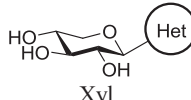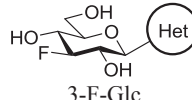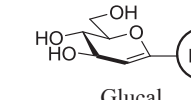© 2015 Elsevier Ltd. All rights reserved.

1. Introduction
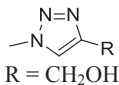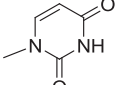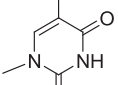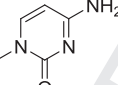
The continuous interest in glycogen phosphorylase inhibitors (GPIs) is primarily derived from the antihyperglycemic potential of such molecules involving their possible application in the medication of patients with type II diabetes.[1] On the other hand, inhibition of glycogen phosphorylase (GP) enzymes has also become an investigational approach in the context of other diseases such as ischemic lesions[2–5] and tumors.[6–10]

The inhibitors targeting the seven binding sites of GP (catalytic, inhibitor, allosteric, new allosteric, glycogen storage, benzimidazole[11] and the recently discovered quercetin binding site[12]) show a large molecular diversity.[13–17] Among them various glucose derivatives bind mostly to the catalytic site of the enzyme.[18,19] At present, N-acyl-β-D-glucopyranosylamines, N-acyl-N′-β-D-glucopyranosyl ureas, glucopyranosylidene-spiro-heterocycles as well as N- and C-glucosylated heterocycles (see Chart 1 for some important representatives of the latter e.g., 1–7, 13–20) belong to the most potent classes of this inhibitor family displaying their activity in or

below the low micromolar range against rabbit muscle GPb (RMGPb).[17–19] X-ray crystallographic studies on the binding modes of several of these molecules elucidated their increased binding strengths in comparison to that of D-glucose (K_i=1.7 and 7.4 mM for the α and β anomers,[20] respectively). Besides the ideal fit of the glucose part of these inhibitors in the active site, the strong binding must be ascribed to the H-bonding c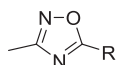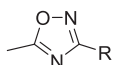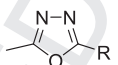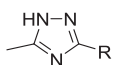apacities of the aglycons as well as van der Waals interactions of an aromatic appendage (if present) in the so-called β-channel of the enzyme.[13] These findings highlight the decisive contribution of the aglycon to the good inhibition and account for the fact that the structure-based inhibitor design of glucose analog GPIs has mainly been focused on the anomeric substitution patterns. Nevertheless, to get a thorough insight into the structure-activity relationships, the exploration of the specificity of the sugar unit is also necessary. Early investigations on the inhibitory and binding properties of different monosaccharides indicated the superior effectiveness of D-glucose.[21,22] Changes in the sugar configuration (e.g., for D-mannose[21] K_i>100 mM against RMGPb) as well as removal or replacement of substituents of the glucose moiety (e.g., for 2-deoxy-D-glucose[21] K_i=27 mM, for D-xylose[21] K_i=> 100 mM, for 3-deoxy-3-fluoro-D-glucose[22] K_i=200 mM against RMGPb) proved

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Sugar part	N-Glycosyl heterocycles				
	triazole, R = CH$_2$OH	uracil	X-uracil	NH$_2$	NHCOPh
Glc	**1** 26[36] 14[37]	**2** 6.1[15] 12[38]	**3** X = F 5.5[15] 7.9[38], **4** X = Cl 1.0[38], **5** X = Br 3.3[38], **6** X = I 1.9[38]	**7** 7.7[15]	-
Xyl	**8** No inh. at 625 μM[28]	-	-	-	-
3-F-Glc	-	**9** 3460[29]	**10** X = F 3670[29]	**11** 4010[29]	**12** 46[29]
	C-Glycosyl heterocycles				
	N–O oxadiazole	O–N oxadiazole	N–N oxadiazole	HN–N triazole	benzimidazole
Glc (R = Me)	**13** No inh. at 625 μM[39]	-	**16** 212[40] 145[41]	**18** 499[42]	**20** 11[40] 8.6[41]
Glc (R = 2-Naphthyl)	**14** 38[43]	**15** 12 (2.4)[39]	**17** 10 % inh. at 625 μM[39]	**19** 0.41[42,44]	
Xyl	-	-	-	**21** 491[24]	**22** No inh. at 625 μM[24]
Glucal	**Target compounds of this study**			-	-

Chart 1. Inhibitory potency (K_i [μM]) of selected N- and C-glycosyl heterocycles against rabbit muscle glycogen phosphorylase b (RMGPb).[36, 37, 38, 42, 44]   Q1

detrimental for the inhibition. Taking into account the crucial role of the aglycon in the efficiency of the glucose based inhibitors alterations of the sugar moiety in cases of some potent heterocyclic glucose derivatives were also examined to test whether the interactions of the anomeric substituent could compensate the impaired binding affinity of the glycon.

As part of this program D-xylose derived analogs of the best glucose based inhibitors were most often studied. Xylopyranosylidene-spiro-hydantoins, the first compounds investigated in this series, proved practically inactive.[23] Xylopyranosylidene-spiro-isoxazolines and oxathiazoles,[24] having the most potent aglycones of the glucopyranosyl series,[25–27] remained also ineffective.[24] Xylopyranosyl counterparts of N-(β-D-glucopyranosyl)-1,2,3-triazoles (e.g., **8** in Chart 1) as well as analogous derivatives of 5-thio-xylose and their oxidized variants (sulfoxides and sulfones) showed also negligible or no inhibition against RMGPb.[28] Very recently, C-β-D-xylopyranosyl-heterocycles were synthesized (e.g., **21** and **22**), and among them only the 2-naphthyl substituted 1,2,4-triazole derivative **21** had modest activity towards RMGPb.[24]

Other studies with N-β-D-glucopyranosyl-pyrimidines[15,17] **2**–**7** and **9**–**12** showed that replacement of the 3-OH group of the glucose moiety by fluorine caused very significant weakening of the inhibitions (see Chart 1 for the directly comparable pairs **2** and **9**, **3** and **10**, and **7** and **11**, respectively).[29] Nevertheless, elongation of the aglycon by a hydrophobic group proved advantageous (compare **11** and **12**) rendering compound **12** to be the first micromolar inhibitor of this class.[29] Furthermore, insertion of an axially oriented hydroxymethyl group into the C-3 position of the glucose part of **2** induced a slightly decreased inhibition (K_i=27.1 μM against RMGPb) in spite of additional molecular interactions of the −CH$_2$OH group that was evidenced by X-ray crystallography.[30]

Additionally, in the frame of a study of conformationally restricted pseudonucleosides, an N-substituted spiro-thiohydantoin ring was constructed at the C-3 position of D-glucose, however, such compounds remained inactive against RMGPb.[31] Glucofuranosylidene-spiro-hydantoins[32] as well as some iminosugar derivatives were also studied to show no significant activity except 1,4-dideoxy-1,4-imino-arabinitol (DAB).[33]

The phosphorolytic cleavage of glycogen catalyzed by GP is supposed to occur via a glycosyliumion-like transition sate.[34] Thus, it can be expected that compounds known to be mimics of this intermediate can bind to the active site of the enzyme as evidenced
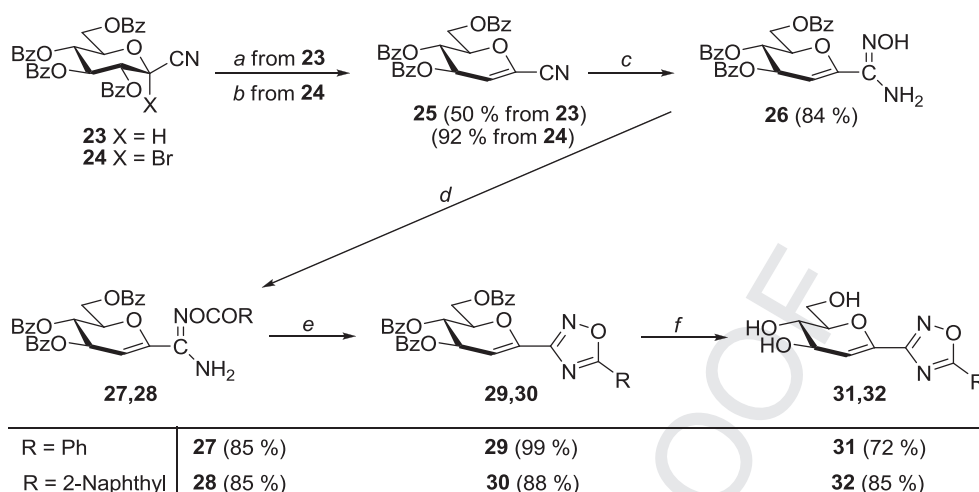
Scheme 1. Reagents and conditions: a) DBU, dry CH$_2$Cl$_2$, rt; b) Zn, N-methylimidazole, dry EtOAc, reflux; c) NH$_2$OH·HCl, dry pyridine, rt; d) RCOCl, dry 1,4-dioxane, rt; e) 1 M TBAF in THF, toluene, reflux; f) cat. NaOMe in dry MeOH, rt.

