JURNAL KARTIKA KIMIA

J. Kartika Kimia, November 2023, 6, (2), 96-101
Journal homepage: http://jkk.unjani.ac.id/index.php/jkk
P-ISSN: 2655-1322 E-ISSN: 2655-0938

Synthesis and Biological Evaluation of N'-Arylidene-4-Hydroxybenzohydrazides against α-Glucosidase Enzyme

Ade Danova[1,*], Elvira Hermawati[1], Warinthorn Chavasiri[2]
[1]Organic Chemistry Division, Department of Chemistry, Faculty of Mathematics and Natural Sciences, Institut Teknologi Bandung, Jl. Ganesha 10, Bandung, West Java, 40132, Indonesia.
[2]Center of Excellence in Natural Products Chemistry, Department of Chemistry, Faculty of Science, Chulalongkorn University, Bangkok, 10330, Thailand.
*E-mail: adedanova@itb.ac.id

DOI: https://doi.org/10.26874/jkk.v6i2.210

Received: 3 Sept 2023, *Revised*: 20 Oct 2023, *Accepted*: 16 Nov 2023, *Online*: 22 Nov 2023

Abstract

*Hydrazides have been reported to have a broad spectrum of bioactivities, such as anticancer, antifungal, antibacterial, and antidiabetic. Thus, our work aimed to synthesize N'-arylidene-4-hydroxybenzohydrazides and investigate their α-glucosidase inhibition. Three compounds (**AD-H$_{1-3}$**) were synthesized with a yield of product from 33 to 65% and then characterized using ^{1}H-and ^{13}C-NMR spectroscopy. The in vitro α-glucosidase inhibition demonstrated that **AD-H$_2$** possessing two hydroxy groups on arylidene moiety showed the best inhibitory activity against α-glucosidase enzyme with an IC$_{50}$ value of 14.4 µM compared with acarbose as a positive control with IC$_{50}$ value of 93.6 µM. Thus, **AD-H$_2$** could be a candidate as a lead compound for antidiabetics.*

Keywords: *antidiabetics, hydrazide, α-glucosidase*

1 Introduction

Type 2 diabetes mellitus (T2DM) is a severe public health issue susceptible to 500 million people worldwide by 2030 [1]. The key features of T2DM include disorders in protein, lipid, and carbohydrate metabolism brought on by a progressive decrease in insulin production from the pancreatic β-cell, typically based on preexisting insulin resistance in skeletal muscle, liver, and adipose tissue [2]. As the primary clinical sign of T2DM, postprandial hyperglycemia can lead to various implications, such as cardiovascular disease, kidney disease, neuropathy, complications in lipid metabolism, and others [3–5].

T2DM and carbohydrate disorders have been linked to α-glucosidase as a key target for therapy [6]. In breaking down carbohydrates, glucose, which is found inside the brush-border surface membrane of intestinal cells, hydrolyzes disaccharides or oligosaccharides into monosaccharides [7]. Thus, delaying the digestion of carbohydrates can alleviate T2DM by reducing α-glucosidase activity [8]. In the market, α-glucosidase inhibitors (Figure 1) including acarbose, voglibose, miglitol, and emiglitate, have been successfully used in clinical trials to manage blood sugar levels and prevent T2DM, despite adverse effects such as vomiting, flatulence, acute stomach pain, allergic responses, and others [9]. Thus, synthesized safe drugs with enhanced efficacy and fewer adverse effects are crucial.

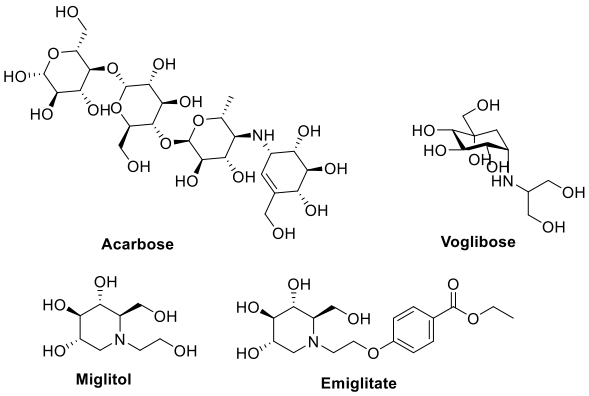


Figure 1. Structures of α-Glucosidase Inhibitors.

