Inorganica Chimica Acta 530 (2022) 120678

Contents lists available at ScienceDirect

Inorganica Chimica Acta

journal homepage: www.elsevier.com/locate/ica

Synthesis and biological evaluation of new silicon(IV) phthalocyanines as carbonic anhydrase and cholinesterase inhibitors

Volkan Çakır [a], Tayfun Arslan [b,*]

[a] Department of Property Protection and Security, Emergency and Disaster Management Program, Espiye Vocational School, Giresun University, 28600 Giresun, Turkey
[b] Department of Chemistry, Faculty of Science, Giresun University, Giresun 28200, Turkey

ARTICLE INFO

Keywords:
Carbonic anhydrase
Cholinesterase
Esterase
Silicon phthalocyanine

ABSTRACT

In this study, novel silicon(IV)phthalocyanines (SiPcs) containing 4-hydroxypyridine (**1a**), N-(2-hydroxyethyl) succinimide (**2a**) and N-(2-hydroxyethyl)phthalimide (**3a**) at their axial positions were synthesized for the first time and surveyed in vitro for key molecular targets. Characterization of new compounds was validated using various spectroscopy techniques. The inhibitory activities of axially disubstituted silicon phthalocyanines were also investigated against carbonic anhydrase isoforms hCA I, II (cytosolic, ubiquitous isozymes), and IX (transmembrane, cancerassociated isozyme) and cholinesterases (AChE and BChE, which are associated with Alzheimer's disease). Against these five esterase enzymes, all synthesized molecules indicated great inhibition effects with IC_{50} values in the range of 49.51–231.05 nM. Among the three silicon(IV)phthalocyanines, **3a** showed most potent inhibitory activity towards both hCA I, AChE and BChE. **1a** demonstrated also the most effective inhibition for hCA II and hCA IX. This work is also the first example of cancer-associated isozyme hCA IX inhibition by silicon(IV)phthalocyanines.

1. Introduction

Alzheimer's disease (AD), which has especially affected the elderly population in recent years, most common type of neurodegenerative illness. Especially after the age of 60, there is an increase in its incidence. Symptoms of Alzheimer's disease appear gradually and over time affect the person to such an extent that they cannot do their daily work on their own. This disease causes simple, tolerable forgetfulness in the initial period. However, it progresses over time, causing the person to be unable to remember what they did in the recent past. The patient cannot recognize friends, family, spouse or even children. In the last period of the disease, the person cannot meet her own basic needs. Although there are many different causes of Alzheimer's, the main causes can be listed as follows: people's lifestyle, advanced age, genetic, cardiovascular diseases, hypertension, diabetes, low levels of acetylcholine, agglomeration of β-amyloid (Aβ) peptides, Vitamin B5 deficiency, τ -protein aggregation and oxidative stress. The exact cause of this disease, which is extremely important for all humanity, has not yet been fully understood and there is no permanent remedy for AD [1–4]. The drugs present in the market reduce only the symptoms of this disease and slow the deterioration of the condition. In particular, recent studies for this illness have focused on cholinesterase inhibitors. Cholinesterases catalyze the hydrolysis of acetylcholine which is neurotransmitter and decrease the level of acetylcholine in the brain but inhibiting cholinesterases enhance cholinergic neurotransmission [5].

CA isoenzymes are vital in reactions where bicarbonate is a substrate, such as pH balance, release of electrolytes, respiration, glycogenesis, lipogenesis and ureagenesis [6–8]. In recent years, human CA inhibitors have been clinically employed for the therapy of various diseases such as, intracranial hypertension, epilepsy, obesity besides diuretics. These inhibitors have been recently commenced to be employed for the treatment of hypoxic tumors, and also accepted as significant drugs for neuropathic pain, arthritis and cerebral ischemia [9–11].

Phthalocyanines, stable synthetic macrocyclic compounds, are widely used in technological and medicinal areas such as photodynamic therapy [12], biosensor [13], solar cells [14] and catalysts [15]. To date, they have showed a wide range of biological properties such as antibacterial [16], antimicrobial [17], antioxidant [18], antifungal [19] and enzyme inhibition [20]. The very low solubility and aggregation of phthalocyanines in water and commonly used organic solvents limits their biological and technological applications. To overcome this

* Corresponding author.
E-mail address: tayfun.arslan@giresun.edu.tr (T. Arslan).

https://doi.org/10.1016/j.ica.2021.120678
Received 15 September 2021; Received in revised form 25 October 2021; Accepted 25 October 2021
Available online 29 October 2021

problem, the introduction of substituents at the axial positions of phthalocyanines is preferred because the axial positions can strongly influence some properties of phthalocyanines such as its solubility and aggregation behavior [20]. For this reason, non-aggregating axially disubstituted silicon(IV)phthalocyanines (SiPcs) can be used in biological applications. Previous researches have also demonstrated that toxicity of SiPcs are very low against cancer cells [21,22].

In the light of above-mentioned information, it has been clearly understood that the discovery of more effective novel and potent enzyme inhibitors has become an urgent task in current scientific research. As research on the biological study of SiPc is also limited in the literature, this study has been carried out in order to support the ongoing studies on this subject. We report herein, the synthesis and characterization of the axially disubstituted novel silicon(IV) phthalocyanines and their hCA I/II/IX, AChE/BChE, inhibitory properties were surveyed for to identify the therapeutic potentials. To the best of our knowledge, the hCA IX inhibitory properties of silicon(IV)phthalocyanines have not been reported in the literature to date. Therefore, their human transmembrane cancer-associated isozyme hCA IX inhibitory properties were also investigated for the first time.

