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Sulfur, selenium and tellurium containing amines act as effective carbonic anhydrase activators

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Abstract. A new series of β -aminochalcogenides were designed and synthesized to identify new carbonic anhydrase activator (CAA) agents as novel tools for the management of several neurodegenerative and metabolic disorders which represent a clinical challenge without effective therapies available. Some β -aminoselenides and β -aminotellurides showed effective CA activating effects and a potent antioxidant activity. CAAs may have applications for memory therapy and CA deficiency syndromes.

Keywords: Carbonic Anhydrases (CAs), Carbonic Anhydrase Activators (CAAs), selenium, metalloenzymes, tellurium.

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1. Introduction

Carbonic anhydrases (CAs; EC 4.2.1.1) are ubiquitous metalloenzymes known for decades to be involved in the reversible hydration of CO₂ to bicarbonate and H⁺ ions.^{1,2} This simple reaction plays a key role in many crucial physiologic functions connected to metabolic pathways in which CO₂ is involved.³⁻⁶ For this reason, their modulation is used for a long time in the treatment of different diseases such as glaucoma,⁷ edema,⁸ epilepsy⁹ and, more recently, inhibitors of CAs are used as a new approach for treatment of neuropathic pain,^{10,11} cerebral ischemia¹² and different forms of tumor.¹³ Conversely, the activation of CAs was been for a long time a debated issue and only in recent years it has been started an exploration of their possible pharmacological applications.^{2,14} Histamine (**Figure 1**) was one of the first CA activator (CAA) discovered¹⁵ and also one of the most investigated scaffold for designing novel ones.^{16,17} X-ray crystal structure of CA-Histamine has been reported in the 90's showing the mechanism of activation, where the molecule participates to the rate-determining step of the catalytic cycle, facilitating the formation of the nucleophilic species of the enzyme.¹⁵ In the last years, our group focused on new molecules with activating propriety against CAs, most of which amines of the general formula Ar-CH₂CH(R)NHR'.¹⁸⁻²⁰ In a recent study from our group,²¹ psychoactive drugs such as amphetamine and related compounds (**Figure 1**) showed strong activating proprieties against different CA isoforms.

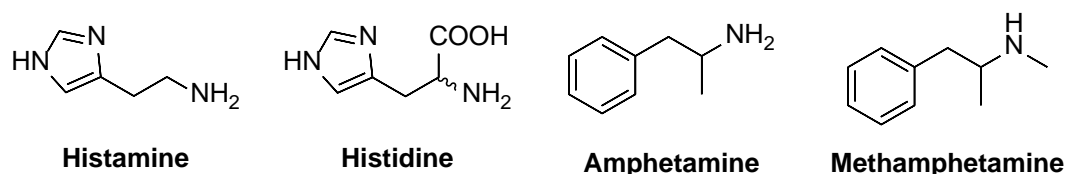


Figure 1. Examples of CAAs: Histamine, Histidine, Amphetamine and Methamphetamine.

In order to continue our studies in this less investigated field, we synthesized, for the first time, organic chalcogenides structurally related to the psychoactive drug Amphetamine. We sought to use N-H

