J. Electrochem. Sci. Eng. **00(0)** (2025) 2867; http://doi.org/10.5599/jese.2867

Open Access : ISSN 1847-9286
www.jESE-online.org

Short communication

Electrochemical assessment of α-amylase inhibition by type 2 diabetes drugs: a cyclic voltammetry study

Touhami Lanez[1,✉], Elhafnaoui Lanez[2] and Aicha Adaika[2]

[1]*University of El Oued, Faculty of Sciences, Chemistry Department, VTRS Laboratory, El Oued, Algeria*

[2]*University of El Oued, Faculty of Nature and Life Sciences, Cellular and Molecular Biology, Department, VTRS Laboratory, El Oued, Algeria*

Corresponding authors: ✉touhami-lanez@univ-eloued.dz

Received: July 1, 2025; Accepted: October 28, 2025; Published: November 2, 2025

Abstract

A new sensitive cyclic voltammetry method was developed for measuring the α-amylase inhibitory activity of six antidiabetic drugs commonly prescribed for type 2 diabetes: acarbose, amaryl, repaglinide, diamecron, dapagliflozin, and metformin. The electrochemical response of an enzyme-starch mixture was measured in the absence and presence of these drugs. Results indicated varying inhibitory potencies with IC_{50} *values ranging from 6.770 to 41.423 µM, with acarbose as the most potent. UV-Vis absorption spectroscopy at 580 nm validated the cyclic voltammetry results, with very good concordance. This study highlights the utility of electrochemical assays for the rapid, quantitative screening of α-amylase inhibitors and supports their integration with spectroscopic methods for the evaluation of antidiabetic drugs.*

Keywords
Enzyme-starch mixture; IC_{50}; electrochemical assay; antidiabetic agents

Introduction

Diabetes mellitus is a chronic metabolic disease characterized by prolonged hyperglycemia resulting from defects in insulin secretion, insulin action, or both [1,2]. Postprandial hyperglycemia is an especially significant contributor to the long-term diabetic microvascular and macrovascular complications [3-5]. A therapeutic strategy to reduce this glucose increase is the inhibition of enzymes involved in carbohydrate digestion, especially α-amylase and α-glucosidase, which hydrolyze dietary starch into absorbable monosaccharides [6-8]. Through the prevention of starch hydrolysis, α-amylase inhibitors suppress the rate of glucose absorption, enabling a more gradual and sustained postprandial blood glucose increase [9-12].

Quantitation of α-amylase inhibitors has traditionally been done by colorimetric or spectrophotometric assays, the most commonly used being the Lugol's method for determining released reducing sugar from starch [13]. Though these are well-established and widely available assays, they are typically characterized by long reaction times, the need for sequential reagent addition, and susceptibility to interference from pigmented or turbid samples [14]. By contrast, electrochemical techniques such as cyclic voltammetry (CV) offer several advantages: rapid data acquisition, sensitivity to redox-active species, and the potential for miniaturized, online sensors [15,16].

Recent studies have demonstrated the promise of CV for enzyme-inhibitor screening [16]. For instance, in situ electrochemical and spectroscopic methods have been used to characterize the inhibitory activity of natural compounds and low-molecular-weight drugs towards hydrolases and oxidases [17,18]. Reports focusing on antidiabetic agents so far are few. Clinically used α-amylase inhibitors (*e.g.* acarbose, miglitol) and sulfonylurea drugs (*e.g*. glyburide, glimepiride) are structurally, mechanistically, and physicochemically heterogeneous but have scarcely been compared directly under electrochemical conditions [19,20].

Here, we set up and validate a CV method for assessing the α-amylase inhibitory activity of six commonly prescribed antidiabetic drugs for type 2 diabetes. We optimize assay parameters, such as enzyme concentration, substrate loading, and scan rate, to achieve maximum sensitivity and reproducibility. Electrochemical results are validated using UV-Vis assays; this new approach aims to render CV a rapid and reliable screening tool for antidiabetic drugs.

Experimental

Chemicals and reagents

All compounds, including porcine pancreatic α-amylase, Lugol solution (Sigma-Aldrich), starch, and pharmaceutical standards (Merck), were used without further purification.

Drug procurement and preparation

The six antidiabetic drugs used in this study, acarbose, Amaryl, repaglinide, Diamecron, dapagliflozin, and metformin, were procured as pharmaceutical-grade tablets from licensed community pharmacies in Guemar, Algeria. Each product was obtained in its commercially available formulation. Upon receipt, tablets were stored at 4 °C in their original blister packs, protected from light and humidity, until use. Prior to assay, each drug was finely ground, and a representative sample was analyzed by UV absorbance to confirm identity and approximate purity in accordance with pharmacopeial standards.

Preparation of Solutions

The α-amylase solution was prepared by mixing 50 mg of α-amylase with 100 mL of a 0.1 M aqueous phosphate buffer solution at pH 6.4. Similarly, the starch solution was prepared by stirring 200 mg of starch in 100 mL of the same buffer solution at the same pH value. The Lugol solution was prepared by dissolving 1mg of iodine-potassium iodide in 100 mL.

Evaluation of α-amylase inhibition

To evaluate the inhibitory potential of the selected test drugs, 200 µL of each drug solution (prepared in 0.1 M phosphate buffer, final concentrations: 7.8, 15.6, 31.2, 62.5, 125, 250 and 500 µM) was mixed with 500 µL of α-amylase solution and incubated at 37 °C for 30 minutes to allow enzyme-inhibitor interaction. Subsequently, 100 µL of a 2 % starch solution was added to initiate the enzymatic reaction, and the mixture was further incubated at 37 °C for an additional 30 minutes.

The reaction was terminated by adding 50 µL of 1 M HCl, followed by 100 µL of iodine-potassium iodide solution, and then heating in a boiling water bath for 5 minutes. After cooling to room temperature, the mixture was diluted to a final volume of 10 mL with distilled water prior to CV measurement.

For each drug, blank tubes were prepared by replacing the enzyme solution with 500 µL of 2 % starch solution in distilled water. The experiments were repeated three times using the same protocol. The α-amylase inhibitory potential was expressed as the inhibition, %. The inhibition was calculated using Equation (1).

$$\text{Inhibition} = \frac{j\text{-}j_0}{j}100 \qquad (1)$$

where, j_0 represents the cathodic peak current density of the control (100 % α-amylase activity), and j is the cathodic peak current density in the presence of increasing concentration of the test drugs.

Cyclic voltammetry assay

Electrochemical measurements were performed at 28±2 °C using a PGZ301 voltammeter, operated with VoltaMaster 4 V 7,08 software (Radiometer Analytical SAS, France). The experimental setup consisted of a three-electrode electrochemical cell with a 10 mL capacity, featuring a glassy carbon (GC) working electrode with a geometric area of 0.013 cm², a platinum wire as the counter electrode, and an Hg/Hg_2Cl_2 paste-covered wire as the reference electrode.