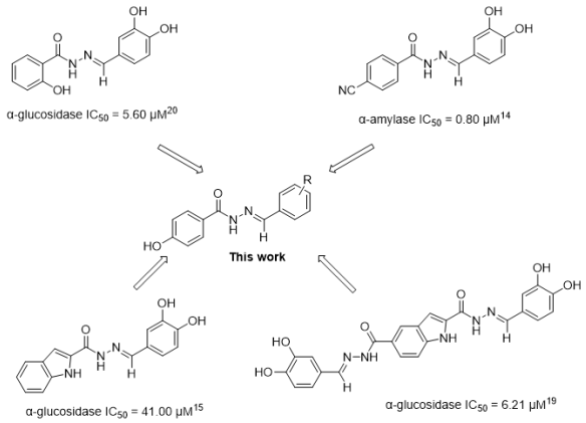


Figure 2. The current study of α-glucosidase inhibitors.

Hydrazide and thiosemicarbazone possess structures that are similar. Currently, hydrazides have been found to be beneficial against cancer, parasites, fungi, bacteria, and antidiabetics [10–14]. In a previous study, Taha et al. presented that synthesizing 2-indolcarbohydrazones and quinoline oxadiazole derivatives showed high inhibitory activity against α-glucosidase with IC_{50} values of 2.3, and 2.6 μM, respectively [15,16]. Wang et al. showed that chromone hydrazone derivatives exhibited high inhibitory activity against α-glucosidase with an IC_{50} value of 20.1 μM and coumarin thiazole derivatives with an IC_{50} value of 6.24 μM [17,18]. Hu et al. reported that indole-based bisacylhydrazone derivatives also demonstrated high inhibitory activity against α-glucosidase with an IC_{50} value of 1.01 μM [19]. Ullah et al. reported that 2-hydroxy-*N*-acylhydrazone and thiazolidine-4-one derivatives showed high inhibitory activity against α-glucosidase with IC_{50} values of 5.60 and 0.40 μM [20].

Based on the previous work, 2-hydroxy-*N*-acylhydrazone or *N'*-arylidene-2-hydroxybenzohydrazide showed high inhibitory activity against α-glucosidase [20]. As presented in Figure 2, our study aimed to synthesize *N'*-arylidene-4-hydroxybenzohydrazides from methyl 4-hydroxybenzoate with hydrazine followed by hydrazide formation with benzaldehyde derivatives in reflux condition. The inhibitory activity of *N'*-arylidene-4-hydroxybenzohydrazides was evaluated against α-glucosidase from *Saccharomyces cerevisiae*.

2 Materials and Method

2.1 Materials

All chemicals were purchased from TCI, Sigma Aldrich, and Merck. Silica gel for column chromatography (CC), (0.063–0.200 mm), was a product of Merck Company. TLC was performed on Merck TLC plates (0.23 mm thickness), with compounds visualized by UV light and vanillin sulphuric acid in ethanol and then heated on a hot plate. The NMR spectra were measured on JEOL JNM-EC500R 500 MHz. Chemical shifts are given in ppm (parts per million) using deuterated solvent (DMSO-d_6). α-Glucosidase enzyme (EC 3.2.1.20) from *Saccharomyces cerevisiae* and p-nitrophenyl-α-D-glucopyranoside were purchased from Sigma Aldrich.

2.2 Method

2.2.1 General Procedure for 4-Hydroxybenzohydrazide (AD-H)

Hydrazine hydrate (16.0 mL, 320 mmol, 8 eq) and methyl 4-hydroxybenzoate (6.08 g, 40 mmol, 1 eq) were refluxed at 90 ^{0}C for 18 hours. The solid was rinsed with hexane to obtain the pure product (5.025 g, 33.0 mmol, 83%) as a brown powder. The ^{1}H- and ^{13}C-NMR spectra as literature [21] at ^{1}H-NMR (500 MHz, DMSO-d_6) δ_H (ppm) 9.49 (br s, 1H), 7.67 (d, J = 8.5 Hz, 2H), 6.76 (d, J = 8.5 Hz, 2H). ^{13}C-NMR (125 MHz, DMSO-d_6) δ_C (ppm) 166.2, 160.8, 128.9, 123.5, 155.1.