by X-ray crystallography for some iminosugar type GP inhibitors.[33] Glycals can also be considered as oxocarbenium ion analogs due to the resemblance of the shape of these molecules (half-chair conformation) to that of the glycosyl cation, as it was demonstrated earlier for glycosidase inhibitors.[35] Based on these considerations we set out to prepare 1-C-hetaryl-glucal derivatives to study their inhibitory potential against GP. Although D-glucal itself is a weak inhibitor of RMGPb (K_i=80 mM),[21] it can be assumed that attaching a heterocycle to the C-1 carbon atom of the glucal may result in favorable interactions with the protein. In this paper we disclose our first steps towards this type of inhibitors and report the syntheses and enzymatic evaluation of each possible isomer of oxadiazoles appended to D-glucal by a C–C bond.

2. Results and discussion

2.1. Syntheses

For the preparation of the target compounds two main routes can be envisaged: a suitably functionalized glucal can be made first followed by the formation of the heterocycle in the final stage or, alternatively, the 1,2-double bond can be introduced into a pre-formed C-glucopyranosyl heterocycle. The rather scarce literature of 1-C-hetaryl pyranoid glycals[45–48] offers possibilities for both strategies and neither of them seems superior.[47] Since the 2,3,4,6-tetra-O-benzoyl-β-D-glucopyranosyl cyanide[49] **23** was shown to be a common precursor toward each isomer of C-glucopyranosyl-oxadiazoles[40,39,43,50] synthesis and transformations of its unsaturated counterpart **25** (Scheme 1) were studied first.

Based on literature analogies[46,47,51,52] DBU induced benzoic acid elimination from **23** as well as Zn/N-methylimidazole mediated reductive elimination[35,53] of the 2,3,4,6-tetra-O-benzoyl-1-bromo-β-D-glucopyranosyl cyanide[49] (**24**) were probed to get the 1-cyano-glucal **25** (Scheme 1). The latter method proved to be more efficient both in terms of purity of the product and overall yields (compare yields under conditions a and b in Scheme 1 taking into account that **24** can be obtained almost quantitatively[49]). By adaptation of a literature method[43] **25** was then transformed into amidoxime **26**, which was acylated by acid chlorides to give compounds **27** and **28** in high yields. Subsequent TBAF promoted ring closure[54] gave oxadiazoles **29** and **30**, which were debenzoylated by the Zemplén protocol to furnish 5-aryl-3-(2′-deoxy-D-arabino-hex-1′-enopyr-anosyl)-1,2,4-oxadiazoles **31** and **32**, respectively, in good yields.

For the preparation of the constitutionally reversed O-perben-zoylated 3-aryl-5-(2′-deoxy-D-arabino-hex-1′-enopyranosyl)-1,2,4-oxadiazoles (**35** and **36**) both main strategies were investigated (Scheme 2). 1,3-Dipolar cycloaddition of in situ generated nitrile oxides[39,50] to the O-perbenzoylated 1-cyano-glucal **25** took place chemoselectively to give unsaturated oxadiazoles **35** and **36**. Additionally, DBU induced β-elimination of PhCOOH from the appropriate O-perbenzoylated 3-aryl-5-β-D-glucopyranosyl-1,2,4-oxadiazoles[39,50] **33** and **34** was also performed. A comparison of the yields for **35** and **36** from **25** on route a and from **33** and **34** on route b, respectively, showed the latter method to be superior. Deprotection of **35** and **36** was carried out by the Zemplén method to yield **37** and **38**, which proved identical with C-glucosyl-1,2,4-oxadiazoles isolated as by-products in the base-catalyzed trans-esterification of compounds **33** and **34**, respectively.[39]

For the formation of 2-(2′-deoxy-D-arabino-hex-1′-enopyr-anosyl)-5-substituted-1,3,4-oxadiazoles (**48**–**50**) the easily avail-able O-perbenzoylated 2-(β-D-glucopyranosyl)-5-substituted-1,3,4-oxadiazoles[40,39] (**39**–**41**) were used as starting materials (Scheme 3). Contrary to the elimination of PhCOOH from 3-aryl-1,2,4-oxadiazole derivatives (**33** and **34** in Scheme 2, route b) introduction of the double bond into the sugar moiety of **39**–**41** by using DBU failed (no or low conversions were observed at either rt. or reflux temperature in CH$_2$Cl$_2$, CHCl$_3$ and toluene). We speculate that this might be due to different acidity of the C-1-H bonds, however, no further investigations were devoted to verify this point. Thereafter, the bromination-reductive elimination sequence was followed to obtain the unsaturated derivatives **45**–**47**. Bromi-nation of **40** and **41** was smoothly accomplished to give **43** and **44**, respectively, by using Br$_2$ under irradiation by a heat lamp.[55] Taking into account the susceptibility of the methyl group to be bromi-nated under the above radical bromination conditions[56] trans-formation of **39** into **42** was achieved by using the KBrO$_3$–Na$_2$S$_2$O$_4$/CH$_2$Cl$_2$–H$_2$O reagent-solvent system[57] where the methyl group remained intact. The brominated compounds **42**–**44** were then subjected to reductive elimination by Zn/N-methylimidazole to obtain **45**–**47**, respectively, in good yields. Removal of the benzoyl protecting groups by the Zemplén method furnished the final products **48**–**50** in high yields.

Structural elucidation of the new compounds was based on proton and carbon NMR data. The ^{1}H NMR spectra for the O-per-benzoylated oxadiazoles **29**, **30**, **35**, **36**, **45**–**47** displayed narrow doublets in the range of 6.2–6.6 ppm for the olefinic H-2′ protons
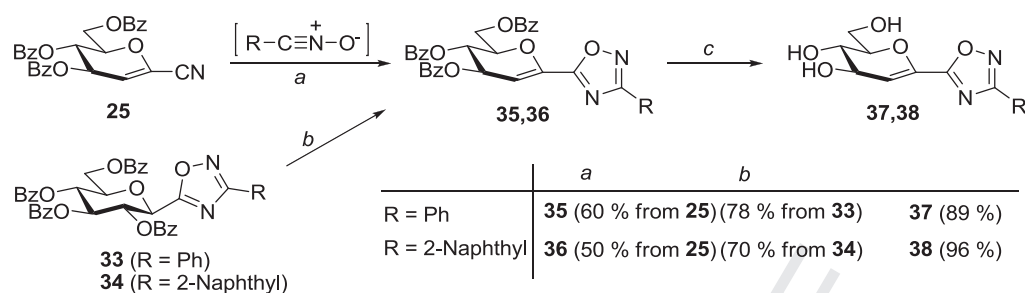
Scheme 2. Reagents and conditions: a) RC(Cl)NOH, dry toluene, Ar, reflux; b) DBU, dry CH_2Cl_2, rt; c) cat. NaOMe in dry MeOH, rt.
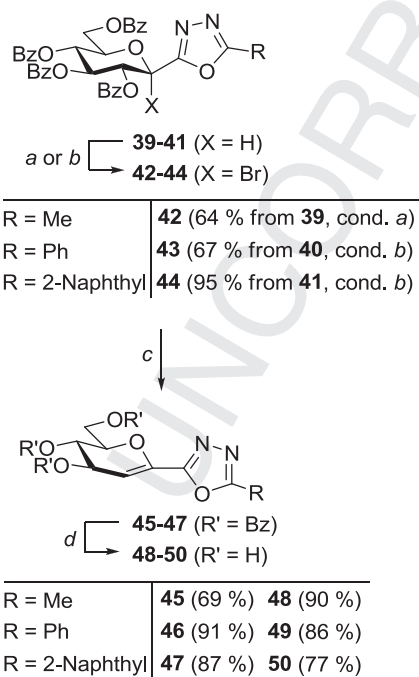
while the same signals appeared in the 5.8–6-1 ppm range for the deprotected derivatives **31**, **32**, **37**, **38**, **48**–**50**. Characteristic ^{13}C resonances are collected in Table 1 to show the C-1′ and C-2′ signals for the double bond in the sugar rings as well as the C-2 or C-3 and C-5 peaks for the heterocycles.