2. Experimental

2.1. Materials and equipments

Silicon phthalocyanine dichloride ($SiPcCl_2$), 4-hydroxypyridine, N-(2-hydroxyethyl)succinimide and N-(2-hydroxyethy)phthalimide were purchased from commercial suppliers. All commercial reagents were used without further purification. All solvents were dried according to standard methods [23]. The IR spectra were determined using a Perkin Elmer 1600 Fourier Transform-Infrared (FT-IR-ATR) spectrophotometer. ^{1}H NMR spectra were recorded on a Bruker Avance III 400 MHz NMR spectrometer in DMSO-d_6, and chemical shifts (δ) are reported in ppm and coupling constants (*J*) are given in hertz (Hz). Mass spectra were measured on a Bruker Microflex LT MALDI-TOF MS spectrometer. Melting points were measured on a Barnstead electrothermal 9200 series digital apparatus. Electronic spectra in the UV–Vis region were recorded on a Perkin Elmer-Lambda 25 spectrophotometer, using 1 cm pathlength cuvettes at room temperature.

2.2. Purification of hCA I and II isoenzymes

hCA I and II isoenzymes were obtained from fresh human erythrocytes by employing affinity chromatography as described in the literature [20].

2.3. hCA I and II isoenzyme activity assays

hCA I and II isoenzyme activity assays were performed according to the Verpoorte method [24]. The inhibitory effects of all synthesized molecules in this part of the study were extensively tested. These derivatives were investigated in triplicate at each concentration employed. The different concentrations were employed for all inhibitors, respectively. In the absence of inhibitor, the control cuvette activity was accepted as 100%. Activities (%)-[Inhibitor] graphs in this study were separately plotted for each inhibitor (Table 1).

2.4. AChE and BChE activity assay

Inhibition activities of all synthesized molecules against AChE and BChE enzymes were determined by employing the Ellman colorimetric process [25]. In this research, neostigmine for AChE assay was employed as a reference drug. IC_{50} values obtained for all molecules tested are summarized in Table 1.

In this study, the stock solutions of all molecules tested (1a-3a) were made ready by dissolving in 1 mgmL-1 of dimethylsulfoxide. Then, the obtained stock solution was diluted ten thousand times with distilled water. So as to establish the inhibition activities of all synthesized molecules with these enzymes, the measurements were conducted in seven different concentrations. The method of this study has been stated in earlier studies in detail [26].

Table 1
hCA I, II, IX, AChE and BChE inhibition data (IC_{50})[a] of all molecules synthesized.

Compounds	hCA I (nM)	hCA II (nM)	hCA IX (nM)	AChE (nM)	BChE (nM)
1a	102.82 ± 1.72	86.64 ± 0.94	49.51 ± 0.57	116.17 ± 2.11	115.52 ± 3.46
2a	173.28 ± 3.43	104.07 ± 1.27	57.76 ± 0.61	166.61 ± 3.72	231.05 ± 4.15
3a	99.02 ± 1.12	100.72 ± 1.16	63.02 ± 0.78	86.25 ± 0.96	112.85 ± 1.46
AZA[b]	250	12	25	–	–
Neostigmine[c]	–	–	–	135.9 ± 1.12	84.0 ± 0.81

[a] IC_{50} values = mean (n ± SD). (Concentration that inhibits 50% in nM, and the data were acquired from triplicate runs).
[b] Acetazolamide (AZA) was employed as a control for hCA I, II and IX [1].
[c] Neostigmine was employed as a control for AChE and BChE.

2.5. Synthesis

2.5.1. Synthesis of silicon phthalocyanine (1a)

A mixture of dichloro(phthalocyaninato) silicon (a) (200 mg, 0.32 mmol) and 4-hydroxypyridine (1) (61 mg, 0.64 mmol) in toluene (20 mL) was stirred and then sodium hydride (26.4 mg, 0.64 mmol) was added to this mixture. After refluxing under nitrogen 24 h, the reaction mixture was cooled to room temperature, and then the solvent was evaporated to dryness under reduced pressure. The crude product was purified by column chromatography which is placed aluminum oxide using $CHCl_3$:CH_3OH (100:2) as solvent system. Dark blue product was obtained (1a). The synthesis of compound 1a shown in Fig. 1. Yield: 97 mg (40%). Mp > 300 °C. FT-IR (KBr, cm^{-1}): 3049 (Ar. CH), 2907 (Aliph. CH), 2843, 1701, 1611, 1517, 1428, 1400, 1332, 1290, 1119, 1077, 929, 721 ; ^{1}H NMR (400 MHz, DMSO-d_6), (δ:ppm): 9.79–9.74 (m, 8H, *Pc*–Hα), 8.31–8.28 (m, 8H, *Pc*–H_β), 7.89–7.80 (m, 4H, Ar–H), 7.03–6.98 (m, 4H, Ar–H); ^{13}C NMR (100 MHz, DMSO-d_6), (δ:ppm): 161.6, 151.1, 149.2, 136.5, 129,2 123.2, 113.4; UV–Vis ($CHCl_3$): λ_{max}, nm (log ε): 674 (5.01), 642 (4.56), 606 (4.42), 354 (4.81) ; MALDI-TOF-MS (*m/z*): Calculated: 728.19; Found: 730.236 $[M + 2H]^{2+}$, 635.418 $[M-C_5H_5NO]^+$