*2.2.2 General Procedure for N'-Arylidene-4-hydroxybenzohydrazide (**AD-H$_{1-3}$**)*

4-Hydroxybenzoylhydrazine (153 mg, 1 mmol, 1 eq) was added to substituted-benzaldehyde (1 mmol, 1 eq) in EtOH (10 mL). The reaction mixture was refluxed for 18 hours. The organic solvent was evaporated and then rinsed using cooled EtOH to obtain the pure product.

(*E*)-4-hydroxy-*N'*-(3-hydroxy-4-methoxybenzylidene)benzohydrazide (**AD-H$_1$**)

Yield: 65%. The ^{1}H- and ^{13}C-NMR spectra as literature [22] at ^{1}H-NMR (500 MHz, DMSO-d_6) δ_H (ppm) 11.48 (s, 1H), 10.12 (s, 1H), 9.32 (s, 1H), 8.27 (s, 1H), 7.79 (d, J = 9.0 Hz, 2H), 7.26 (s, 1H), 7.03 (d, J = 8.0 Hz, 1H), 6.96 (d, J = 8.0 Hz, 1H), 6.85 (d, J = 8.5 Hz, 2H), 3.79 (s, 3H). ^{13}C-NMR (125 MHz, DMSO-d_6) δ_C (ppm) 162.6, 160.6, 149.7, 147.1, 146.9, 129.6, 127.4, 124.1, 120.2, 115.1, 112.3, 111.9, 55.6.

(*E*)-*N'*-(3,4-dihydroxybenzylidene)-4-hydroxybenzohydrazide (**AD-H$_2$**)

Yield: 33.7%. The ^{1}H- and ^{13}C-NMR spectra as literature [23] at ^{1}H-NMR (500 MHz, DMSO-*d*$_6$) δ_H (ppm) 11.42 (s, 1H), 10.08 (br s, 1H), 9.35 (br s, 2H), 8.23 (s, 1H), 7.78 (d, *J* = 8.5 Hz, 2H), 7.23 (s, 1H), 6.90 (dd, J = 8.0, 1.0 Hz, 1H), 6.84 (d, *J* = 8.5 Hz, 2H), 6.77 (d, *J* = 8.0 Hz, 1H). ^{13}C-NMR (125 MHz, DMSO-*d*$_6$) δ_C (ppm) 162.6, 160.6, 147.8, 147.5, 145.8, 129.6, 126.0, 124.2, 120.5, 115.6, 115.1, 112.6.

(*E*)-*N'*-(4-fluorobenzylidene)-4-hydroxybenzohydrazide (**AD-H$_3$**)

Yield: 37.5%. The ^{1}H- and ^{13}C-NMR spectra as literature [24] at ^{1}H-NMR (500 MHz, DMSO-*d*$_6$) δ_H (ppm) 9.51 (s, 1H), 8.72 (s, 1H), 7.94 (dd, J_{H-H}= 9.0 Hz, J_{H-F}= 5.5 Hz, 2H), 7.68 (d, *J* = 8.50 Hz, 2H), 7.35 (t, $J_{H-H\ \&\ H-F}$ = 9.0 Hz, 2H), 6.77 (d, *J* = 8.5 Hz, 2H). ^{13}C-NMR (125 MHz, DMSO-*d*$_6$) δ_C (ppm) 166.0, 160.6, 160.1, 130.8, 128.9, 123.9, 116.3, 116.1, 114.9.