2.2. Enzyme kinetic studies

The unprotected oxadiazoles **31**, **32**, **37**, **38**, and **48**–**50** were assayed against rabbit muscle glycogen phosphorylase *b* as described earlier[58] with maximal inhibitor concentrations of 625 µM. Under these circumstances none of the compounds inhibited the enzyme (Table 2, entry 2). A comparison with the 'parent' *C*-glucopyranosyl oxadiazoles (Table 2, entry 1) shows that either the removal of the 2-OH or the change in the conformation of the sugar ring or both resulted in a complete loss of the activity. The alterations of the sugar ring could not be compensated by the additional interactions of the aglycons in the β-channel of the enzyme, and among others this might be due to a move in their position as a consequence of the conformational change of the

pyranoid ring. Nevertheless, the effect of glucals attached to better aglycons (e.g., 1,2,4-triazoles **19** in Chart 1) remains an open question, and further studies in this direction are in progress in our laboratory.

In conclusion, syntheses of ᴅ-glucals conjugated to each isomer of oxadiazoles by a C–C bond were carried out. These compounds could be prepared by introducing the double bond in the precursor glucopyranosyl cyanide followed by further manipulations to cyclize the aglycon or formation of the unsaturated sugar ring in the *C*-glucosyl heterocycle. Either base induced elimination of benzoic acid from the O-perbenzoylated starting compounds or Zn/N-methylimidazole mediated reductive elimination from the 1-bromoglucosyl precursor molecules could be applied for the formation of the hex-1-enopyranosyl rings. However, no general sequence of the above steps could be established, therefore, specific syntheses had to be elaborated for each target compound. None of the studied glucal derived oxadiazoles showed inhibition against RMGP*b* indicating that the binding of the these aglycons was not strong enough to override the detrimental effects of the changes in the sugar parts of the molecules.

3. Experimental

3.1. General methods

Melting points were measured on a Kofler hot-stage and are uncorrected. Optical rotations were determined with a Perkin–Elmer 241 polarimeter at rt. NMR spectra were recorded with Bruker 360 (360/90 MHz for ^1H/^{13}C) or Bruker 400 (400/100 MHz for ^1H/^{13}C) spectrometers. Chemical shifts are referenced to Me_4Si (^1H), or to the residual solvent signals (^{13}C). Mass spectra were obtained by a Thermo Scientific LTQ XL instrument (sample injection in 50:50:0.1 CH_3CN–H_2O–HCOOH or 50:50:0.1 MeOH–H_2O–CH_3COONH_4). TLC was performed on DC-Alurolle Kieselgel 60 F_{254} (Merck) plates, visualized under UV light and by gentle heating. For column chromatography Kieselgel 60 (Merck, particle size 0.063–0.200 mm) was used. Toluene, CH_2Cl_2, $CHCl_3$, EtOAc were distilled from P_4O_{10} and stored over 4 Å molecular sieves or sodium wires. MeOH was purified by distillation after refluxing for a couple of hours with magnesium turnings and iodine. 1,4-Dioxane was distilled from sodium benzophenone ketyl and stored over sodium wires. 2,3,4,6-Tetra-O-benzoyl-β-ᴅ-glucopyranosyl cyanide[49] (**23**), 2,3,4,6-tetra-O-benzoyl-1-bromo-1-deoxy-β-ᴅ-glucopyranosyl cyanide[49] (**24**), N-hydroxy-arenecarboximidoyl chlorides,[39] 3-aryl-5-(2′,3′,4′,6′-tetra-O-benzoyl-β-ᴅ-glucopyranosyl)-1,2,4-oxadiazoles[39] (**33** and **34**) and 2-(2′,3′,4′,6′-tetra-O-benzoyl-β-ᴅ-glucopyranosyl)-5-substituted-1,2,4-oxadiazoles[40,39] (**39**–**41**) were synthesized according to published procedures.

Scheme 3. Reagents and conditions: a) $KBrO_3$, $Na_2S_2O_4$, CH_2Cl_2, H_2O, rt; b) Br_2, dry $CHCl_3$, K_2CO_3, hν, reflux; c) Zn, N-methylimidazole, dry EtOAc, reflux; d) cat. NaOMe in dry MeOH, rt.

5

Table 1
Characteristic ^{13}C resonances of the oxadiazole derivatives

		C-1′	C-2′	C-3	C-5			C-1′	C-2′	C-3	C-5
R=Ph	**29**	144.5	103.2	164.9	175.9	**31**		141.3	110.4	164.8	174.6
R=2-naphthyl	**30**	144.5	103.2	164.9	175.9	**32**		141.3	110.5	164.9	174.8
R=Ph	**35**	141.7	105.2	168.8	170.3	**37**		141.1	112.9	169.8	172.9
R=2-naphthyl	**36**	141.7	105.3	168.9	170.4	**38**		138.7	113.0	168.0	171.3
				C-2						C-2	
R=Me	**45**	141.3	102.4	160–166[a]		**48**		140.2	110.2	166.1[b]	162.0[b]
R=Ph	**46**	141.2	102.6	159–166[a]		**49**		138.2	110.1	163.7[b]	159.9[b]
R=2-naphthyl	**47**	141.3	102.7	159–166[a]		**50**		138.3	110.2	163.9[b]	160.0[b]

[a] The range of the C-2, C-5 and CO signals without assignment.
[b] Interchangable assignments.

3.2. General procedure I for the preparation of 3,4,6-tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl derivatives (**25**, **35**, **36**) by DBU induced PhCOOH elimination

To a solution of the corresponding 2,3,4,6-tetra-O-benzoyl-β-D-glucopyranosyl derivative (**23**, **33**, **34**) in anhydrous CH_2Cl_2 (1 mmol/10 mL) DBU (1.5–2.0 equiv) was added and the reaction mixture was stirred at rt. When the TLC (1:4 EtOAc-hexane) showed total consumption of the starting material the mixture was diluted with CH_2Cl_2, extracted with satd aq $KHSO_4$ solution then with water. The organic phase was dried over $MgSO_4$, filtered and concentrated under diminished pressure. The residue was purified by column chromatography.

3.3. General procedure II for the synthesis of 3,4,6-tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl derivatives (**25**, **45**, **47**) by Zn/N-methylimidazole mediated reductive elimination

The corresponding 2,3,4,6-tetra-O-benzoyl-1-bromo-1-deoxy-β-D-glucopyranosyl derivative (**24**, **42**–**44**) was dissolved in anhydrous EtOAc (1 mmol/10 mL), activated zinc dust[35] (10 equiv) was added and the mixture was stirred at reflux temperature. N-Methylimidazole (5 equiv) was added to the boiling suspension, and the heating was continued until the starting material disappeared (TLC, 3:7 EtOAc-hexane). Charcoal was added to the hot mixture and the solids were filtered off through a pad of Celite. The filtrate was then extracted with 1 M aq HCl solution, satd aq $NaHCO_3$ solution and water. The organic phase was dried over $MgSO_4$, filtered and evaporated. The residue was purified by column chromatography.