2.5.2. Synthesis of silicon phthalocyanine (2a)

A mixture of $SiPcCl_2$ (a) (200 mg, 0.32 mmol), N-(2-hydroxyethyl) succinimide (2) (92 mg, 0.64 mmol) and NaH (26.4 mg, 0.64 mmol) in toluene (20 mL) was refluxed for 24 h. After evaporating the solvent in vacuo, the residue was purified by aluminum oxide column chromatography with $CHCl_3$:CH_3OH (100:2) as solvent system. Pure product was obtained (2a). The synthesis of compound 2a shown in Fig. 1. Yield: 103 mg (37%). Mp > 300 °C. FT-IR (KBr, cm^{-1}): 2973 (Aliph. CH), 2874, 1637, 1611, 1516, 1471, 1429, 1332, 1289, 1190, 1118, 1067, 910, 825, 724 ; ^{1}H NMR (400 MHz, DMSO-d_6), (δ:ppm): 9.66–9.63 (m, 8H, *Pc*–Hα), 8.36–8.32 (m, 8H, *Pc*–H_β), 3.67 (m, 4H, O-CH_2), 3.41 (m, 4H, N-CH_2), 2.64 (m, 4H, –CH_2); ^{13}C NMR (100 MHz, DMSO-d_6), (δ:ppm): 175.2, 149.1, 135.8, 129.3, 123.7, 59.8, 37.4, 28.9; UV–Vis ($CHCl_3$): λ_{max}, nm (log ε): 676 (5.03), 643 (4.58), 607 (4.43), 352 (4.79); MALDI-TOF-MS (*m/z*): Calculated: 824.23; Found: 825.092 $[M + H]^+$, 683.177 $[M-C_6H_9NO_3]^+$.

2.5.3. Synthesis of silicon phthalocyanine (3a)

The synthesis of silicon phthalocyanine (3a) procedure was applied according to the compound (2a) by using N-(2-hydroxyethyl)

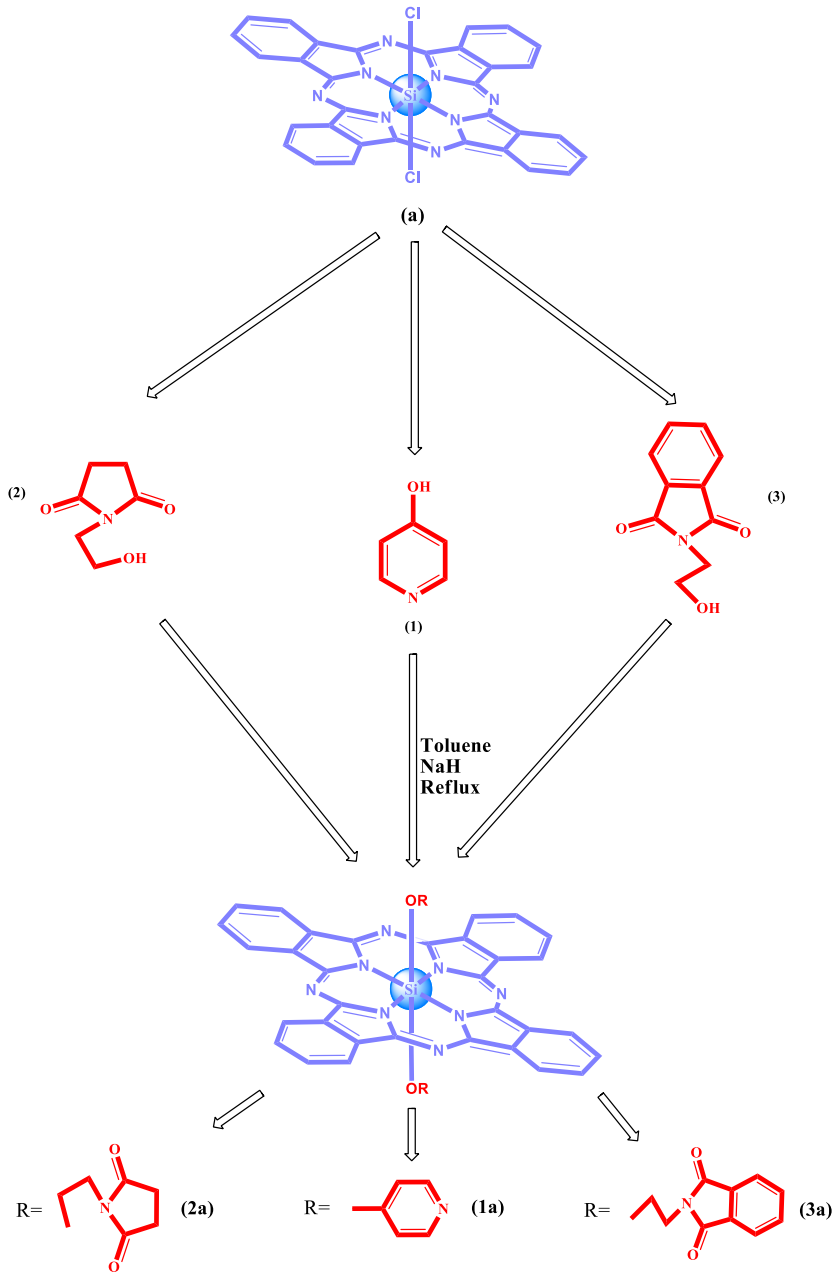

Fig. 1. The synthesis of silicon phthalocyanines 1a, 2a and 3a.