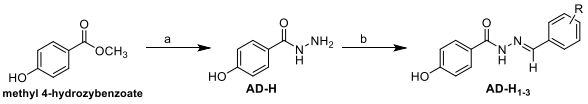
2.2.3 In Vitro α-Glucosidase Assay

According to the previous method [25], α-glucosidase assay has been conducted. The α-glucosidase (0.1 U/mL) and substrate (1 mM p-nitrophenyl-α-D-glucopyranoside) were dissolved in 0.1 M phosphate buffer (pH 6.9). A 10 µL test sample was pre-incubated with α-glucosidase (40 µL) at 37ºC for 10 min. A substrate solution (50 µL) was then added to the reaction mixture and incubated at 37ºC for an additional 20 min, and terminated by adding 1 M Na_2CO_3 solution (100 µL). Enzymatic activity was quantified by measuring the absorbance at 405 nm (ALLSHENG AMR-100 microplate reader). The percentage inhibition of activity was calculated as follows: % Inhibition = $[(A_0-A_1)/A_0] \times 100$, where: A_0 is the absorbance without the sample; A_1 is the absorbance with the sample. The IC_{50} value was deduced from the plot of % inhibition versus the concentration of the test sample. Acarbose was used as standard control and the experiment was performed in triplicate.

3 Result and Discussion

3.1 Chemistry

N'-Arylidene-4-hydroxybenzohydrazides (**AD-H$_{1-3}$**) were synthesized according to the previous work through two steps (Scheme 1) [21]. In the first step, 4-hydroxybenzoylhydrazine was successfully synthesized from methyl 4-hydroxybenzoate and hydrazine hydrate with a ratio of 1:8 in reflux condition for 18 hours to obtain 83% of the product yield. This product was formed from ester benzoic acid and hydrazine, because OH from benzoic acid was not a good leaving group. Thus, the carboxylic acid group was not easily substituted with hydrazine. In the second step, *N'*-Arylidene-4-hydroxybenzohydrazide was obtained from the reaction between 4-hydroxybenzohydrazide (**AD-H**) and substituted-benzaldehydes with a ratio of 1:1 in reflux condition for 18 hours to obtain the product (Figure 3) with the yield around 33-65%. However, this condition did not obtain a high yield compared to the previous work for 3 hours at 80^{0}C in ethanol solvent [23]. This result could be the hydrazone decomposition due to a long reaction of around 18 hours in reflux conditions.



Scheme 1. Reaction conditions. a) $N_2H_4.xH_2O$, 90^{0}C, 18 h; b) substituted-benzaldehydes, EtOH, reflux, 18 h.

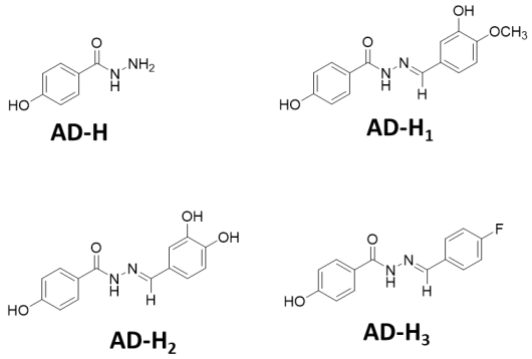


Figure 3. Four synthesized compounds (**AD-H** and **AD-H$_{1-3}$**).

All synthesized compounds were characterized using ^{1}H- and ^{13}C-NMR spectroscopy. **AD-H** and **AD-H$_2$** were selected to explain the structural elucidation. Compound **AD-H** (Figure 2) possessed the molecular formula $C_7H_8N_2O_2$. The spectra of ^{1}H-NMR of **AD-H** showed three proton signals at δ_H 9.49 (br s, 1H), 7.67 (d, *J* = 8.5 Hz, 2H), and 6.76 (d, *J* = 8.5 Hz, 2H). Moreover, the spectra of ^{13}C NMR of AD-H showed five carbon signals at δ_C 166.2 (C=O), 160.8 (=C-OH), 128.9 (2 x =CH), 123.5 (=Cq), and 155.1 (2 x =CH). Meanwhile, Compound **AD-H$_2$** showed the 1H-NMR spectra at δ_H 11.42 (s, 1H) belonged to the proton of amide (NH), while proton at δ_H 10.08 (br s, 1H), 9.33 (br s, 2H), and 8.23 (s, 1H) belonged

to the three hydroxy and an imine group. The two proton signals at δ_H 7.78 (d, *J* = 8.5 Hz, 2H) and 6.84 (d, *J* = 8.5 Hz, 2H) belonged to the benzohydrazide, while three proton signals at δ_H 7.23 (s, 1H), 6.90 (dd, *J* = 8.0, 1.0 Hz, 1H), 6.77 (d, *J* = 8.0 Hz, 1H) belonged to the arylidene. In addition, the spectra of ^{13}C-NMR of **AD-H$_2$** showed twelve carbon signals at δ_C 162.6 (C=O), 160.6 (=C-OH), 147.8 (=C-OH), 147.5 (N=CH), 145.8 (=C-OH), 129.6 (2 x =CH), 126.0 (=Cq), 124.2 (=Cq), 120.5 (=CH), 115.6 (=CH), 115.1 (2 x =CH), 112.6 (=CH).