3.4. General procedure III for the synthesis of O-aroyl-C-(3,4,6-tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl) formamidoximes (**27** and **28**)

To the solution of C-(3,4,6-tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyrano-syl)formamidoxime (**26**) in anhydrous 1,4-dioxane (1 mmol/5 mL) an acid chloride (1.1 equiv) was added and the reaction mixture was stirred at rt. After total consumption of the starting material (TLC 1:2 EtOAc-hexane) the solvent was removed under reduced pressure. The crude product was then purified by column chromatography.

Table 2
Inhibition (K_i [µM]) of RMGPb by C-glycosyl oxadiazoles

Entry	Sugar part	Heterocycle						
		R=Ph	R=2-naphthyl	R=Ph	R=2-naphthyl	R=Me	R=Ph	R=2-naphthyl
1		**14** 10% inh. at 625 µM[43]	38[43]	64[39]	**15** 12 (2.4)[39]	**16** 212[40] 145[41]	10% inh. at 625 µM[39]	**17** 10% inh. at 625 µM[39]
2		**31** No inhibition at 625 µM concentration.	**32**	**37**	**38**	**48**	**49**	**50**

3.5. General procedure IV for the synthesis of 5-aryl-3-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-hex-1′-enopyranosyl)-1,2,4-oxadiazoles (29 and 30)

An O-aroyl-C-(3,4,6-tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl)formamidoxime (27 or 28) was dissolved in toluene (1 mmol/15 mL), a 1 M solution of Bu₄NF in THF (0.1 equiv) was added and the mixture was refluxed. After completion of the reaction monitored by TLC (1:2 EtOAc-hexane) the solvent was removed and the residue was purified by column chromatography.

3.6. General procedure V for removal of benzoyl protecting groups by the Zemplén protocol

To a solution of an O-perbenzoylated compound in anhydrous MeOH (5 mL/100 mg, a few drops of anhydrous CHCl₃ were added in case of incomplete dissolution) a catalytic amount of a NaOMe solution (1 M in MeOH) was added and the mixture was left at rt. After completion of the reaction monitored by TLC (9:1 CHCl₃–MeOH) the mixture was neutralized with a cation exchange resin Amberlyst 15 (H⁺ form), then the resin was filtered off and the solvent was removed. The crude product was purified by column chromatography.

3.7. General procedure VI for the synthesis of 3-aryl-5-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-hex-1′-enopyranosyl)-1,2,4-oxadiazole (35 and 36) from cyanide 25 by 1,3-dipolar cycloaddition

3,4,6-Tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl cyanide (25, 0.50 g, 1.03 mmol) and the corresponding N-hydroxy-arenecarboximidoyl chloride (5.0 equiv) were dissolved in anhydrous toluene (12 mL) and stirred at reflux temperature under Ar atmosphere. To this mixture a solution of Et₃N (1.08 mL, 7.76 mmol, 7.5 equiv) in anhydrous toluene (12 mL) was added with a syringe pump in 8 h. The reaction mixture was heated for an additional 12 h and then the solvent was removed under reduced pressure. The residue was purified by column chromatography.

3.8. General procedure VII for the preparation of 5-aryl-2-(2′,3′,4′,6′-tetra-O-benzoyl-1′-bromo-1′-deoxy-β-D-glucopyranosyl)-1,3,4-oxadiazoles (43 and 44)

A 5-aryl-2-(2′,3′,4′,6′-tetra-O-benzoyl-β-D-glucopyranosyl)-1,3,4-oxadiazole (40 or 41, 2.0 g) was dissolved in anhydrous CHCl₃ (30 mL) in an Erlenmeyer flask, and bromine (3 equiv) and some solid K₂CO₃ were added. The mixture was placed above a heat lamp (375 W, distance from the lamp 2–3 cm, height of the solution 1–2 cm), and refluxed. After total consumption of the starting material monitored by TLC (1:3 EtOAc-hexane) the mixture was diluted with CHCl₃ (100 mL) and extracted with 1 M aq Na₂SO₃ solution (80 mL), satd aq NaHCO₃ solution (2×80 mL) and with water (80 mL). The organic phase was dried over MgSO₄, filtered and evaporated under diminished pressure. The residual crude product was purified either by crystallization or by column chromatography.

3.9. Synthesis and characterization of the compounds

3.9.1. 3,4,6-Tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl cyanide (25)

A: Prepared from cyanide 23[49] (10.00 g, 16.50 mmol) and DBU (4.92 mL, 33.00 mmol) according to General procedure **I** (Section 3.2). Reaction time: 3 h. Purified by column chromatography (1:4 EtOAc-hexane) to yield 3.97 g (50%) colorless syrup.

B: Prepared from cyanide 24[49] (4.13 g, 6.04 mmol) according to General procedure **II** (Section 3.3). Reaction time: 2 h. Purified by column chromatography (1:4 EtOAc-hexane) to yield 2.08 g (92%) colorless syrup. R_f: 0.39 (1:4 EtOAc-hexane); $[\alpha]_D$ −43 (c 0.50, CHCl₃); ¹H NMR (CDCl₃) δ (ppm): 8.05–7.97 (6H, m, aromatics), 7.60–7.54 (3H, m, aromatics), 7.46–7.39 (9H, m, aromatics), 5.99 (1H, d, J=4.0 Hz, H-2), 5.81 (1H, pseudo t, J=5.9, 5.3 Hz, H-4), 5.76 (1H, pseudo t, J=5.3, 4.0 Hz, H-3), 4.85 (1H, ddd, J=5.9, 5.3, < 1 Hz, H-5), 4.75–4.66 (2H, m, H-6a, H-6b); ¹³C NMR (CDCl₃) δ (ppm): 165.9, 165.2, 164.7 (C=O), 133.8, 133.7, 133.3 (aromatics), 131.0 (C-1), 129.8–128.4 (aromatics), 112.9 (C≡N), 112.1 (C-2), 75.8, 66.3, 66.0 (C-3–C-5), 61.0 (C-6). MS-ESI (m/z, positive mode): Calcd for $C_{28}H_{25}N_2O_7^+$ [M+NH₄]⁺: 501.2. Found: 501.8.

3.9.2. C-(3,4,6-Tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl)formamidoxime (26)

3,4,6-Tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl cyanide (25, 3.00 g, 6.21 mmol) and hydroxylamine hydrochloride (1.08 g, 15.53 mmol, 2.5 equiv) were stirred in anhydrous pyridine (20 mL) at rt. When the TLC (1:1 EtOAc-hexane) showed total disappearance of the starting material (2 d) the reaction mixture was diluted with EtOAc (200 mL) and extracted with water (200 mL). The organic phase was then washed with 1 M aq HCl solution (200 mL), with satd aq NaHCO₃ solution (200 mL) and with water (200 mL), respectively. The separated organic phase was dried over MgSO₄, filtered and the solvent was removed by diminished pressure. The resulted colorless oil (2.70 g, 84%) was then used without further purification. R_f: 0.47 (1:1 EtOAc-hexane); $[\alpha]_D$ −23 (c 0.50, CHCl₃); ¹H NMR (CDCl₃) δ (ppm): 8.04–7.97 (6H, m, aromatics), 7.56–7.48 (3H, m, aromatics), 7.43–7.35 (6H, m, aromatics), 5.83–5.78 (3H, m, H-2, H-3, H-4), 4.91 (2H, s, NH₂), 4.80–4.69 (3H, m, H-5, H-6a, H-6b); ¹³C NMR (CDCl₃) δ (ppm): 166.1, 165.6, 165.0 (C=O), 147.6, 146.3 (C-1, C=N), 133.5–128.4 (aromatics), 97.0 (C-2), 74.7, 67.5 (2) (C-3–C-5), 61.6 (C-6). MS-ESI (m/z, positive mode): Calcd for $C_{28}H_{25}N_2O_8^+$ [M+H]⁺: 517.2. Found: 517.7.