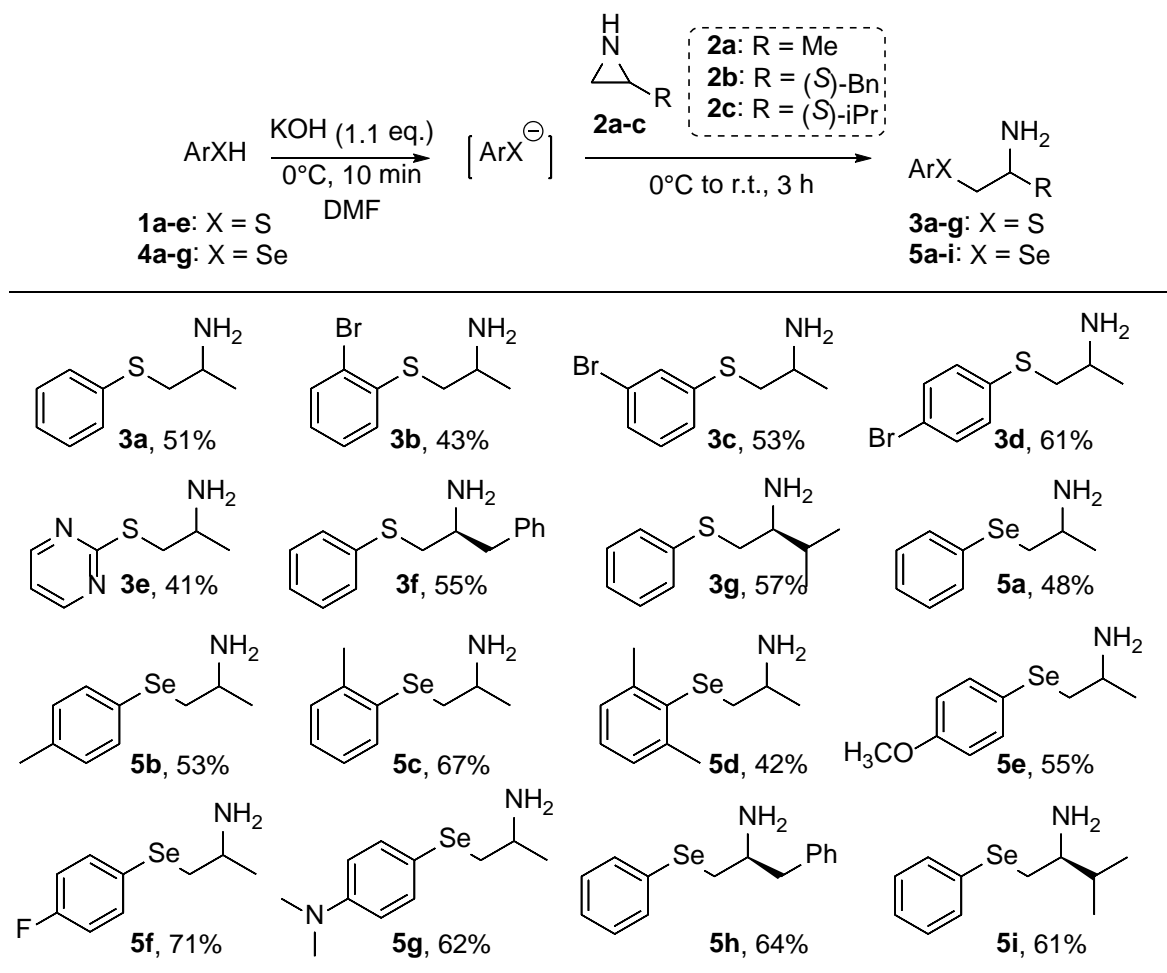
unactivated aziridines in reactions with suitable nucleophiles for preparing these derivatives. Indeed, ring opening reactions of strained heterocycles with chalcogen nucleophiles, including silyl chalcogenides²²⁻²⁷ represents a versatile way to obtain a large variety of biologically and synthetically valuable organosulfur,²⁸ organoselenium,²⁹ and organotellurium³⁰ compounds.

2. Results and discussion

2.1 Compounds Design and Synthesis.

We began our investigations by establishing the conditions required to obtain aminosulfide **2a** through the ring opening reaction of 2-methylaziridine **2a** with thiophenol **1a**. Among several organic or inorganic bases (Et₃N, DMAP, Cs₂CO₃/TBAI) and solvents (THF, CH₂Cl₂) evaluated, the use of KOH in DMF proved to be the most effective conditions in promoting the desired transformation, allowing to access **3a** in acceptable yield. Evaluation of different temperatures (-15°C, r.t.) gave no yield improvement. Having found suitable conditions for the synthesis of β-aminosulfides bearing a free amino function, we next explored the scope of this procedure. Thus, *o*-, *m*-, and *p*-bromothiophenol (**1b-d**) were smoothly converted into the corresponding β-aminosulfides **3b-d** upon reaction with **2a** under the same conditions. Interestingly, the yield proved to be strongly influenced by the position of the bromine on the aromatic ring, increasing from 44% (for the more hindered *o*-Br-C₆H₄ substituent) to 63% (for the less hindered *p*-Br-C₆H₄). Furthermore, the reaction of 2-mercaptopyrimidine **1e** with 2-methylaziridine resulted in the formation of the heteroaromatic aminosulfide **3e**. In order to evaluate the effect of the substituent on the stereogenic centre and to further enlarge the scope of the reaction, we also synthesized enantioenriched aminosulfides **3f,g** from thiophenol and (*S*)-*N*-H aziridines **2b,c**. Having obtained different novel sulfur-containing hCA activator candidates, we next evaluated the reactivity of selenols^{31,32} with 2-methylaziridine, in order to access new β-aminoselenides with potential activity as hCA activators. Therefore, a series of differently substituted aryl selenols (**4a-g**) were treated with aziridines **2a** in the presence of KOH to afford the corresponding β-aminoselenides **5a-g**.