phthalimide (3) (122 mg, 0.64 mmol). The synthesis of compound 3a shown in Fig. 1. Yield: 128 mg (42%). Mp > 300 °C. FT-IR (KBr, cm^{-1}): 3054 (Ar. CH), 2932 (Aliph. CH), 2819, 1701, 1610, 1518, 1429, 1334, 1291, 1119, 1079, 905, 828, 727 ; ^{1}H NMR (400 MHz, DMSO-d_6), (δ: ppm): 9.63–9.60 (m, 8H, *Pc*–Hα), 8.33–8.30 (m, 8H, *Pc*–H_β), 7.92–7.89 (m, 4H, Ar-H), 7.79–7.75 (m, 4H, Ar-H), 3.90 (m, 4H, O-CH_2), 3.78 (m, 4H, N-CH_2) ; ^{13}C NMR (100 MHz, DMSO-d_6), (δ:ppm): 169.7, 148.9, 136.1, 133.4, 132.8, 129.7, 124.6, 123.1, 59.2, 42.4; UV–Vis ($CHCl_3$): λ_{max}, nm (log ε): 672 (4.99), 641 (4.54), 604 (4.40), 353 (4.80); MALDI-TOF-MS (*m/z*): Calculated: 920.23; Found: 919.789 $[M-H]^+$, 730.723 $[M- C_{10}H_9NO_3]^+$.

3. Results and discussion

3.1. Synthesis and characterization

The synthetic route of the compounds presented in this work is shown in Fig. 1. Compounds 4-hydroxypyridine (**1**), N-(2-hydroxyethyl) succinimide (**2**) and N-(2-hydroxyethy)phthalimide (**3**) was purchased from commercial suppliers. Reaction of silicon phthalocyanine dichloride (a) with 4-hydroxypyridine (1), N-(2-hydroxyethyl)succinimide (2) and N-(2-hydroxyethyl)phthalimide (3) in the present of NaH in toluene led to the axially substituted silicon phthalocyanines 1a (in 40%), 2a (in 37%) and 3a (in 42%). Silicon(IV) phthalocyanines (1a, 2a and 3a) were purified by column chromatography which is placed aluminum oxide using $CHCl_3$:CH_3OH (100:2) as solvent system. The phthalocyanines (1a, 2a and 3a) show good solubility in common organic solvents, e.g., dichloromethane, chloroform, and DMF. All synthesized new axially disubstituted silicon phthalocyanines (1a, 2a and 3a) were verified by IR, UV–vis, MS and NMR spectroscopic techniques.

In the IR spectrum of silicon phthalocyanines (1a, 2a and 3a) were clearly confirmed by disappearance of the —OH bands. The characteristic vibrations corresponding to aliphatic –CH stretching bands were

observed at around 2944–2861 cm^{-1} for 1a, 2a and 3a. Aromatic –CH peaks were observed at around 3050 cm^{-1} for 1a, and 3a. The ^{1}H NMR spectrum of axially disubstituted silicon(IV)phthalocyanine complexes **1a**, **2a** and **3a** showed peaks belonging to H_{α} and H_{β} protons at between 9,79–9,74 and 8.31–8.28 ppm for complex **1a**, 9.66–9.63 and 8.36–8.32 ppm for complex **2a**, 9.63–9.60 and 8.33–8.30 ppm for complex **3a**, respectively. The ^{1}H NMR spectrum of silicon phthalocyanine complexes **1a** and **3a,** the observation of new signals at between 7.89 and 7.80 and 7.03–6.98 ppm for complex **1a**, 7.92–7.89 and 7.79–7.75 ppm for complex **3a** belonging to aromatic protons on the substituents proved the synthesis of this phthalocyanines. On the other hand, the appearance of new signal at $\delta = 3.67$, 3.41, 2.64 and 3.90, 3.78 ppm belonging to aliphatic protons also confirmed the formation of silicon(IV) phthalocyanines **2a** and **3a**. The ^{13}C NMR spectra of SiPc **1a**, **2a**, **3a** demonstrated signals for relative carbon atoms. In the ^{13}C NMR spectrum of **2a** and **3a** exhibited the carbonyl carbon atoms at $\delta = 175.2$ and 169.7 ppm. respectively. The ^{13}C NMR spectra of **1a**, the aromatic carbon atoms resonated between 151.1 and 113.4 ppm.

Generally, two absorption bands are observed for phthalocyanine compounds in their electronic absorption spectra. One of them is observed at around 600–750 nm and is known as Q. The other is observed in the ultra violet region of the spectrum at around 300–450 nm and is known as the B or Soret band [27]. The UV–vis spectra of silicon phthalocyanines 1a, 2a and 3a in $CHCl_3$ at room temperature are shown in Fig. 2. The Q band of novel axially disubstituted silicon phthalocyanines (1a, 2a and 3a) were observed at 674, 676 and 672 nm respectively is caused from transitions from π-HOMO to π*-LUMO energy levels. B or Soret band, is caused from electronic transitions from deeper level of HOMO to LUMO for these compounds 1a, 2a and 3a were observed at 354, 352 and 353 nm, respectively (Fig. 2).