UV-Vis spectroscopic validation

Absorption spectra were measured on a UV-Vis spectrometer (Shimadzu, Japan, model 1800). The measurements were conducted in a 2 ml cell with an optical path length of 10 mm. These parallel inhibition assays employed the Lugol method [21,22], monitoring absorbance at 580 nm to calculate percent inhibition relative to drug-free controls.

α-amylase inhibition assay

Cyclic voltammetry technique

The α-amylase inhibitory activity was assessed using a modified Caraway-Somogyi iodine/potassium iodide (IKI) method [23,24], adapted for electrochemical detection. The procedure was based on protocols established by Yang *et al.* [25] and Zengin *et al.* [26], with minor modifications to optimize assay sensitivity. Briefly, 25 µL of the test sample was mixed with 50 µL of α-amylase solution (prepared in phosphate buffer, pH 6.4, containing 6 mM sodium chloride) and incubated at 37 °C for 10 minutes to allow for enzyme interaction. The reaction was then initiated by adding 100 µL of a 2 % (w/v) starch solution as the substrate.

A corresponding enzyme-free blank was prepared under identical conditions, omitting the α-amylase. After 10 minutes of incubation at 37 °C, the enzymatic reaction was terminated by adding 50 µL of 1 M hydrochloric acid, followed by 100 µL of IKI reagent. The resulting mixture was then subjected to cyclic voltammetry using a three-electrode setup (glassy carbon working electrode, Pt counter electrode, Hg/Hg_2Cl_2 reference electrode). Voltammograms were recorded in the range -0.2 to +0.8 V at a scan rate of 50 mV s^{-1}. The anodic peak current of the I^-/I_3^- redox couple was monitored, with decreased current intensity indicating higher inhibition. IC_{50} values were calculated as the inhibitor concentration required to suppress 50 % of α-amylase activity.

Electronic spectroscopy techniques

The effect of the drugs on α-amylase activity was measured using the method described by Yang *et al.* [25] and Zengin *et al.* [26]. This was achieved by determining the absorbance at 580 nm to assess α-amylase activity and calculating the percentage decrease in the maltose production rate as a measure of α-amylase inhibitory activity.

Electrochemical characterization of Lugol's solution

In the current research, Lugol's solution, characterized by its electroactive iodide ions, was chosen for its advantageous redox properties and enhanced sensitivity under moderate experimental conditions. Figure 1 presents a cyclic voltammogram of iodide in a 1 % Lugol solution, obtained using a glassy carbon electrode dipped in 0.1 M phosphate buffer (pH 6.4) at a scan rate of 0.1 V s^{-1}, in a range of -0.1 to +1.2 V, under conditions maintained at 28 °C.

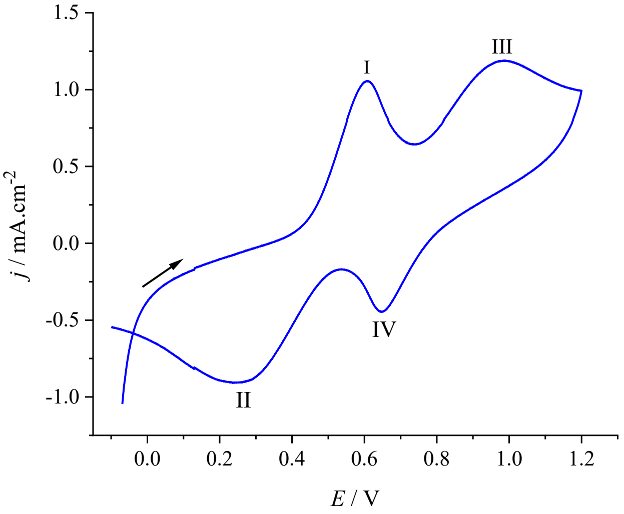

Figure 1. *Electrochemical response of iodide in 1 % Lugol's solution recorded at 0.1 V s^{-1} potential sweep rate on GC disk electrode at 301 K in a 1.0 M phosphate buffer solution at pH 6.4*

Under these optimized conditions, four distinct redox peaks were observed: two anodic (I and III) and two cathodic (II and IV). The first anodic peak (E_{paI} = 0.608 V) and its corresponding cathodic peak (E_{pcII} = 0.251 V) represent the iodide/triiodide (I^-/I_3^-) redox couple. The second anodic peak (E_{paIII} = 0.976 V) and cathodic peak (E_{pcIV} = 0.648 V) correspond to the iodine/iodide (I_2/I^-) redox couple. These two pairs of redox peaks indicate a two-step quasi-reversible electrochemical process involving iodide species, in agreement with reported mechanisms of iodide electrochemistry [27-29].

As described above, iodide exhibited two quasi-reversible anodic-cathodic peak pairs (I and II), corresponding to the iodide/triiodide and iodine/iodide redox couples, Equation (2):

$$3I^- \leftrightarrow I^{3-} + 2e^- \tag{2}$$

while the second pair (III and IV) is attributed to the iodine/iodide redox process, Equation (3):

$$I_2 + 2e^- \leftrightarrow 2I^- \tag{3}$$

By confirming that iodide dominates the redox response under the selected conditions, we established a reliable baseline for evaluating how α-amylase activity and its inhibition by antidiabetic drugs modulate the electrochemical signal. This validation step allows us to attribute subsequent signal changes specifically to enzyme-inhibitor interactions mediated through iodide chemistry.

To further confirm the electrochemical nature of the iodide system, diagnostic tests for semi-reversibility were performed according to the criteria outlined by Greef *et al.* [30]. The analysis of peak separation and scan-rate dependence supported the quasi-reversible behavior of the two redox couples observed. Furthermore, control experiments were performed with Lugol's solution and each drug in the absence of α-amylase. These control voltammograms showed no significant modification of the iodide redox profile, thereby confirming that the variations observed in subsequent experiments arise from enzyme-drug interactions rather than from direct drug-iodine reactions.

Data analysis

Linear regression of inhibition versus concentration yielded IC_{50} values. Statistical significance was assessed via ANOVA with Tukey's *post-hoc* test ($p < 0.05$).

Results and discussion

Having confirmed that iodide is the only electroactive species within the selected potential range, this finding serves as the foundation for a novel electrochemical strategy to assess α-amylase inhibition, a key indicator of antidiabetic activity. Building on this, we investigated the electrochemical behaviour of a starch-amylase-iodine reaction system using cyclic voltammetry. The reaction mixture was composed of 100 µL starch solution (2 % w/v), 500 µL α-amylase solution (0.5 % w/v), and 1 mL of Lugol's iodine solution (5 % w/v), prepared in 10 mL of 0.1 M phosphate buffer (pH 6.4) with 6.7 mM NaCl added to simulate physiological ionic strength.