3.9.3. O-Benzoyl-C-(3,4,6-tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl)form-amidoxime (27)

Prepared from amidoxime 26 (1.50 g, 2.90 mmol) and benzoyl chloride (0.37 mL, 3.19 mmol) according to General procedure **III** (Section 3.4). Reaction time: 1 d. Purified by column chromatography (1:3 EtOAc-hexane) to yield 1.53 g (85%) white amorphous solid. R_f: 0.53 (1:1 EtOAc-hexane); $[\alpha]_D$ −13 (c 0.50, CHCl₃); ¹H NMR (CDCl₃) δ (ppm): 8.04–7.37 (20H, m, aromatics), 6.22 (1H, d, J=3.9 Hz, H-2), 5.86 (1H, pseudo t, J=4.7, 3.9 Hz, H-3), 5.80 (1H, pseudo t, J=5.5, 4.7 Hz, H-4), 5.43 (2H, s, NH₂), 4.86–4.69 (3H, m, H-5, H-6a, H-6b); ¹³C NMR (CDCl₃) δ (ppm): 166.3, 165.4, 165.1, 163.7 (C=O), 152.1, 144.7 (C=N, C-1), 133.7–128.6 (aromatics), 100.1 (C-2), 75.5, 67.5, 66.9 (C-3–C-5), 61.6 (C-6). MS-ESI (m/z, positive mode): Calcd for $C_{35}H_{29}N_2O_9^+$ [M+H]⁺: 621.2. Found: 621.3.

3.9.4. O-(2-Naphthoyl)-C-(3,4,6-tri-O-benzoyl-2-deoxy-D-arabino-hex-1-enopyranosyl)-formamidoxime (28)

Prepared from amidoxime 26 (2.00 g, 3.87 mmol) and 2-naphthoyl chloride (0.81 g, 4.26 mmol) according to General procedure **III** (Section 3.4). Reaction time: 1 d. Purified by column chromatography (1:3 EtOAc-hexane) to yield 2.21 g (85%) white amorphous solid. R_f: 0.33 (1:3 EtOAc-hexane); $[\alpha]_D$ −10 (c 0.50, CHCl₃); ¹H NMR (CDCl₃) δ (ppm): 8.58 (1H, s, aromatic), 8.04–7.34 (21H, m, aromatics), 6.27 (1H, d, J=4.0 Hz, H-2), 5.88 (1H, pseudo t, J=5.3, 4.0 Hz, H-3), 5.84 (1H, pseudo t, J=5.9, 5.3 Hz, H-4), 5.57 (2H, s, NH₂), 4.86–4.70 (3H, m, H-5, H-6a, H-6b); ¹³C NMR (CDCl₃) δ (ppm): 166.3, 165.4, 165.1, 163.9 (C=O), 152.2, 144.7 (C=N, C-1), 135.4–124.9 (aromatics), 100.1 (C-2), 75.4, 67.5, 67.0 (C-3–C-5), 61.6

(C-6). MS-ESI (*m/z*, positive mode): Calcd for $C_{39}H_{31}N_2O_9^+$ [M+H]$^+$: 671.2. Found: 671.3.

3.9.5. 5-Phenyl-3-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-hex-1′-enopyranosyl)-1,2,4-oxadiazole (29)

Prepared from compound 27 (1.00 g, 1.61 mmol) according to General procedure IV (Section 3.5). Reaction time: 1 d. Purified by column chromatography (1:2 EtOAc-hexane) to yield 0.96 g (99%) white solid. Mp: 154–155 °C; [α]$_D$ +24 (*c* 0.50, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ (ppm): 8.16–7.39 (20H, m, aromatics), 6.46 (1H, d, *J*=3.9 Hz, H-2′), 5.96–5.94 (2H, m, H-3′, H-4′), 5.05 (1H, ddd, *J*=6.3, 5.5, 4.7 Hz, H-5′), 4.87 (1H, dd, *J*=12.5, 6.3 Hz, H-6′a), 4.80 (1H, dd, *J*=12.5, 4.7 Hz, H-6′b); ^{13}C NMR (CDCl$_3$) δ (ppm): 175.9 (oxadiazole C-5), 166.2, 165.7, 165.2, 164.9 (C=O, oxadiazole C-3), 144.5 (C-1′), 133.6–123.8 (aromatics), 103.2 (C-2′), 75.0, 67.4, 67.0 (C-3′–C-5′), 61.7 (C-6′). MS-ESI (*m/z*, positive mode): Calcd for $C_{35}H_{27}N_2O_8^+$ [M+H]$^+$: 603.2. Found: 603.3.

3.9.6. 5-(2-Naphthyl)-3-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-hex-1′-enopyranosyl)-1,2,4-oxadiazole (30)

Prepared from compound 28 (1.00 g, 1.49 mmol) according to General procedure IV (Section 3.5). Reaction time: 1 d. Purified by column chromatography (1:2 EtOAc-hexane) to yield 0.86 g (88%) white amorphous solid. *Rf*: 0.42 (1:2 EtOAc-hexane); [α]$_D$ +20 (*c* 0.50, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ (ppm): 8.68 (1H, s, aromatic), 8.14–7.38 (21H, m, aromatics), 6.53 (1H, d, *J*=3.3 Hz, H-2′), 6.00–5.97 (2H, m, H-3′, H-4′), 5.07 (1H, ddd, *J*=5.9, 5.3, 4.6 Hz, H-5′), 4.91 (1H, dd, *J*=11.9, 5.9 Hz, H-6′a), 4.83 (1H, dd, *J*=11.9, 4.6 Hz, H-6′b); ^{13}C NMR (CDCl$_3$) δ (ppm): 175.9 (oxadiazole C-5), 166.2, 165.7, 165.1, 164.9 (C=O, oxadiazole C-3), 144.5 (C-1′), 135.3–120.9 (aromatics), 103.2 (C-2′), 74.9, 67.4, 67.0, (C-3′–C-5′), 61.7 (C-6′). MS-ESI (*m/z*, positive mode): Calcd for $C_{39}H_{29}N_2O_8^+$ [M+H]$^+$: 653.2. Found: 653.2.

3.9.7. 3-(2′-Deoxy-D-arabino-hex-1′-enopyranosyl)-5-phenyl-1,2,4-oxadiazole (31)

Prepared from compound 29 (0.30 g, 0.50 mmol) according to General procedure V (Section 3.6). Reaction time: 1 h. Purified by column chromatography (9:1 CHCl$_3$–MeOH) to yield 0.10 g (72%) white solid. Mp: 212–214 °C; [α]$_D$ +16 (*c* 0.50, DMSO); ^{1}H NMR (DMSO-d_6) δ (ppm): 8.12 (2H, d, *J*=7.3 Hz, aromatics), 7.72 (1H, t, *J*=7.3 Hz, aromatic), 7.64 (2H, t, *J*=7.3 Hz, aromatics), 5.88 (1H, d, *J*=2.0 Hz, H-2′), 5.36, 5.30, 4.76 (3×1H, 3×OH), 4.19–4.15, 3.92–3.89, 3.85–3.81, 3.77–3.71, 3.62–3.56 (5×1H, 5m, H-3′, H-4′, H-5′, H-6′a, H-6′b); ^{13}C NMR (DMSO-d_6) δ (ppm): 174.6 (oxadiazole C-5), 164.8 (oxadiazole C-3), 141.3 (C-1′), 133.4, 129.6 (2), 127.9 (2), 123.2 (aromatics), 110.4 (C-2′), 80.8, 68.4, 68.1 (C-3′–C-5′), 59.9 (C-6′). MS-ESI (*m/z*, negative mode): Calcd for $C_{16}H_{17}N_2O_7$ [M+AcO]$^-$: 349.1. Found: 349.7.

3.9.8. 3-(2′-Deoxy-D-arabino-hex-1′-enopyranosyl)-5-(2-naphthyl)-1,2,4-oxadiazole (32)

Prepared from compound 30 (0.30 g, 0.50 mmol) according to General procedure V (Section 3.6). Reaction time: 1 h. Purified by column chromatography (9:1 CHCl$_3$–MeOH) to yield 0.13 g (85%) white solid. Mp: 199–200 °C; [α]$_D$ +20 (*c* 0.50, DMSO); ^{1}H NMR (DMSO-d_6) δ (ppm): 8.81 (1H, s, aromatic), 8.21–7.64 (6H, m, aromatics), 5.93 (1H, s, H-2′), 5.05 (3H, br signal, OH), 4.20–4.19, 3.91, 3.86–3.83, 3.78–3.74, 3.64–3.59 (5×1H, 5m, H-3′, H-4′, H-5′, H-6′a, H-6′b); ^{13}C NMR (DMSO-d_6) δ (ppm): 174.8 (oxadiazole C-5), 164.9 (oxadiazole C-3), 141.3 (C-1′), 134.8, 132.3, 129.4, 129.3, 129.1, 128.9, 127.9, 127.5, 123.5, 120.4 (aromatics), 110.5 (C-2′), 80.8, 68.4, 68.3 (C-3′–C-5′), 60.0 (C-6′). MS-ESI (*m/z*, negative mode): Calcd for $C_{20}H_{19}N_2O_7$ [M+AcO]$^-$: 399.1. Found: 399.3.