In the mass spectra of axially disubstituted silicon phthalocyanines (1a, 2a and 3a), the molecular ion peaks were observed at m/z = 730.236 $[M + 2H]^{2+}$, 635.418 $[M-C_5H_5NO]^{+}$, 825.092 $[M + H]^{+}$, 683.177 $[M-C_6H_9NO_3]^{+}$ and 919.789 $[M-H]^{+}$, 730.723 $[M-C_{10}H_9NO_3]^{+}$ respectively, confirmed the proposed structures.

3.2. Biochemical studies

Many studies have shown that there is a link between Alzheimer's illness and cholinesterases. Recently, several studies shed also light on carbonic anhydrases as possible new targets for AD treatment [28,29]. Inhibitory effects of various heterocyclic compounds such as imidazolium, pyrazole, hydrazone on carbonic anhydrases and cholinesterases were surveyed [30–33]. Both metal-free and metallophthalocyanines containing various substituents were tested for their CA I/II and AChE/BChE inhibitory effects [34–35]. In an earlier report from our group, the inhibitory ability of some silicon(IV)phthalocyanines compounds on carbonic anhydrase isozymes (hCA I and II) [20]. In another study, the ability of some phthalocyanines to inhibit both carbonic anhydrases (CA I/II/IX) and cholinesterases (AChE/BChE) enzymes was also investigated [1,36]. Hence, it was decided to investigate for these enzymes in molecules of like structure. Inhibitory effects of silicon phthalocyanines 1a, 2a and 3a on enzyme activities were tested for the first time under in vitro conditions; IC_{50} values of all molecules synthesized are given in Table 1. In this study, neostigmine were used as reference drugs for AChE and BChE enzymes and Acetazolamide was used for carbonic anhydrase enzymes.

1. Newly synthesized silicon(IV)phthalocyanines against the slow cytosolic isozyme hCA I exhibited enzyme inhibition activity at concentrations between 99.02 and 173.28 nM. It was determined that all molecules synthesized in this study showed activity at much

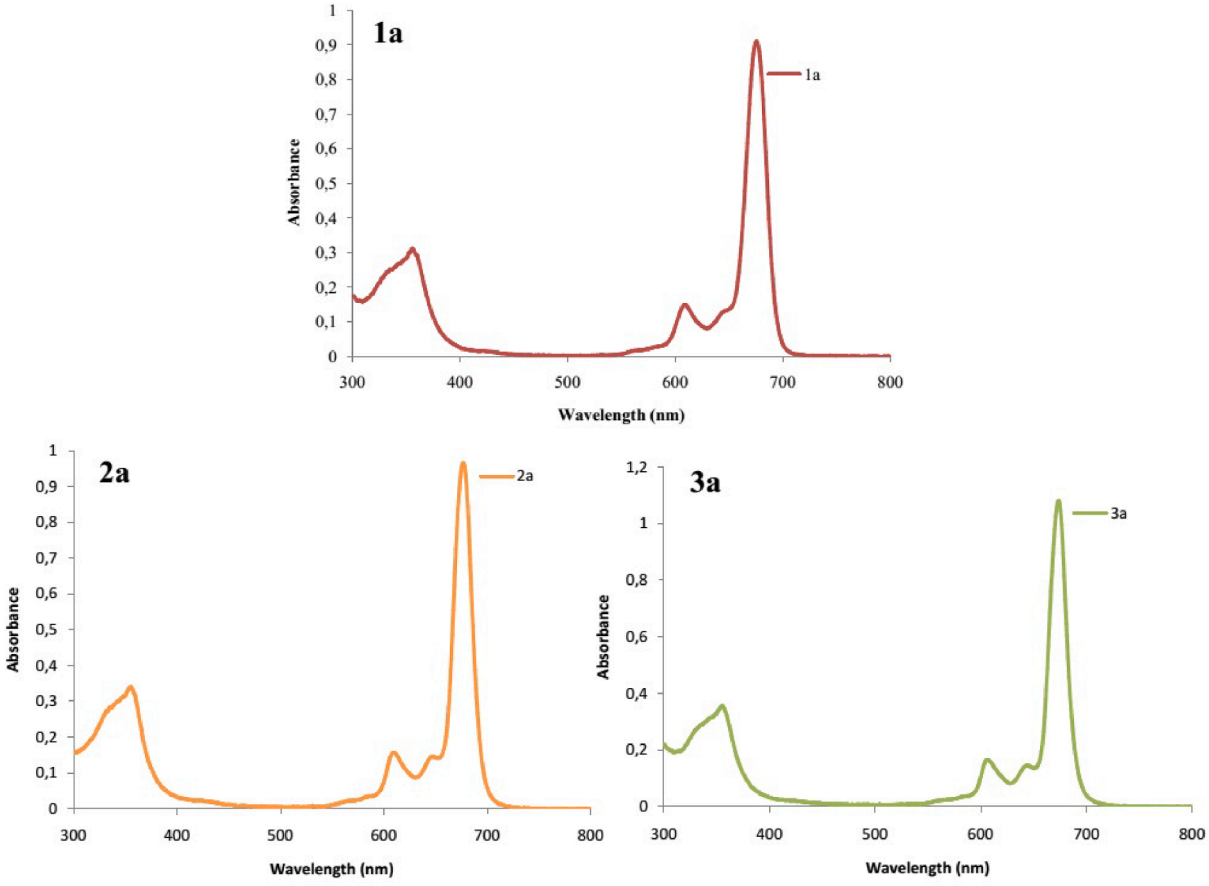

Fig. 2. The UV–Vis spectra of silicon phthalocyanines 1a, 2a and 3a in chloroform at room temperature (1×10^{-5} M).