3.2 Biological Evaluation

Four synthesized compounds (Figure 3) were screened for inhibition against the α-glucosidase enzyme according to the previous work [25]. As shown in Figure 4, all synthesized compounds showed less inhibitory activity against α-glucosidase, except for compound **AD-H$_2$**. Although, 4-hydroxybenzohydrazide (**AD-H, AD-H$_1$**, **AD-H$_3$**) showed less potency (25-50%) with inhibitory activity around 34.4%, 29.2% and 24.7%, respectively. However, this result was not in line with previous work using 2-hydroxybenzohydrazide as the starting material that showed moderate (50-75%) to high potency (75-100%) [20,26]. Therefore, 2-hydroxybenzohydrazide derivatives possessed good pharmacophores for α-glucosidase inhibitors. Nevertheless, compound **AD-H$_2$** possessing two hydroxy groups on arylidene moiety exhibited the best inhibitory activity against α-glucosidase around 83% in line with previous work [14,20], but the substitution of hydroxy with methoxy group decreased an inhibitory activity around 29.2%. The IC_{50} value of **AD-H$_2$** showed higher than acarbose as a positive control, such as 14.4 and 93.6 µM, respectively. This suggests that two hydroxy groups on the arylidene moiety might be crucial for the interaction between the inhibitor and amino residues in the active site of α-glucosidase.

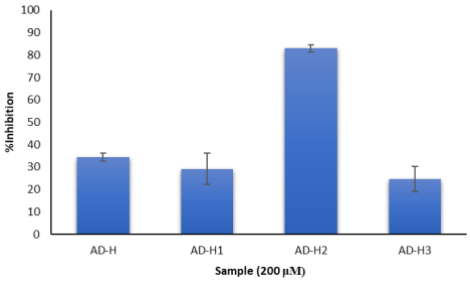


Figure 4. Screening of α-glucosidase inhibition of **AD-H** and **AD-H$_{1-3}$**.

4 Conclusion

In this study, four compounds (**AD-H**, **AD-H$_{1-3}$**) were successfully synthesized and characterized using ^{1}H-and ^{13}C-NMR spectroscopy. Among four compounds, **AD-H$_2$** showed the most potent antidiabetic against α-glucosidase. Therefore, it suggests that **AD-H$_2$** could be the lead candidate for the treatment of type 2 diabetes mellitus (T2DM).

Acknowledgment

Royal Golden Jubilee supported this work for the Ph.D. program (No. PhD/0017/2561), Thailand Research Fund.

References

[1] Forouhi, N.G. and Wareham, N.J. 2014. Epidemiology of diabetes. *Medicine (Abingdon, England : UK Ed)*, England. 42 (12) 698–702. 10.1016/j.mpmed.2014.09.007

[2] Kokil, G.R., Veedu, R.N., Ramm, G.A., Prins, J.B. and Parekh, H.S. 2015. Type 2 Diabetes Mellitus: Limitations of Conventional Therapies and Intervention with Nucleic Acid-Based Therapeutics. *Chemical Reviews*, American Chemical Society (ACS). 115 (11) 4719–43. 10.1021/cr5002832

[3] Kousaxidis, A., Petrou, A., Lavrentaki, V., Fesatidou, M., Nicolaou, I. and Geronikaki, A. 2020. Aldose reductase and protein tyrosine phosphatase 1B inhibitors as a promising therapeutic approach for diabetes mellitus. *European Journal of Medicinal Chemistry*, Elsevier BV. 207 112742. 10.1016/j.ejmech.2020.112742

[4] Kumar, K., Suebsuwong, C., Wang, P., Garcia-Ocana, A., Stewart, A.F. and DeVita, R.J. 2021. DYRK1A Inhibitors as Potential Therapeutics for β-Cell Regeneration for Diabetes. *Journal of Medicinal Chemistry*, American Chemical Society (ACS). 64 (6) 2901–22. 10.1021/acs.jmedchem.0c02050