Cyclic voltammetry experiments were conducted both in the absence and in the presence of acarbose (50 µg mL^{-1}), a known α-amylase inhibitor, to examine its effect on the redox profile of the system. The scans were performed over a potential range of -0.1 to +1.2 V at a scan rate of 0.1 V s^{-1}. The observed changes in current response reflect modulation of enzyme activity by the inhibitor and are illustrated in Figure 2, highlighting the viability of this electrochemical system for monitoring enzymatic inhibition.

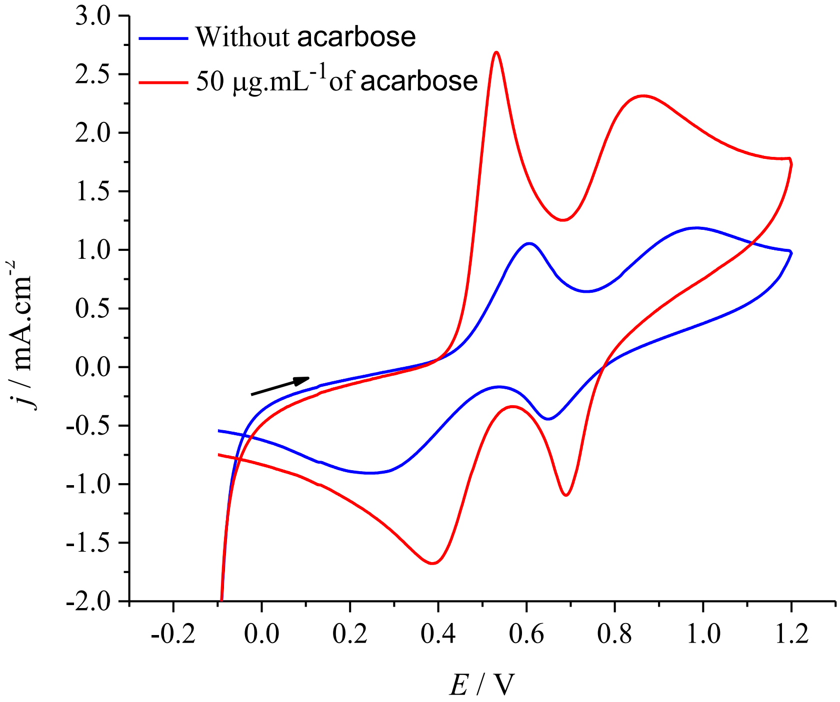

Figure 2. *Cyclic voltammograms of the starch-amylase-iodine system recorded at 0.1 V s^{-1} potential sweep rate on GC electrode at 301 K in 0.1 M phosphate buffer (pH 6.4), showing the control trace without inhibitor (blue) and the trace in the presence of acarbose (red)*

The voltammetric data in Figure 2 clearly indicate that the presence of acarbose results in a notable increase in current density at both the anodic and cathodic iodide peaks. Additionally, a slight negative shift in the anodic peak potential was observed, suggesting subtle changes in the redox environment. Importantly, no new peaks emerged following the addition of acarbose, reinforcing the conclusion that iodide from Lugol's solution remains the sole electroactive species within the studied potential window.

The observed enhancement in current density is attributed to a greater extent of iodide oxidation, which correlates with the inhibition of α-amylase activity by acarbose. As a result, more residual starch remains unhydrolyzed, allowing it to form an electroactive complex with Lugol's iodine, which intensifies the redox signal. This mechanism highlights the viability of using iodide electroactivity as an indirect reporter for monitoring enzymatic inhibition.

Building upon this preliminary validation, the present study expands the approach to evaluate the α-amylase inhibitory potential of six commonly prescribed antidiabetic drugs: acarbose, amaryl, repaglinide, diamecron, dapagliflozin, and metformin.

To evaluate the kinetics of inhibition, we investigated the effect of incubation time on the electrochemical response of the starch-amylase-iodine system in the presence of acarbose. Cyclic voltammograms were recorded after 0, 10, 20 and 30 minutes of incubation at 37 °C. As shown in Figure 3, the cathodic peak current shows a slight decrease within the first 10 minutes, after which no further significant change was observed. The voltammogram shapes remained stable, with peak positions essentially unchanged beyond this period. These results indicate that the inhibitory interaction between α-amylase and acarbose proceeds rapidly and reaches equilibrium within 10 minutes or less. Therefore, a 10-minute pre-incubation was adopted as the standard condition in subsequent assays.

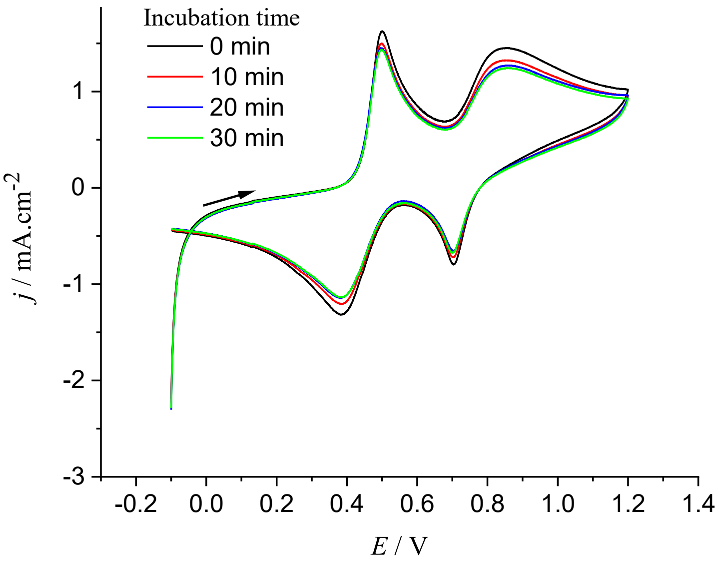

Figure 3. *Time-dependent cyclic voltammograms of the starch-amylase-iodine system in the presence of acarbose, recorded after 0, 10, 20 and 30 minutes of incubation at 37 °C*

Evaluation of the antidiabetic potential

The study of the cathodic behavior of starch dissociation by α-amylase (Figure 3), in the absence and presence of increasing concentrations of the drugs, was used to determine the antidiabetic potential of α-amylase in combination with the studied drugs. This study of the electrochemical behavior was conducted using cyclic voltammetry in a 0.1 M phosphate buffer solution at pH 6.4, with a potential window ranging from -0.1 to +1.2 V and a scan rate of 0.1 V s^{-1}. Cyclic voltammograms were initially started at -0.1 V and reversed at +1.2 V.