lower concentrations than AZA (IC_{50} = 250 nM) employed as a reference drug. Compound **2a** (IC_{50} = 173.28 nM) showed the weakest inhibitory activity against hCA I. It was determined that the most active molecule against this enzyme was compound **3a** (IC_{50} = 99.02 nM). This molecule was calculated to be approximately 2.5 times more active than the reference drug AZA. It can be said that this situation arises because phthalocyanines, which have a larger molecular structure than some other derivatives, interact more with the active site of the enzyme. In many previous studies by different research groups, it was emerged that natural phenolic molecules, pesticides, sulphonamide derivatives, molecules containing nitro groups uracil derivatives and great number of the diverse chemicals was indicated to provide similar results with these molecules synthesized against the hCA I isoenzyme [1,8,20,37].

2. In the rapid cytosolic isozyme hCA II assay, it was determined that all synthesized SiPc exhibited activity with IC_{50} values ranging from 86.64 to 104.07 nM towards hCA II (Table 1). Also, the phthalocyanines we tested in this study showed lower activity than the reference molecule AZA (12 nM). Among the SiPc tested, compound 1a containing the pyridine ring (IC_{50} = 86.64 nM) was determined to be the most potent inhibitor for hCA II.
3. Against the cancer-related isozyme hCA IX, SiPc 1a containing the pyridine ring behaved as good inhibitor (IC_{50} = 49.51 nM). The weakest inhibitor against hCA IX was 3a. As seen in Table 1, it was determined that all synthesized molecules inhibited hCA IX more potently than hCA I and II isoenzymes. In addition, it was determined that phthalocyanines with lower volume sidegroups have higher enzyme inhibition activities than other derivatives, since they interact more with the active center of the enzyme. As a result, it can be said that these molecules have the potential to be used in the treatment of some types of cancer due to their effectiveness at lower doses for the hCA IX enzyme.
4. In AChE assay, It was determined that **1a** (116.1 nM) and **3a** (86.2 nM) SiPcs outperformed the reference molecule neostigmine (135.9 nM), while **2a** had an activity close to neostigmine. Sipc **2a** (IC_{50} = 166.61 nM) exhibited the weakest inhibitory effect against AChE (Table 1). **3a** was determined to be approximately 1.5 times more active than the reference drug neostigmine. In a previous study, it was determined by our group that the values obtained for phthalocyanines containing pyridine side group gave similar results [1]. In conclusion, the results revealed that overall, all examined SiPCs showed promising AChE inhibitory activities.
5. Silicon(IV) phthalocyanines (1a) and (3a) displayed the strongest inhibitory effects to BChE with IC_{50} values of 115.52 ± 3.46 and 112.85 ± 1.46 nM. These results were similar to IC_{50} value of neostigmine (84.0 ± 0.81 nM) against BChE. Similar to the AChE enzyme, 3a, the molecule with the largest side group, showed the highest enzyme inhibition activity. When the results were examined, it was observed that the newly synthesized especially 1a and 3a SiPc derivatives showed promising BChE inhibitory activities.

In prior research, phthalocyanines containing different substituents have shown inhibitory effects against various enzymes including cholinesterases, carbonic anhydrases, α-glucosidase. Considering the results, the alterations on their lateral chain and internal core have a important effect on their inhibition ability [1,5,20,30,38]. SiPcs displayed distinct inhibitory impacts on cholinesterases and carbonic anhydrases. When the IC_{50} values of the SiPc for hCA II and IX were studied, 1a, which did bear pyridine ring in its structure, had the most inhibitory effect. When the inhibition values of cholinesterases were examined, 3a showed the best inhibitory effect. It can be said that these SiPcs, which show an inhibitory potential close to neostigmine and acetazolamide which is used as a cholinesterase and carbonic anhydrase inhibitors, have the potential to be drugs in the treatment of various neurodegenerative diseases.

4. Conclusion

In the presented study, novel SiPcs bearing pyridine, phthalimide and succinimide rings were synthesized for the first time. We investigated compounds 1a, 2a and 3a for their inhibition activities against hCA isoforms (I, II, and IX) and cholinesterases (AChE and BChE). As a result of this study, the new silicon(IV)phthalocyanine analogues showed inhibition at the nanomolar levels against these enzymes. The newly synthesized compounds were good inhibitors for hCA I, II and IX isoenzymes and AChE/BChE enzymes. This study is also the first example of transmembrane tumor-associated hCA IX inhibition by SiPcs. Thus, this type of phthalocyanines may form the cornerstone for the development of novel antiglaucoma, antiepileptic and anti-Alzheimer therapeutic molecules.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgement

We thank Prof. Dr. Murat Sentürk for contributing to the interpretation of enzyme inhibition.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ica.2021.120678.