3.9.9. 3-Phenyl-5-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-1′-enopyranosyl)-1,2,4-oxadiazole (35)

A: Prepared from oxadiazole 33[39] (0.20 g, 0.28 mmol) and DBU (62 μL, 0.41 mmol) according to General procedure I (Section 3.2). Reaction time: 1 d. Purified by column chromatography (1:7 EtOAc-hexane) to yield 0.13 g (78%) pale yellow syrup.

B: Prepared from cyanide 25 (0.50 g, 1.03 mmol) and N-hydroxy-benzenecarboximidoyl chloride[39] (0.80 g, 5.17 mmol) according to General procedure VI (Section 3.7). Purified by column chromatography (1:8 EtOAc-hexane) to yield 0.37 g (60%) pale yellow syrup. *Rf*: 0.67 (1:2 EtOAc-hexane); [α]$_D$ −11 (*c* 0.50, CHCl$_3$). ^{1}H NMR (CDCl$_3$) δ (ppm): 8.12–8.02 (8H, m, aromatics), 7.63–7.41 (12H, m, aromatics), 6.55 (1H, d, *J*=4.7 Hz, H-2′), 5.92–5.91 (2H, m, H-3′, H-4′), 5.07 (1H, ddd, *J*=7.0, 6.3, 4.7 Hz, H-5′), 4.89 (1H, dd, *J*=11.7, 6.3 Hz, H-6′a), 4.78 (1H, dd, *J*=11.7, 4.7 Hz, H-6′b); ^{13}C NMR (CDCl$_3$) δ (ppm): 170.3, 168.8 (oxadiazole C-3, C-5), 166.0, 165.4, 164.9 (C=O), 141.7 (C-1′), 133.7–126.2 (aromatics), 105.2 (C-2′), 75.2, 67.0, 66.3 (C-3′–C-5′), 61.2 (C-6′). MS-ESI (*m/z*, positive mode): Calcd for $C_{35}H_{27}N_2O_8^+$ [M+H]$^+$: 603.2. Found: 603.8.

3.9.10. 3-(2-Naphthyl)-5-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-hex-1′-enopyrano-syl)-1,2,4-oxadiazole (36)

A: Prepared from oxadiazole 34[39] (0.5 g, 0.65 mmol) and DBU (145 μL, 0.97 mmol) according to General procedure I (Section 3.2). Reaction time: 2 h. Purified by column chromatography (1:4 EtOAc-hexane) to yield 0.30 g (70%) white solid.

B: Prepared from cyanide 25 (0.5 g, 1.03 mmol) and N-hydroxy-naphthalene-2-carboximidoyl chloride[39] (1.06 g, 5.17 mmol) according to General procedure VI (Section 3.7). Purified by column chromatography (toluene) to yield 0.34 g (50%) white solid. Mp: 184–185 °C; [α]$_D$ −6 (*c* 0.50, CHCl$_3$). ^{1}H NMR (CDCl$_3$) δ (ppm): 8.66 (1H, s, aromatic), 8.24–7.41 (21H, m, aromatics), 6.60 (1H, d, *J*=4.7 Hz, H-2′), 5.94–5.93 (2H, m, H-3′, H-4′), 5.10 (1H, ddd, *J*=6.3, 4.7, 3.9 Hz, H-5′), 4.92 (1H, dd, *J*=12.5, 6.3 Hz, H-6′a), 4.80 (1H, dd, *J*=12.5, 4.7 Hz, H-6′b); ^{13}C NMR (CDCl$_3$) δ (ppm): 170.4, 168.9 (oxadiazole C-3, C-5), 166.0, 165.4, 164.9 (C=O), 141.7 (C-1′), 134.6–123.4 (aromatics), 105.3 (C-2′), 75.2, 67.0, 66.2 (C-3′–C-5′), 61.2 (C-6′). MS-ESI (*m/z*, positive mode): Calcd for $C_{39}H_{29}N_2O_8^+$ [M+H]$^+$: 653.2. Found: 653.1.

3.9.11. 5-(2′-Deoxy-D-arabino-hex-1′-enopyranosyl)-3-phenyl-1,2,4-oxadiazole[39] (37)

Prepared from compound 35 (0.20 g, 0.33 mmol) according to General procedure V (Section 3.6). Reaction time: 8 h. Purified by column chromatography (9:1 CHCl$_3$–MeOH) to yield 85 mg (89%) white solid. Mp: 198–199 °C; [α]$_D$ −8 (*c* 0.45, DMSO); ^{1}H NMR (CD$_3$OD) δ (ppm): 8.06 (2H, d, *J*=7.8 Hz, aromatic), 7.58–7.51 (3H, m, aromatics), 6.13 (1H, d, *J*=2.3 Hz, H-2′), 4.35 (1H, dd, *J*=7.0, 2.3 Hz, H-3′), 4.08 (1H, ddd, *J*=7.8, 5.5, 2.3 Hz, H-5′), 4.03 (1H, dd, *J*=12.5, 2.3 Hz, H-6′a), 3.94 (1H, dd, *J*=12.5, 5.5 Hz, H-6′b), 3.76 (1H, dd, *J*=7.8, 7.0 Hz, H-4′); ^{13}C NMR (CD$_3$OD) δ (ppm): 172.9, 169.8 (oxadiazole C-3, C-5), 141.1 (C-1′), 132.7–127.7 (aromatics), 112.9 (C-2′), 82.3, 70.3, 69.7 (C-3′–C-5′), 61.9 (C-6′).

3.9.12. 5-(2′-Deoxy-D-arabino-hex-1′-enopyranosyl)-3-(2-naphthyl)-1,2,4-oxadiazole[39] (38)

Prepared from compound 36 (0.19 g, 0.29 mmol) according to General procedure V (Section 3.6). Reaction time: 4 h. Purified by column chromatography (9:1 CHCl$_3$–MeOH) to yield 95 mg (96%) white solid. Mp: 202–203 °C; [α]$_D$ −8 (*c* 0.50, DMSO); ^{1}H NMR (DMSO-d_6+1–2 drops of D$_2$O) δ (ppm): 8.55 (1H, s, aromatic), 8.06–7.95 (4H, m, aromatics), 7.63–7.56 (2H, m, aromatics), 6.05 (1H, d, *J*=3.1 Hz, H-2′), 4.19 (1H, dd, *J*=6.3, 3.1 Hz, H-3′), 3.98 (1H, ddd, *J*=8.6, 5.5, 2.3 Hz, H-5′), 3.83 (1H, dd, *J*=12.5, 2.3 Hz, H-6′a), 3.74 (1H, dd, *J*=12.5, 5.5 Hz, H-6′b), 3.59 (1H, dd, *J*=8.6, 6.3 Hz, H-

4′); ^{13}C NMR (DMSO-d_6) δ (ppm): 171.3, 168.0 (oxadiazole C-3, C-5), 138.7 (C-1′), 134.2−123.2 (aromatics), 113.0 (C-2′), 81.1, 67.9, 67.7 (C-3′−C-5′), 59.6 (C-6′). MS-ESI (m/z, negative mode): Calcd for $C_{20}H_{19}N_2O_7$ [M+AcO]$^-$: 399.1. Found: 399.2.