As the concentration of the tested drugs increased, the cathodic peak current density increased in a stepwise manner (Figure 4). However, the starch redox system remained reversible throughout the experiment.

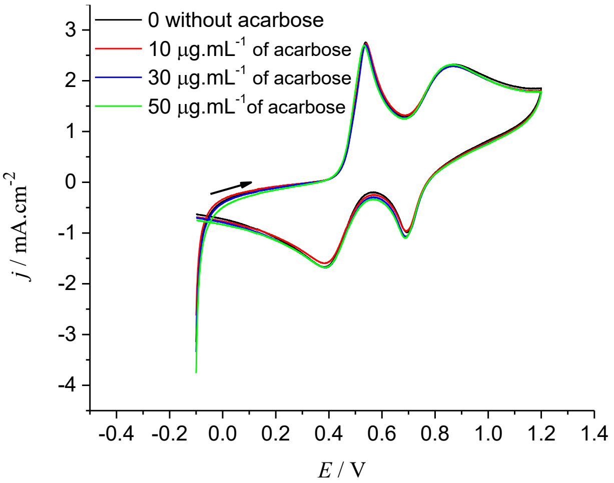

Figure 4. *Cyclic voltammograms of starch-Lugol solution recorded in the presence of acarbose in the absence of α-amylase.*

Control experiments

To ensure that the changes observed were not due to direct electrochemical activity of the drugs, additional control experiments were conducted under identical conditions: (i) Lugol's solution with each drug in the absence of α-amylase, (ii) α-amylase with each drug in the absence of Lugol, and (iii) starch with each drug in the absence of both Lugol and enzyme. In all cases, the voltammetric profiles showed no significant differences compared to the respective controls, confirming that the drugs do not directly catalyze iodine electrochemistry or contribute additional electroactive signals. Notably, α-amylase alone does not exhibit an electrochemical response within the studied potential range; therefore, the corresponding voltammograms are not presented. The obtained voltammograms for other control tests are presented in Figures 4 and 5. Altogether, these results demonstrate that the variations observed in the leading figures are exclusively due to enzyme-drug interactions.

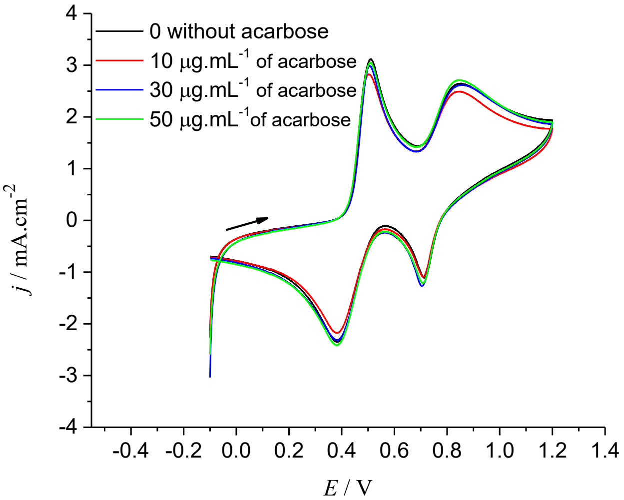

Figure 5*. Cyclic voltammograms of α-amylase-Lugol solution in phosphate-buffered saline solution recorded at 0.1 V·s^{-1} potential sweep rate on GC disk electrode at 301 K in the absence and presence of acarbose*

The voltammograms in Figure 5 illustrate the redox behavior of the α-amylase-Lugol system in the absence and presence of acarbose, confirming that the enzyme itself does not significantly alter the iodide response. This experiment establishes a clean electrochemical baseline for subsequent assays involving the full starch-amylase-iodine mixture. Building on this control, the following set of voltammograms (Figure 6) presents the complete reaction system, enabling direct visualization of how increasing drug concentrations modulate the enzyme-catalyzed redox signals.