References

[1] T. Arslan, M.B. Ceylan, H. Baş, Z. Biyiklioglu, M. Senturk, Design, synthesis, characterization of novel peripherally tetra 1,2,3-triazole substituted phthalocyanines and inhibitor effect on cholinesterases (AChE/BChE) and carbonic anhydrases (hCA I, II and IX), Dalton Trans. 49 (2020) 203–209.
[2] J.A. Lima, T.W.R. Costa, A.C.C. Fonseca, R.F. Amaral, M.D.S.B. Nascimento, O. A. Santos-Filho, A.L. Miranda, D.C.F. Neto, F.R.S. Lima, L. Hamerski, L.W. Tinoco, Geissoschizoline, a promising alkaloid for Alzheimer's disease: Inhibition of human cholinesterases, anti-inflammatory effects and molecular docking, Bioorg. Chem. 104 (2020), 104215.
[3] S. Shaikha, P. Dhavanb, G. Pavalea, M.M.V. Ramanaa, B.L. Jadhavb, Design, synthesis and evaluation of pyrazole bearing α-aminophosphonate derivatives as potential acetylcholinesterase inhibitors against Alzheimer's disease, Bioorg. Chem. 96 (2020), 103589.
[4] S. Srivastava, R. Ahmad, S.K. Khare, Alzheimer's disease and its treatment by different approaches: a review, Eur. J. Med. Chem. 216 (2021) 113320, https://doi.org/10.1016/j.ejmech.2021.113320.
[5] B. Barut, U. Demirbaş, Synthesis, anti-cholinesterease, a-glucosidase inhibitory, antioxidant and DNA nuclease properties of non-peripheral triclosan substituted metal-free, copper(II), and nickel(II) phthalocyanines, J. Organomet. Chem. 923 (2020) 121423.
[6] Claudiu T. Supuran, Carbonic anhydrases: novel therapeutic applications for inhibitors and activators, Nat. Rev. Drug. Discov. 7 (2) (2008) 168–181.
[7] İ. Fidan, R.E. Salmas, M. Arslan, M. Şentürk, S. Durdağı, D. Ekinci, E. Şentürk, S. Coşgun, C.T. Supuran, Carbonic anhydrase inhibitors: design, synthesis, kinetic, docking and molecular Dynamics analysis of novel glycine and phenylalanine sulphonamide derivatives, Bioorg. Med. Chem. 23 (2015) 7353–7358.
[8] Semra Işık, Daniela Vullo, Serdar Durdagi, Deniz Ekinci, Murat Şentürk, Ahmet Çetin, Esra Şentürk, Claudiu T. Supuran, Interaction of carbonic anhydrase isozymes I, II, and IX with some pyridine and phenol hydrazinecarbothioamide derivatives, Bioorg. Med. Chem. Lett. 25 (23) (2015) 5636–5641.
[9] Claudiu T. Supuran, Agents for the prevention and treatment of age-related macular degeneration and macular edema: a literature and patent review, Expert. Opin. Ther. Pat. 29 (10) (2019) 761–767.
[10] V. Burianova, S. Kalinin, C.T. Supuran, M. Krasavin, Radiotracers for positron emission tomography (PET) targeting tumour-associated carbonic anhydrase isoforms, Eur. J. Med. Chem. 213 (2021), 113046.
[11] Halis T. Balaydın, Murat Şentürk, Abdullah Menzek, Synthesis and carbonic anhydrase inhibitory properties of novel cyclohexanonyl bromophenol derivatives, Bioorg. Med. Chem. Lett. 22 (3) (2012) 1352–1357.
[12] K. Plaetzer, B. Krammer, J. Berlanda, F. Berr, T. Kiesslich, Photophysics and photochemistry of photodynamic therapy: fundamental aspects, Lasers Med. Sci. 24 (2) (2009) 259–268.