3.9.13. 2-Methyl-5-(2′,3′,4′,6′-tetra-O-benzoyl-1′-bromo-1′-deoxy-β-D-glucopyranosyl)-1,3,4-oxadiazole (42)

2-Methyl-5-(2′,3′,4′,6′-tetra-O-benzoyl-β-D-glucopyranosyl)-1,3,4-oxadiazole[40] (39, 0.85 g, 1.28 mmol) was dissolved in CH_2Cl_2 (25 mL), and an aqueous solution of $KBrO_3$ (1.29 g, 7.70 mmol, 6 equiv in 25 mL water) was added. To the stirred heterogenous mixture an aqueous solution of $Na_2S_2O_4$ (1.34 g, 7.70 mmol, 6 equiv in 25 mL water) was added dropwise over 10 min. The mixture was then stirred at rt and the reaction was monitored by TLC (1:1 EtOAc-hexane). After disappearance of the starting material (1 d) the reaction mixture was diluted with CH_2Cl_2 (50 mL), extracted with 1 M aq solution of Na_2SO_3 (30 mL), with satd aq $NaHCO_3$ (2×30 mL) and with water (30 mL), respectively. The organic phase was dried over $MgSO_4$, filtered and evaporated under diminished pressure. The residue was purified by column chromatography (1:2 EtOAc-hexane) to yield 0.61 g (64%) white amorphous solid. R_f: 0.49 (1:1 EtOAc-hexane); [α]$_D$ +141 (c 0.50, CHCl₃); ^{1}H NMR (CDCl₃) δ (ppm): 8.07−7.25 (20H, m, aromatics), 6.30 (1H, pseudo t, J=9.4, 9.4 Hz, H-3′ or H-4′), 6.07 (1H, d, J=9.4 Hz, H-2′), 5.95 (1H, pseudo t, J=9.4, 9.4 Hz, H-3′ or H-4′), 4.92 (1H, ddd, J=9.4, 4.7, 2.3 Hz, H-5′), 4.73 (1H, dd, J=12.5, 2.3 Hz, H-6′a), 4.61 (1H, dd, J=12.5, 4.7 Hz, H-6′b), 2.48 (3H, s, CH₃); ^{13}C NMR (CDCl₃) δ (ppm): 165.8, 165.4, 165.0, 164.8, 164.3, 162.4 (C=O, oxadiazole C-2, C-5), 133.6−128.2 (aromatics), 90.8 (C-1′), 74.8, 71.9, 71.4, 67.5 (C-2′−C-5′), 61.7 (C-6′), 10.9 (CH₃). MS-ESI (m/z, positive mode): Calcd for $C_{37}H_{30}BrN_2O_{10}$ [M+H]$^+$: 741.1. Found: 741.5.

3.9.14. 2-Phenyl-5-(2′,3′,4′,6′-tetra-O-benzoyl-1′-bromo-1′-deoxy-β-D-glucopyranosyl)-1,3,4-oxadiazole (43)

Prepared from oxadiazole 40[39] (2.00 g, 2.76 mmol) according to General procedure VII (Section 3.8). Reaction time: 2 h. Purified by crystallization from EtOH to yield 1.49 g (67%) white solid. Mp: decomposition above 152 °C; [α]$_D$ +137 (c 0.50, CHCl₃); ^{1}H NMR (CDCl₃) δ (ppm): 8.08, 8.00, 7.96, 7.92, 7.85 (5×2H, 5d, J=7.3 Hz in each, aromatics), 7.60−7.26 (15H, m, aromatics), 6.32 (1H, pseudo t, J=9.2, 9.2 Hz, H-3′ or H-4′), 6.12 (1H, d, J=9.2 Hz, H-2′), 5.96 (1H, pseudo t, J=9.9, 9.2 Hz, H-3′ or H-4′), 4.95 (1H, ddd, J=9.9, 4.6, 2.6 Hz, H-5′), 4.77 (1H, dd, J=12.6, 2.6 Hz, H-6′a), 4.65 (1H, dd, J=12.6, 4.6 Hz, H-6′b); ^{13}C NMR (CDCl₃) δ (ppm): 165.9, 165.6, 165.5, 164.9, 164.4, 162.2 (C=O, oxadiazole C-2, C-5), 133.7−127.2 (aromatics), 90.9 (C-1′), 74.9, 72.0, 71.5, 67.8 (C-2′−C-5′), 61.8 (C-6′). MS-ESI (m/z, positive mode): Calcd for $C_{42}H_{32}BrN_2O_{10}$ [M+H]$^+$: 803.1. Found: 803.2.

3.9.15. 2-(2-Naphthyl)-5-(2′,3′,4′,6′-tetra-O-benzoyl-1′-bromo-1′-deoxy-β-D-glucopy-ranosyl)-1,3,4-oxadiazole (44)

Prepared from oxadiazole 41[39] (2.00 g, 2.58 mmol) according to General procedure VII (Section 3.8). Reaction time: 2 h. Purified by column chromatography (1:3 EtOAc-hexane) to yield 2.09 g (95%) colorless syrup. R_f: 0.35 (EtOAc-hexane 1:2); [α]$_D$ +104 (c 0.50, CHCl₃); ^{1}H NMR (CDCl₃) δ (ppm): 8.37 (1H, s, aromatic), 8.10−7.25 (26H, m, aromatics), 6.36 (1H, pseudo t, J=9.4, 9.4 Hz, H-3′ or H-4′), 6.15 (1H, d, J=9.4 Hz, H-2′), 6.00 (1H, pseudo t, J=9.4, 9.4 Hz, H-3′ or H-4′), 4.99 (1H, ddd, J=9.4, 4.7, 2.3 Hz, H-5′), 4.80 (1H, dd, J=12.5, 2.3 Hz, H-6′a), 4.69 (1H, dd, J=12.5, 4.7 Hz, H-6′b); ^{13}C NMR (CDCl₃) δ (ppm): 165.9, 165.8, 165.5, 164.9, 164.4, 162.3 (C=O, oxadiazole C-2, C-5), 134.8−120.1 (aromatics), 90.9 (C-1′), 74.9, 72.0, 71.5, 67.7 (C-2′−C-5′), 61.8 (C-6′). MS-ESI (m/z, positive mode): Calcd for $C_{46}H_{34}BrN_2O_{10}$ [M+H]$^+$: 853.1. Found: 853.3.

3.9.16. 2-Methyl-5-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-hex-1′-enopyranosyl)-1,3,4-oxadiazole (45)

Prepared from compound 42 (0.40 g, 0.54 mmol) according to General procedure II (Section 3.3). Reaction time: 10 min. Purified by column chromatography (1:1 EtOAc-hexane) to yield 0.20 g (69%) colorless syrup. R_f: 0.40 (1:1 EtOAc-hexane); [α]$_D$ −29 (c 0.50, CHCl₃); ^{1}H NMR (CDCl₃) δ (ppm): 8.05−7.39 (15H, m, aromatics), 6.24 (1H, d, J=3.9 Hz, H-2′), 5.90 (1H, pseudo t, J=5.5, 4.7 Hz, H-3′ or H-4′) 5.86 (1H, pseudo t, J=4.7, 3.9 Hz, H-3′ or H-4′), 5.01 (1H, ddd, J=6.3, 5.5, 4.7 Hz, H-5′), 4.85 (1H, dd, J=11.7, 6.3 Hz, H-6′a), 4.75 (1H, dd, J=11.7, 4.7 Hz, H-6′b), 2.57 (3H, s, CH₃); ^{13}C NMR (CDCl₃) δ (ppm): 166.0, 165.5, 164.9, 164.2, 159.9 (C=O, oxadiazole C-2, C-5), 141.3 (C-1′), 133.7−128.4 (aromatics), 102.4 (C-2′), 75.1, 67.0, 66.5 (C-3′−C-5′), 61.3 (C-6′), 11.0 (CH₃). MS-ESI (m/z, positive mode): Calcd for $C_{30}H_{25}N_2O_8$ [M+H]$^+$: 541.2. Found: 541.7.