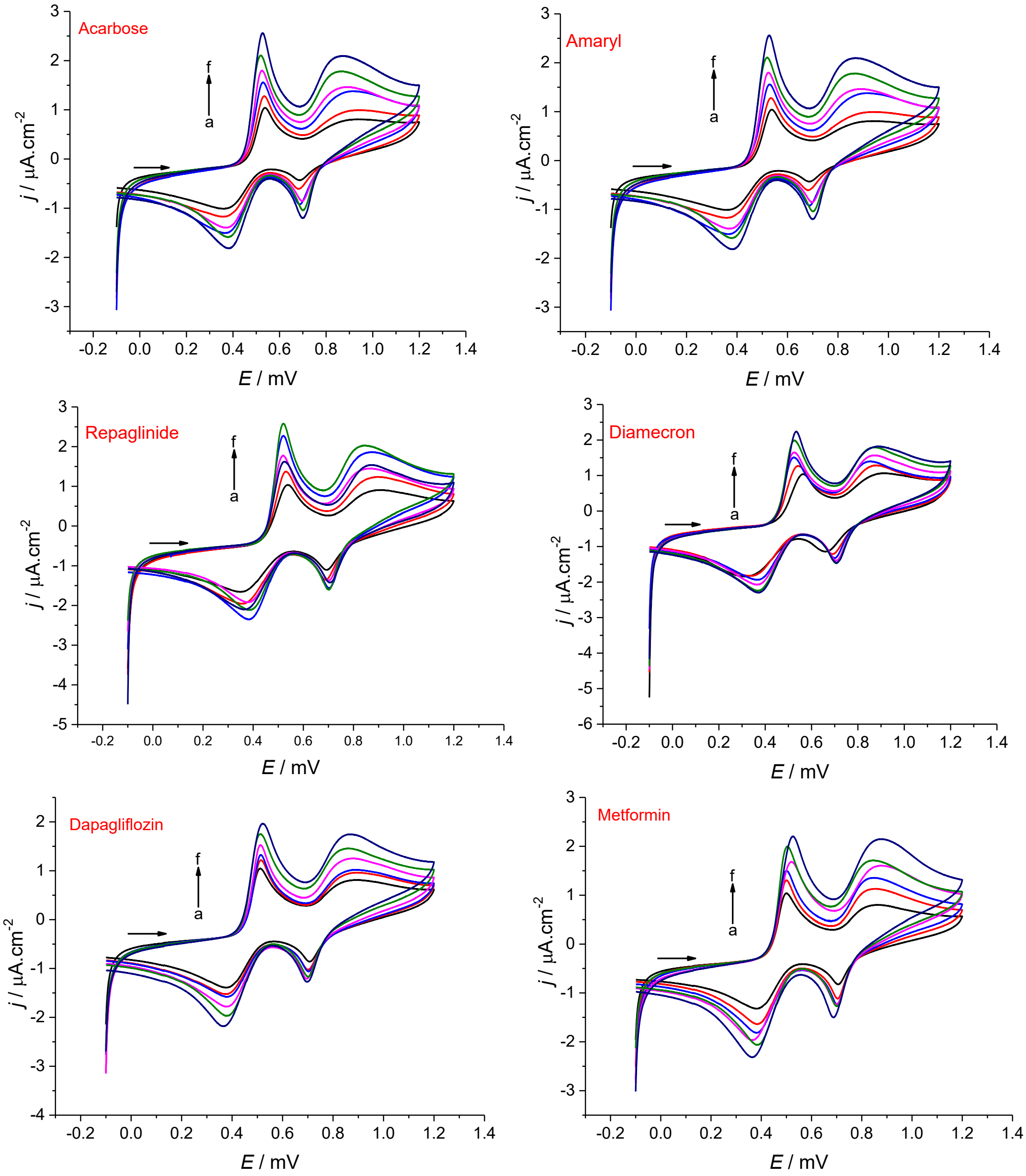

***Figure 6**. Cyclic voltammograms of starch-amylase-iodine reaction in phosphate-buffered saline solution recorded at 0.1 V s^{-1} potential sweep rate on GC disk electrode at 301 K in the absence of drug (a) and presence of 10 µg (b), 20 µg (c), 30 µg (d), 40 µg (e), and 50 µg (f) of different drugs*

While Figure 3 provides visual evidence of the inhibition of α-amylase activity by the selected antidiabetic drugs in cyclic voltammetry profiles, quantitative insight is gained by plotting the inhibition degree against drug concentration. Figure 7 presents such dose-response calibration curves, presenting the percent inhibition of α-amylase against the concentration of each studied drug. While Figure 3 provides visual evidence of the inhibition of α-amylase activity by the selected antidiabetic drugs in cyclic voltammetry profiles, quantitative insight is gained by plotting the inhibition degree against drug concentration. Figure 7 presents such dose-response calibration curves, presenting the percent inhibition of α-amylase against the concentration of each studied drug. These curves allow determination of IC_{50} values, the drug concentration at which 50 % inhibition is achieved, by linear regression. Each curve's slope and shape determine the relative drug potency and serve as the basis for comparing their inhibitory potential under controlled electrochemical conditions.

To obtain quantitative values with precision from the inhibition curves in Figure 7, the regression equations and corresponding correlation coefficients (R^2) were calculated.

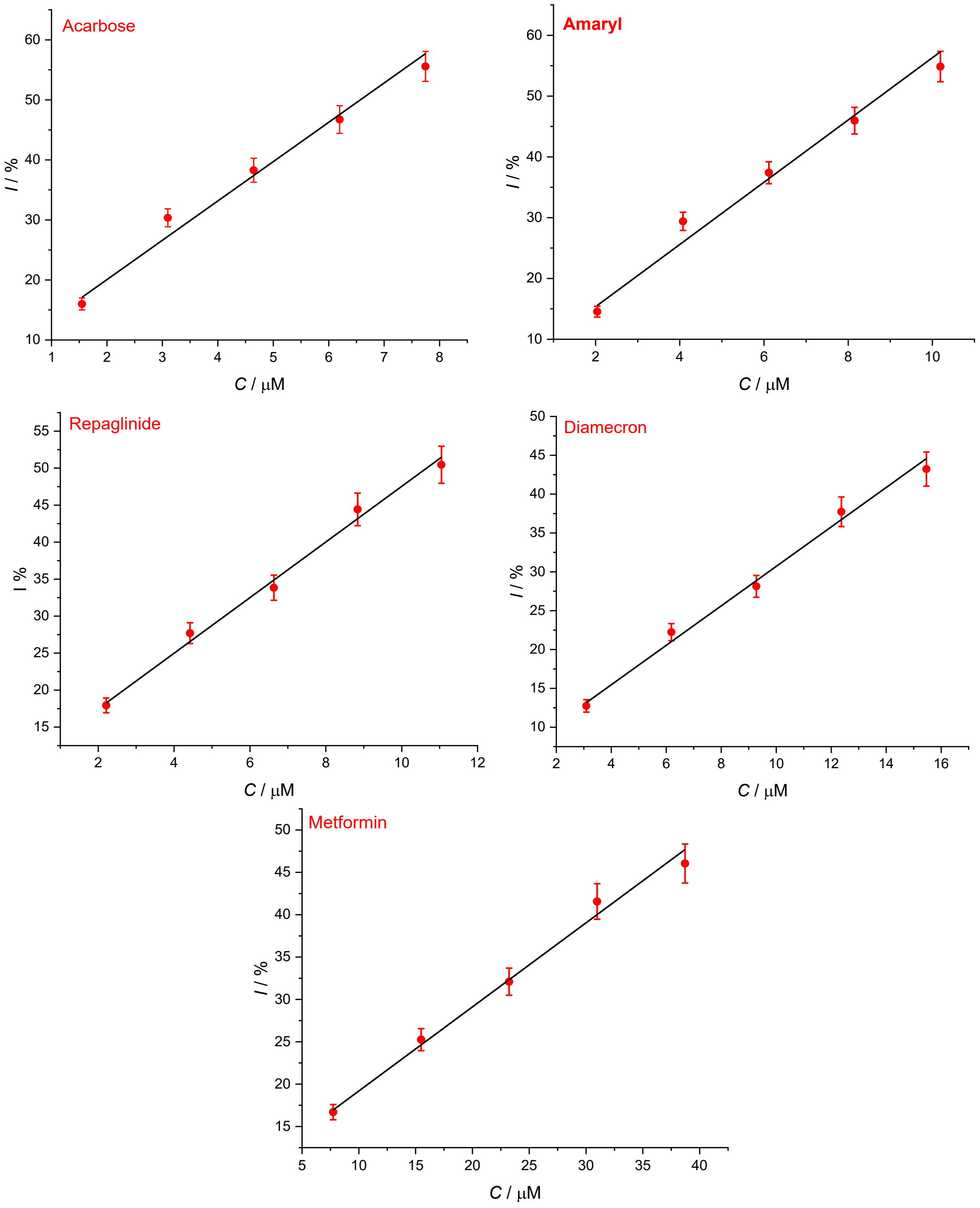

Figure 7. *Concentration-dependent inhibition of α-amylase by the tested drugs determined by cyclic voltammetry. Data points represent mean inhibition values ± standard deviation from three independent experiments (*n *= 3)*

These are summarized in Table 1, which includes the calculated IC_{50} values for each drug, based on the dose-response relationships developed using cyclic voltammetry.