[13] G. Guillaud, J. Simon, J.P. Germain, Metallophthalocyanines, Coord. Chem. Rev. 178-180 (1998) 1433–1484.
[14] Yulong Wang, Xiaoyuan Liu, Haiquan Shan, Qian Chen, Tuo Liu, Xize Sun, Dian Ma, Zihao Zhang, Jiaju Xu, Zong-Xiang Xu, Tetra-alkyl-substituted copper (II) phthalocyanines as dopant-free holetransport layers for planar perovskite solar cells with enhanced open circuit voltage and stability, Dyes Pigm. 139 (2017) 619–626.
[15] Volkan Çakır, Ece Tuğba Saka, Zekeriya Bıyıklıoğlu, Halit Kantekin, Highly selective oxidation of benzyl alcohol catalyzed by new peripherally tetra-substituted Fe(II) and Co(II) phthalocyanines, Synth. Met. 197 (2014) 233–239.
[16] M. Rudrapal, R.S. Satyanandam, T.S. Swaroopini, T.N. Lakshmi, S.K. Jaha, S. Zaheera, Synthesis and antibacterial activity of some new hydrazones, Med. Chem. Res. 22 (2013) 2840-2846.
[17] R. Gupta, R.P. Chaudhary, Synthesis, antimicrobial and DFT studies of novel fused thiazolopyrimidine derivatives, Heterocycl. Commun. 19 (2013) 207–214.
[18] A. Günsel, A.T. Bilgiçli, C. Kandemir, R. Sancak, G. Arabacı, M.N. Yaraşır, Comparison of novel tetra-substituted phthalocyanines with their quaternized derivatives: antioxidant and antibacterial properties, Synth. Met. 260 (2020), 116288.
[19] F. Tang, F. Gao, W. Xie, S. Li, B. Zheng, M. Ke, J. Huang, Carboxymethyl chitosan-zinc(II) phthalocyanine conjugates: synthesis, characterization and photodynamic antifungal therapy, Carbohydr. Polym. 235 (2020), 115949.
[20] Tayfun Arslan, Zekeriya Biyiklioglu, Murat Şentürk, The synthesis of axially disubstituted silicon phthalocyanines, their quaternized derivatives and first inhibitory effect on human cytosolic carbonic anhydrase isozymes hCA I and II, RSC Adv. 8 (19) (2018) 10172–10178.
[21] İpek Ömeroğlu, Esra Nur Kaya, Meltem Göksel, Vesselin Kussovski, Vanya Mantareva, Mahmut Durmuş, Axially substituted silicon(IV) phthalocyanine and its quaternized derivative as photosensitizers towards tumor cells and bacterial pathogens, Bioorg. Med. Chem. 25 (20) (2017) 5415–5422.
[22] K. Li, L. Qiu, Q. Liu, G. Lv, X. Zhao, S. Wang and J. Lin, J. Photochem. Photobiol. B, Conjugate of biotin with silicon(IV) phthalocyanine for tumor-targeting photodynamic therapy, 174 (2017) 243–250.
[23] D.D. Perrin, W.L.F. Armarego, Purification of Laboratory Chemicals, Pergamon Press, Oxford, 1989.
[24] Jacob A. Verpoorte, Suchinta Mehta, John T. Edsall, Esterase activities of human carbonic anhydrases B and C, J. Biol. Chem. 242 (18) (1967) 4221–4229.
[25] George L. Ellman, K.Diane Courtney, Valentino Andres, Robert M. Featherstone, A new and rapid colorimetric determination of acetylcholinesterase activity, Biochem. Pharmacol. 7 (2) (1961) 88–95.
[26] Huseyin Cavdar, Murat Senturk, Murat Guney, Serdar Durdagi, Gulru Kayik, Claudiu T. Supuran, Deniz Ekinci, Inhibition of acetylcholinesterase and butyrylcholinesterase with uracil derivatives: kinetic and computational studies, J. Enzy. Inhibit. Med. Chem. 34 (1) (2019) 429–437.
[27] K. D. Modibane, Grahamstown, Rhodes University, (2009).
[28] D. Taylor, C. Paton, K. Shitij. Maudsley Prescribing Guidelines in Psychiatry, 11th ed., Wiley-Blackwell, West Sussex, 2012.
[29] G. Provensi, F. Carta, A. Nocentini, C.T. Supuran, F. Casamenti, M.B. Passani, S. Fossati, A new kid on the block? carbonic anhydrases as possible new targets in Alzheimer's disease, Int. J. Mol. Sci. 20 (2019) 4724.
[30] P. Taslimi, F. Turkan, A. Cetin, H. Burhan, M. Karaman, I. Bildirici, İ. Gulcin, F. Şen, Pyrazole[3,4-*d*]pyridazine derivatives: molecular docking and explore of acetylcholinesterase and carbonic anhydrase enzymes inhibitors as anticholinergics potentials, Bioorg. Chem. 92 (2019), 103213.
[31] Fikret Turkan, Adnan Cetin, Parham Taslimi, Muhammet Karaman, İlhami Gulçin, Synthesis, biological evaluation and molecular docking of novel pyrazole derivatives as potent carbonic anhydrase and acetylcholinesterase inhibitors, Bioorg. Chem. 86 (2019) 420–427.
[32] Kaan Kucukoglu, Halise Inci Gul, Parham Taslimi, Ilhami Gulcin, Claudiu T. Supuran, Investigation of inhibitory properties of some hydrazone compounds on hCA I, hCA II and AChE enzymes, Bioorg. Chem. 86 (2019) 316–321.
[33] S. Bal, R. Kaya, Y. Gok, P. Taslimi, A. Aktas, M. Karaman, İ. Gulcin, Novel 2-methylimidazolium salts: synthesis, characterization, molecular docking, and carbonic anhydrase and acetylcholinesterase inhibitory properties, Bioorg. Chem. 94 (2020), 103468.
[34] Furkan Özen, Aslıhan Günel, Arif Baran, DNA-binding, enzyme inhibition, and photochemical properties of chalconecontaining metallophthalocyanine compounds, Bioorg. Chem. 81 (2018) 71–78.
[35] E. Güzel, Ü.M. Koçyiğit , P. Taslimi, İ. Gülçin, S. Erkan, M. Nebioğlu , B.S. Arslan, İ. Şişman, Phthalocyanine complexes with (4-isopropylbenzyl)oxy substituents: preparation and evaluation of anti-carbonic anhydrase, anticholinesterase enzymes and molecular docking studies, J. Biomol. Struct. Dyn. doi:10.1080/07391102.2020.1818623.
[36] T. Arslan, N. Cakır, T. Keleş, Z. Biyiklioglu, M. Senturk, Triazole substituted metal-free, metallo-phthalocyanines and their water soluble derivatives as potential cholinesterases inhibitors: Design, synthesis and *in vitro* inhibition study, Bioorg. Chem. 90 (2019), 103100.
[37] E.A. Kazancioglu, M. Şentürk, Synthesis of N-phenylsulfonamide derivatives and investigation of some esterase enzymes inhibiting properties, Bioorg. Chem. 104 (2020), 104279.
[38] Tian Tian, Liwei Weng, Shaoru Wang, Xiaocheng Weng, Lixia Zhang, Xiang Zhou, Cationic tetrapyrrolic macromolecules as new acetylcholinesterase inhibitors, J. Porphyr. Phthalocyan. 13 (08n09) (2009) 893–902.