3.9.17. 2-Phenyl-5-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-hex-1′-enopyranosyl)-1,3,4-oxadiazole (46)

Prepared from compound 43 (1.00 g, 1.24 mmol) according to General procedure II (Section 3.3). Reaction time: 15 min. Purified by column chromatography (2:3 EtOAc-hexane) to yield 0.68 g (91%) colorless syrup. R_f: 0.33 (2:3 EtOAc-hexane); [α]$_D$ −7 (c 0.50, CHCl₃); ^{1}H NMR (CDCl₃) δ (ppm): 8.08−7.39 (20H, m, aromatics), 6.38 (1H, d, J=3.9 Hz, H-2′), 5.96−5.92 (2H, m, H-3′, H-4′), 5.08 (1H, ddd, J=6.3, 5.5, 4.7 Hz, H-5′), 4.94 (1H, dd, J=11.7, 6.3 Hz, H-6′a), 4.79 (1H, dd, J=11.7, 4.7 Hz, H-6′b); ^{13}C NMR (CDCl₃) δ (ppm): 165.9, 165.4, 164.8 (2), 159.4 (C=O, oxadiazole C-2, C-5), 141.2 (C-1′), 133.6−123.1 (aromatics), 102.6 (C-2′), 75.0, 67.0, 66.4 (C-3′−C-5′), 61.2 (C-6′). MS-ESI (m/z, positive mode): Calcd for $C_{35}H_{27}N_2O_8$ [M+H]$^+$: 603.2. Found: 603.3.

3.9.18. 2-(2-Naphthyl)-5-(3′,4′,6′-tri-O-benzoyl-2′-deoxy-D-arabino-hex-1′-enopyrano-syl)-1,3,4-oxadiazole (47)

Prepared from compound 44 (1.00 g, 1.17 mmol) according to General procedure II (Section 3.3). Reaction time: 15 min. Purified by column chromatography (2:3 EtOAc-hexane) to yield 0.67 g (87%) colorless syrup. R_f: 0.34 (1:2 EtOAc-hexane); [α]$_D$ +9 (c 0.50, CHCl₃); ^{1}H NMR (CDCl₃) δ (ppm): 8.53 (1H, s, aromatic), 8.13−7.40 (21H, m, aromatics), 6.43 (1H, d, J=3.9 Hz, H-2′), 5.98−5.93 (2H, m, H-3′, H-4′), 5.10 (1H, ddd, J=6.3, 5.5, 4.7 Hz, H-5′), 4.96 (1H, dd, J=11.7, 6.3 Hz, H-6′a), 4.81 (1H, dd, J=11.7, 4.7 Hz, H-6′b); ^{13}C NMR (CDCl₃) δ (ppm): 166.0, 165.5, 165.1, 164.9, 159.5 (C=O, oxadiazole C-2, C-5), 141.3 (C-1′), 134.7−120.3 (aromatics), 102.7 (C-2′), 75.1, 67.1, 66.5 (C-3′−C-5′), 61.2 (C-6′). MS-ESI (m/z, positive mode): Calcd for $C_{39}H_{29}N_2O_8$ [M+H]$^+$: 653.2. Found: 653.1.

3.9.19. 5-(2′-Deoxy-D-arabino-hex-1′-enopyranosyl)-2-methyl-1,3,4-oxadiazole (48)

Prepared from compound 45 (0.21 g, 0.38 mmol) according to General procedure V (Section 3.6). Reaction time: 15 min. Purified by column chromatography (9:1 CHCl₃−MeOH) to yield 0.08 g (90%) white solid. Mp: 154−155 °C; [α]$_D$ −8 (c 0.50, DMSO); ^{1}H NMR (CD₃OD) δ (ppm): 5.79 (1H, d, J=3.1 Hz, H-2′), 4.31 (1H, dd, J=7.0, 3.1 Hz, H-3′), 4.02−3.96 (2H, m, H-5′, H-6′a), 3.88 (1H, dd, J=11.7, 6.3 Hz, H-6′b), 3.69 (1H, dd, J=9.3, 7.0 Hz, H-4′), 2.57 (3H, s, CH₃); ^{13}C NMR (CD₃OD) δ (ppm): 166.1, 162.0 (oxadiazole C-2, C-5), 140.2 (C-1′), 110.2 (C-2′), 82.0, 70.2, 69.9 (C-3′−C-5′), 62.1 (C-6′), 10.7 (CH₃). MS-ESI (m/z, negative mode): Calcd for $C_{11}H_{15}N_2O_7$ [M+AcO]$^-$: 287.1. Found: 287.6.

3.9.20. 5-(2′-Deoxy-D-arabino-hex-1′-enopyranosyl)-2-phenyl-1,3,4-oxadiazole (49)

Prepared from compound 46 (0.30 g, 0.50 mmol) according to General procedure V (Section 3.6). Reaction time: 1 h. Purified by column chromatography (9:1 CHCl₃−MeOH) to yield 0.12 g (86%)

white solid. Mp: 193–194 °C; $[\alpha]_D$ +8 (c 0.50, DMSO); ^{1}H NMR (DMSO-d_6) δ (ppm): 8.05 (2H, dd, J=8.2, 1.4 Hz, aromatics), 7.67–7.59 (3H, m, aromatics), 5.89 (1H, d, J=3.4 Hz, H-2′), 5.41–5.37 (2H, 2×OH), 4.81 (1H, OH), 4.19–4.15, 3.96–3.92, 3.85–3.80, 3.77–3.71, 3.62–3.57 (5×1H, 5m, H-3′, H-4′, H-5′, H-6′a, H-6′b); ^{13}C NMR (DMSO-d_6) δ (ppm): 163.7, 159.9 (oxadiazole C-2, C-5), 138.2 (C-1′), 129.6, 126.8, 126.5, 123.0 (aromatics), 110.1 (C-2′), 81.01, 68.0, 67.9 (C-3′–C-5′), 59.7 (C-6′). MS-ESI (m/z, negative mode): Calcd for $C_{16}H_{17}N_2O_7^-$ [M+AcO]$^-$: 349.1. Found: 349.7.

3.9.21. 5-(2′-Deoxy-D-arabino-hex-1′-enopyranosyl)-2-(2-naphthyl)-1,3,4-oxadiazole (50)

Prepared from compound 47 (0.20 g, 0.31 mmol) according to General procedure V (Section 3.6). Reaction time: 1 h. Purified by column chromatography (9:1 CHCl$_3$–MeOH) to yield 0.08 g (77%) white solid. Mp: decomposition above 174 °C; $[\alpha]_D$ +18 (c 0.50, DMSO); ^{1}H NMR (DMSO-d_6) δ (ppm): 8.68 (1H, s, aromatic), 8.18–8.03 (4H, m, aromatics), 7.70–7.63 (2H, m, aromatics), 5.96 (1H, d, J=2.7 Hz, H-2′), 5.43–5.40 (2H, 2×OH), 4.83 (1H, OH), 4.22–4.18, 3.99–3.95, 3.87–3.82, 3.78–3.74, 3.65–3.59 (5×1H, 5m, H-3′, H-4′, H-5′, H-6′a, H-6′b); ^{13}C NMR (DMSO-d_6) δ (ppm): 163.9, 160.0 (oxadiazole C-2, C-5), 138.3 (C-1′), 134.2, 132.4, 129.3, 129.0, 128.8, 128.1, 127.7, 127.2, 120.3 (aromatics), 110.2 (C-2′), 81.0, 68.0, 67.9 (C-3′–C-5′), 59.7 (C-6′). MS-ESI (m/z, positive mode): Calcd for $C_{18}H_{17}N_2O_5^+$ [M+H]$^+$: 341.1. Found: 341.3.

3.10. Enzyme assay

Glycogen phosphorylase b was prepared from rabbit skeletal muscle according to the method of Fischer and Krebs,[59] using dithiothreitol instead of L-cysteine, and recrystallized at least three times before use with a specific activity of 55 U/mg protein. Kinetic experiments were performed in the direction of glycogen synthesis as described previously.[58] Kinetic data for the inhibition of rabbit skeletal muscle glycogen phosphorylase were collected using 4 mM of α-D-glucose-1-phosphate constant concentrations of glycogen (1% w/v) and AMP (1 mM), and various concentrations of inhibitor. Inhibitor was dissolved in dimethyl sulfoxide (DMSO) and diluted in the assay buffer (final concentrations between 6.25 and 625 μM) (50 mM triethanolamine, 100 mM KCl, 1 mM EDTA and 1 mM dithiothreitol) so that the DMSO concentration in the assay should be lower than 1%.

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