Table 1. *IC_{50} values for antidiabetic drugs obtained using cyclic voltammetry assays*

Entry	Equation	R^2	IC_{50}, µM
Acarbose	$y = 6.167x + 8.731$	0.981	6.770 ± 0.372
Amaryl	$y = 4.768x + 7.278$	0.979	8.956 ± 0.464
Repaglinide	$y = 3.701x + 10.336$	0.990	10.720 ± 0.306
Diamecron	$y = 2.473x + 5.876$	0.989	17.844 ± 0.989
Dapagliflozin	$y = 3.071x + 2.701$	0.995	15.407 ± 0.524
Metformin	$y = 0.969x + 9.823$	0.987	41.423 ± 0.676

This table not only enables direct comparison of inhibitory capacity among drugs but also demonstrates the method's reliability, with high linear correlation values. Lower IC_{50} values for drugs indicate greater inhibitory activity against α-amylase, which is significant for the treatment of postprandial glucose in type 2 diabetes.

Validation of the CV results using UV-Vis spectrophotometric measurements

To validate the inhibitory results obtained from CV measurements, a parallel assessment of α-amylase inhibition by the selected antidiabetic drugs was carried out using UV-Vis spectrophotometric techniques. For this purpose, a reaction system was prepared containing 2 % w/v starch, 0.5 % w/v α-amylase, and 5 % w/v Lugol's iodine solution, all dissolved in 0.1 M phosphate buffer (pH 6.4) supplemented with 6.7 mM sodium chloride to mimic physiological ionic strength. Each drug was introduced incrementally at increasing concentrations, enabling the observation of dose-dependent inhibitory effects as reflected in absorbance changes. This approach ensures comparative validation of the electrochemical data and supports the reproducibility of α-amylase inhibition trends across two independent analytical platforms.

The resulting solutions were analyzed spectrophotometrically at 580 nm, the characteristic wavelength at which the Lugol-iodide-starch complex exhibits peak absorbance. A gradual increase in absorbance intensity was observed with increasing drug concentration, indicating greater retention of unhydrolyzed starch due to enzyme inhibition. This absorbance change forms the basis for calculating the percentage of α-amylase inhibition, as described in Equation (4). The approach provides a quantitative and visual means to assess the extent to which each compound inhibits enzymatic starch breakdown.

$$\text{Inhibition} = \left(\frac{A - A_0}{A}\right)100 \tag{4}$$

where A_0 represents the absorbance of the negative control (enzyme activity in the absence of the inhibitor), and A is the absorbance of the test assay (enzyme activity in the presence of the inhibitor).

Figure 8 is the α-amylase inhibition plots yielded by UV-Vis spectrophotometric measurements through the Lugol's method colorimetric assay. The plots show a decrease in α-amylase activity with increasing concentrations of the antidiabetic drugs, consistent with CV-based inhibition plots. The eye-fit correspondence between curves in Figures 7 and 8 is applied to confirm the reproducibility and quality of the inhibition trends, thereby establishing the use of cyclic voltammetry as a screening method for enzyme inhibition.

To systematically compare the inhibitory profiles obtained through spectrophotometry, Table 2 summarizes the corresponding regression equations, R^2 values, and IC_{50} values calculated from the UV-Vis inhibition curves shown in Figure 8. This Table enables a direct side-by-side comparison of the IC_{50} values obtained via both analytical methods, cyclic voltammetry and spectrophotometry, highlighting any variations in drug potency measurements.

Table 2. *IC_{50} values for antidiabetic drugs obtained using UV-Vis absorbance measurements*

Entry	Equation	R^2	IC_{50}, µg mL^{-1}
Acarbose	$y = 4.741x + 19.857$	0.916	6.359 ± 0.37
Amaryl	$y = 3.443x + 19.930$	0.973	8.734 ± 1.19
Repaglinide	$y = 3.444x + 14.210$	0.978	10.395 ± 0.74
Diamecron	$y = 2.328x + 8.992$	0.998	17.601 ± 1.31
Dapagliflozin	$y = 3.174x + 1.715$	0.995	15.213 ± 1.09
Metformin	$y = 1.051x + 6.972$	0.970	40.940 ± 1.05