[5] Padhi, S., Nayak, A.K. and Behera, A. 2020. Type II diabetes mellitus: a review on recent drug based therapeutics. *Biomedicine & Pharmacotherapy*, Elsevier BV. 131 110708. 10.1016/j.biopha.2020.110708

[6] Hossain, U., Das, A.K., Ghosh, S. and Sil, P.C. 2020. An overview on the role of bioactive α-glucosidase inhibitors in ameliorating diabetic complications. *Food and Chemical Toxicology : An International Journal Published for the British Industrial Biological Research Association*, 2020/09/09. England. 145 111738. 10.1016/j.fct.2020.111738

[7] Saini, R.K., Goyal, D. and Goyal, B. 2020. Targeting Human Islet Amyloid Polypeptide Aggregation and Toxicity in Type 2 Diabetes: An Overview of Peptide-Based Inhibitors. *Chemical Research in Toxicology*, American Chemical Society (ACS). 33 (11) 2719–38. 10.1021/acs.chemrestox.0c00416

[8] Ghani, U. 2015. Re-exploring promising α-glucosidase inhibitors for potential development into oral anti-diabetic drugs: Finding needle in the haystack. *European Journal of Medicinal Chemistry*, Elsevier BV. 103 133–62. 10.1016/j.ejmech.2015.08.043

[9] Rasouli, H., Yarani, R., Pociot, F. and Popović-Djordjević, J. 2020. Anti-diabetic potential of plant alkaloids: Revisiting current findings and future perspectives. *Pharmacological Research*, Elsevier BV. 155 104723. 10.1016/j.phrs.2020.104723

[10] El Shehry, M.F., Abbas, S.Y., Farrag, A.M., Eissa, S.I., Fouad, S.A. and Ammar, Y.A. 2018. Design, synthesis and biological evaluation of quinoxaline N-propionic and O-propionic hydrazide derivatives as antibacterial and antifungal agents. *Medicinal Chemistry Research*, Springer Science and Business Media LLC. 27 (10) 2287–96. 10.1007/s00044-018-2235-4

[11] Kumar, A.K.A., Bodke, Y.D., Sambasivam, G. and Lakra, P.S. 2017. Design, synthesis, and evaluation of novel hydrazide hydrochlorides of 6-aminopyrazolo[1,5-a]pyrimidine-3-carboxamides as potent Aurora kinase inhibitors. *Monatshefte Für Chemie - Chemical Monthly*, Springer Science and Business Media LLC. 148 (10) 1767–80. 10.1007/s00706-017-1943-7

[12] Ahmed, W., Rani, M., Khan, I.A., Iqbal, A., Khan, K.M., Haleem, M.A. et al. 2010. Characterisation of hydrazides and hydrazine derivatives as novel aspartic protease inhibitors. *Journal of Enzyme Inhibition and Medicinal Chemistry*, Informa UK Limited. 25 (5) 673–8. 10.3109/14756360903508430

[13] Liang, J.-W., Li, S.-L., Wang, S., Li, W.-Q. and Meng, F.-H. 2020. Synthesis and biological evaluation of novel (E)-N'-benzylidene hydrazides as novel c-Met inhibitors through fragment based virtual screening. *Journal of Enzyme Inhibition and Medicinal Chemistry*, England. 35 (1) 468–77. 10.1080/14756366.2019.1702655

[14] Rahim, F., Taha, M., Ullah, H., Wadood, A., Selvaraj, M., Rab, A. et al. 2019. Synthesis of new arylhydrazide bearing Schiff bases/thiazolidinone: α-Amylase, urease activities and their molecular docking studies. *Bioorganic Chemistry*, Elsevier BV. 91 103112. 10.1016/j.bioorg.2019.103112

[15] Taha, M., Ismail, N.H., Javaid, K., Imran, S., Anouar, E.H., Wadood, A. et al. 2015. Evaluation of 2-indolcarbohydrazones as potent α-glucosidase inhibitors, in silico studies and DFT based stereochemical predictions. *Bioorganic Chemistry*, Elsevier BV. 63 24–35. 10.1016/j.bioorg.2015.09.001