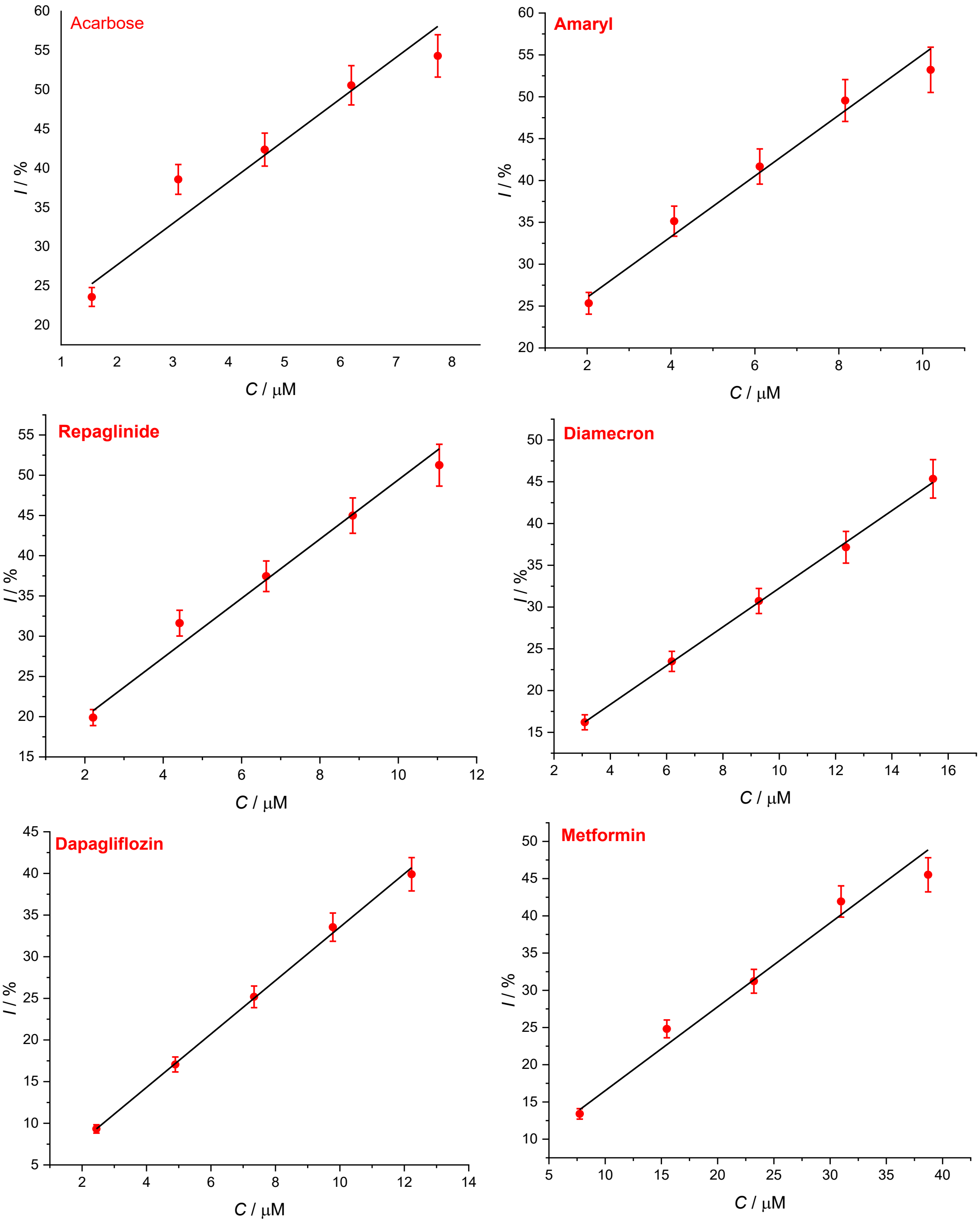

Figure 8. *Concentration-dependent inhibition of α-amylase by the tested drugs determined by UV-Vis spectrophotometry. Data points represent mean inhibition values ± standard deviation from three independent experiments (n = 3)*

The consistent patterns across both techniques underscore the reproducibility of the results and reinforce the potential of electrochemical methods as complementary or alternative approaches for assessing α-amylase inhibition.

The cyclic voltammetry assay proved to be a sensitive and efficient technique for determining α-amylase inhibition, as evidenced by sharp linear responses and precise IC_{50} estimation (Table 1). Among the tested compounds, acarbose exhibited the strongest inhibition with an IC_{50} of 6.77 μM, followed closely by amaryl (8.956 μM) and repaglinide (10.720 μM). Metformin, in contrast, showed the weakest inhibitory activity (IC_{50} = 41.423 μM), consistent with its known mechanism of action, which does not directly target α-amylase.

The UV-Vis absorbance method using the Lugol's assay confirmed these findings, with IC_{50} values that trended and were nearly identical in magnitude (Table 2). The eye-fit similarity between the inhibition plots in Figures 7 and 8 further supports the reproducibility and validity of the voltametric method. The consistent correlation coefficients (with most R^2 values exceeding 0.97) across both techniques reflect robust assay performance and minimal variation.

Importantly, both datasets demonstrate a direct correlation between IC_{50} values and the antidiabetic drug class or mechanism. Drugs known to interact with carbohydrate metabolism (*e.g*. acarbose, a known α-amylase inhibitor) predictably show greater potency, while those with insulinotropic effects (*e.g*. metformin) have diminished inhibition profiles, as expected.

Conclusions

This work successfully demonstrated the utility of cyclic voltammetry (CV) as a sensitive and rapid electrochemical method for the evaluation of the inhibitory activity of antidiabetic drugs against α-amylase. Using iodide as the redox-active mediator in the starch-amylase system, we have established a reproducible and stable electrochemical interface for the determination of inhibition of the enzyme. Of the six clinically proven antidiabetic drugs tested - acarbose, amaryl, repaglinide, diamecron, dapagliflozin, and metformin - acarbose was most active (IC_{50} = 6.77 μM), with the lowest shown by metformin, as would be expected from their proven pharmacological activity.

Electrochemical outcomes were confirmed by concurrent UV-Vis spectrophotometric analysis of the DNS colorimetric measurements. The high congruence in IC_{50} values and inhibition profiles between the two tests confirms the CV-based method as a plausible alternative for enzyme inhibition screening.

Acknowledgement: *The authors extend thanks and gratitude to the Directorate-General of Scientific Research and Technological Development (DGRSDT) of the Algerian Ministry of Higher Education and Research for financial support*

Conflict of interest: *The authors declare no conflict of interest.*

References

[1] U. Galicia-Garcia, A. Benito-Vicente, S. Jebari, A. Larrea-Sebal, H. Siddiqi, K.B. Uribe, H. Ostolaza, C. Martín, Pathophysiology of Type 2 Diabetes Mellitus, *International Journal of Molecular Sciences* **21** (2020) 6275. https://doi.org/10.3390/ijms21176275

[2] N. A. ElSayed, G. Aleppo, R. R. Bannuru, D. Bruemmer, B. S. Collins, L. Ekhlaspour, J. L. Gaglia, M. E. Hilliard, E. L. Johnson, K. Khunti, I. Lingvay, G. Matfin, R. G. McCoy, M. Lou Perry, S. J. Pilla, S. Polsky, P. Prahalad, R. E. Pratley, A. R. Segal, J. J. Seley, E. Selvin, R. C. Stanton, R. A. Gabbay, 2. Diagnosis and Classification of Diabetes: Standards of Care in Diabetes-2024, *Diabetes Care* **47(Suppl 1)** (2024) S20-S42. https://dx.doi.org/10.2337/dc24-S002

[3] A. Ceriello, Postprandial hyperglycemia and diabetes complications: Is it time to treat? *Diabetes* **54** (2005) 1-7. https://doi.org/10.2337/diabetes.54.1.1

[4] K. Node, T. Inoue, Postprandial hyperglycemia as an etiological factor in vascular failure, *Cardiovascular Diabetology* **8** (2009) 23. https://doi.org/10.1186/1475-2840-8-23

[5] D. L. Eizirik, F. Szymczak, R. Mallone. why does the immune system destroy pancreatic β-cells but not α-cells in type 1 diabetes? *Nature Reviews Endocrinology* **19** (2023) 425-434. https://doi.org/10.1038/s41574-023-00826-3

[6] P. Sudha, S.S. Zinjarde, S.Y. Bhargava, A.R. Kumar, Potent α-amylase inhibitory activity of Indian Ayurvedic medicinal plants, *BMC Complementary Medicine and Therapies* **11** (2011) 5. https://doi.org/10.1186/1472-6882-11-5

[7] H. E. Lebovitz, Alpha-glucosidase inhibitors, *Endocrinology and Metabolism Clinics of North America* **26** (1997) 539-551. https://doi.org/10.1016/S0889-8529(05)70266-8

[8] R. Tundis, M. R. Loizzo, F. Menichini, Natural Products as α-amylase and α-glucosidase Inhibitors and their Hypoglycaemic Potential in the Treatment of Diabetes: An Update, *Mini-Reviews in Medicinal Chemistry* **10** (2010) 315-331. http://dx.doi.org/10.2174/138955710791331007

[9] F. Laar. Alpha-glucosidase inhibitors in the early treatment of type 2 diabetes, *Vascular Health and Risk Management* **4** (2008) 1189-1195. https://doi.org/10.2147/VHRM.S3119

[10] K. Ninomiya, S. Ina, A. Hamada, Y. Yamaguchi, M. Akao, F. Shinmachi, H. Kumagai, H. Kumagai, Suppressive effect of the α-amylase inhibitor albumin from buckwheat (*Fagopyrum esculentum Moench*) on postprandial hyperglycaemia, *Nutrients* **10** (2018) 1503. https://doi.org/10.3390/nu10101503

[11] L. Guo, J. Xia, S. Yu, J. Yan, F. He, M. Zhang, Q. Fan, R. Yang, W. Zhao, Natural edible materials made of protein-functionalized aerogel particles for postprandial hyperglycemia management. *International Journal of Biological Macromolecules* **167** (2021) 279-288. https://doi.org/10.1016/j.ijbiomac.2020.11.186

[12] J. L. Chiasson, R. G. Josse, R. Gomis, M. Hanefeld, A. Karasik, M. Laakso. Acarbose for prevention of type 2 diabetes mellitus: The STOP-NIDDM randomised trial, *The Lancet* **359** (2002) 2072-2077. https://doi.org/10.1016/s0140-6736(02)08905-5

[13] G. L. Miller, Use of Dinitrosalicylic Acid Reagent for Determination of Reducing Sugar, *Analytical Chemistry* **31** (1959) 426-428. https://doi.org/10.1021/ac60147a030

[14] P. Bernfeld, Amylases, α and β, in *Methods in Enzymology*, S. P. Colowick, N.O. Kaplan, Eds., Academic Press, New York, United States, 1955, pp. 149-158. https://doi.org/10.1016/0076-6879(55)01021-5

[15] D. R. Thévenot, K. Toth, R. A. Durst, G. S. Wilson, Electrochemical biosensors: Recommended definitions and classification, *Biosensors and Bioelectronics* **16** (2001) 121-131. https://doi.org/10.1016/S0956-5663(01)00115-4

[16] J. Wang, Electrochemical biosensors: Towards point-of-care cancer diagnostics. *Biosensors and Bioelectronics* **21** (2006) 1887-1892. https://doi.org/10.1016/j.bios.2005.10.027

[17] M. Bae, N. Kim, E. Cho, T. Lee, J.-H. Lee, Recent Advances in Electrochemical Biosensors for Neurodegenerative Disease Biomarkers, *Biosensors* **15** (2025) 151. https://doi.org/10.3390/bios15030151

[18] S. Li, K. Kerman, Electrochemical biosensors for biometal-protein interactions in neurodegenerative diseases, *Biosensors and Bioelectronics* **179** (2021) 113035. https://doi.org/10.1016/j.bios.2021.113035

[19] K. Yokoyama, Y. Kayanuma, Cyclic Voltammetric Simulation for Electrochemically Mediated Enzyme Reaction and Determination of Enzyme Kinetic Constants, *Analytical Chemistry* **70** (1998) 3368-3376. https://doi.org/10.1021/ac9711807

[20] M. Mohiuddin, D. Arbain, A.K.M.Shafiqul Islam, M. S. Ahmad, M. N. Ahmad, Alpha-Glucosidase Enzyme Biosensor for the Electrochemical Measurement of Antidiabetic Potential of Medicinal Plants, *Nanoscale Research Letters* **11** (2016) 95. https://doi.org/10.1186/s11671-016-1292-1

[21] Z. Xiao, R. Storms, A. Tsang,. A quantitative starch-iodine method for measuring alpha-amylase and glucoamylase activities. *Analytical Biochemistry* **351(1)** (2006) 146-148. https://doi.org/10.1016/J.AB.2006.01.036

[22] L. Mogole, W. Omwoyo, F. Mtunzi, Phytochemical screening, anti-oxidant activity and α-amylase inhibition study using different extracts of loquat (*Eriobotrya japonica*) leaves, *Heliyon* **6** (2020) e04736. https://doi.org/10.1016/J.HELIYON.2020.E04736

[23] M. Somogyi, Determination of blood sugar, *Journal of Biological Chemistry* **160** (1945) 69-73. https://doi.org/10.1016/S0021-9258(18)43098-0

[24] Y. Bekkar, E. Lanez, T. Lanez, L. Bourougaa, A. Adaika, A. Benine, Z. Saada, Combined In Vitro and In Silico analysis of ferrocenylmethylaniline derivatives: Antibacterial potential, DFT calculations, and molecular dynamics insights, *Journal of Organometallic Chemistry* 1032 (2025) 123618. https://doi.org/10.1016/J.JORGANCHEM.2025.123618

[25] X.-W. Yang, M.-Z. Huang, Y.-S. Jin, L.-N. Sun, Y. Song, H.-S. Chen, Phenolics from Bidens bipinnata and their amylase inhibitory properties, *Fitoterapia* **83** (2012) 1169-1175. https://doi.org/10.1016/j.fitote.2012.07.005

[26] G. Zengin, C. Sarikurkcu, A. Aktumsek, R. Ceylan, O. Ceylan, A comprehensive study on phytochemical characterization of Haplophyllum myrtifolium Boiss. endemic to Turkey and its inhibitory potential against key enzymes involved in Alzheimer, skin diseases and type II diabetes, *Industrial Crops and Products* **53** (2014) 244-251. https://doi.org/10.1016/j.indcrop.2013.12.043

[27] V. A. Stenger, J. D. McLean, R. M. Van Effen. Polarographic Methods in the Testing of Reagent Chemicals. *Analytical Chemistry* **57** (1985) 27A-36A. https://doi.org/10.1021/ac00279a716

[28] I. Kosta, H. Grande, R. Tena-Zaera, Dimethylformamide-free processing of halide perovskite solar cells from electrodeposited PbI2 precursor films, *Electrochimica Acta* **246** (2017) 1193-1199. https://doi.org/10.1016/j.electacta.2017.06.104

[29] I. S. El-Hallag. Electrochemical oxidation of iodide at a glassy carbon electrode in methylene chloride at various temperatures. *Journal of the Chilean Chemical Society* **55** (2010) 67-73. http://dx.doi.org/10.4067/S0717-97072010000100016

[30] R. Greef, L. M. Peat, L. M. Peter, D. Pletcher, J. Robinson, *Instrumental Methods in Electrochemistry*, Ellis Horwood, New York, United States, 1990, p. 188. ISBN 978-0745803692.