[16] Taha, M., Rahim, F., Imran, S., Ismail, N.H., Ullah, H., Selvaraj, M. et al. 2017. Synthesis, α -glucosidase inhibitory activity and in silico study of tris -indole hybrid scaffold with oxadiazole ring: As potential leads for the management of type-II diabetes mellitus. *Bioorganic Chemistry*, Elsevier BV. 74 30–40. 10.1016/j.bioorg.2017.07.009

[17] Wang, G., Chen, M., Wang, J., Peng, Y., Li, L., Xie, Z. et al. 2017. Synthesis, biological evaluation and molecular docking studies of chromone hydrazone derivatives as α -glucosidase inhibitors. *Bioorganic & Medicinal Chemistry Letters*, Elsevier BV. 27 (13) 2957–61. 10.1016/j.bmcl.2017.05.007

[18] Wang, G., He, D., Li, X., Li, J. and Peng, Z. 2016. Design, synthesis and biological

evaluation of novel coumarin thiazole derivatives as α-glucosidase inhibitors. *Bioorganic Chemistry*, Elsevier BV. 65 167–74. 10.1016/j.bioorg.2016.03.001

[19] Hu, C.-M., Zheng, Y.-Y., Lin, A.-T., Zhang, X., Wu, X.-Z., Lin, J. et al. 2023. Design, synthesis and evaluation of indole-based bisacylhydrazone derivatives as α-glucosidase inhibitors. *Journal of Molecular Structure*, Elsevier BV. 1271 134124. 10.1016/j.molstruc.2022.134124

[20] Ullah, H., Uddin, I., Rahim, F., Khan, F., Sobia, Taha, M. et al. 2022. In vitro α-glucosidase and α-amylase inhibitory potential and molecular docking studies of benzohydrazide based imines and thiazolidine-4-one derivatives. *Journal of Molecular Structure*, Elsevier BV. 1251 132058. 10.1016/j.molstruc.2021.132058

[21] Leigh, D.A., Marcos, V., Nalbantoglu, T., Vitorica-Yrezabal, I.J., Yasar, F.T. and Zhu, X. 2017. Pyridyl-Acyl Hydrazone Rotaxanes and Molecular Shuttles. *Journal of the American Chemical Society*, American Chemical Society (ACS). 139 (20) 7104–9. 10.1021/jacs.7b03307

[22] Fun, H.-K., Horkaew, J. and Chantrapromma, S. 2011. (E)-4-Hy-droxy-N'-(3-hy-droxy-4-meth-oxy-benzyl-idene)benzohydrazide. *Acta Crystallographica Section E, Structure Reports Online*, 2011/09/14. United States. 67 (Pt 10) o2644–5. 10.1107/S1600536811036579

[23] Branković, J., Milivojević, N., Milovanović, V., Simijonović, D., Petrović, Z.D., Marković, Z. et al. 2022. Evaluation of antioxidant and cytotoxic properties of phenolic N-acylhydrazones: structure-activity relationship. *Royal Society Open Science*, England. 9 (6) 211853. 10.1098/rsos.211853

[24] Parisi, E., Borbone, F., Carella, A., Lettieri, S., Capobianco, A., Peluso, A. et al. 2023. Winning Strategy toward Acentric Crystals: Transverse Dipole Moment Molecules. *Crystal Growth & Design*, American Chemical Society (ACS). 23 (6) 4538–44. 10.1021/acs.cgd.3c00295

[25] Le, T.-K.-D., Danova, A., Aree, T., Duong, T.-H., Koketsu, M., Ninomiya, M. et al. 2022. α-Glucosidase Inhibitors from the Stems of *Knema globularia*. *Journal of Natural Products*, American Chemical Society (ACS). 85 (4) 776–86. 10.1021/acs.jnatprod.1c00765

[26] Gholamhose, A., Fallah, H., Sharifi-Fa, F. and Mirtajaddi, M. 2008. Alpha Mannosidase Inhibitory Effect of Some Iranian Plant Extracts. *International Journal of Pharmacology*, Science Alert. 4 (6) 460–5. 10.3923/ijp.2008.460.465