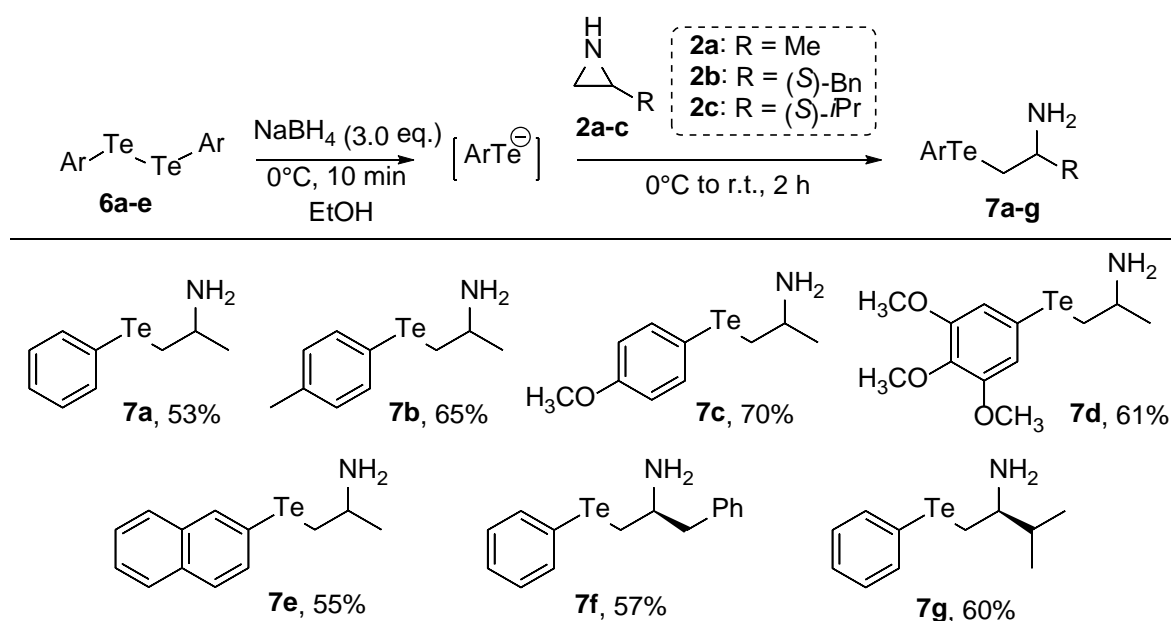
Furthermore, chiral enantioenriched β -phenylseleno amines **5h,i** were successfully achieved from the corresponding (*S*)-aziridines **2b,c** and benzeneselenol **4a**. These findings describe the first example of ring opening reaction of *N*-unactivated aziridines with stable aryl selenols.



Scheme 1: Synthesis of β -amino sulfides **3a-g** and β -amino selenides **5a-i** from *N*-H aziridines, Yields refer to isolated products

Having synthesised a wide variety of arylthio- and arylseleno-amines, we also investigated the reactivity of unprotected aziridines with tellurium-containing nucleophiles. Owing to the instability of tellurols, we sought to exploit the reactivity of *in situ* generated aryltellurolate anions. Thus, differently substituted diaryl ditellurides **6a-e** were treated with NaBH_4 and, then, reacted with *N*-H aziridine **2a**, affording the corresponding β -amino aryltellurides **7a-e**, bearing different substituted phenyl rings (**7a-d**) or the

naphthyl group (**7e**). Furthermore, enantioenriched β -phenyltelluro amines **7e,f** were achieved from ditelluride **6a** and (*S*)-2-benzyl- or (*S*)-2-isopropyl-aziridine, respectively.



Scheme 2. Synthesis of β -amino tellurides **7a-g** from *N*-H aziridines. Yields refer to isolated products.

2.2 Carbonic anhydrase activation.

To the best of our knowledge, this is the first study which evaluates the activation profile of different β -aminochalcogenides. For this reason, we investigated the CA activating properties of amines **3a-g**, **5a-i** and **7a-g** against 4 catalytically active and physiologically relevant hCA isoforms, the hCA I, II, VA and VII. Amphetamine (**AMP**) and histamine (**HIST**) were used as standard activators and shown in **Table 1**.

Table 1. Activation data of human CA isoforms I, II, VA and VII with compounds **3a-g**, **5a-i**, **7a-g**, **AMP** and **HIST** by a stopped flow CO₂ hydrase assay.³³

Cmp	K _A (μ M)*			
	hCA I	hCA II	hCA VA	hCA VII
3a	11.8	>100	12.4	23.4

3b	9.5	>100	13.3	23.2
3c	13.5	>100	12.9	11.4
3d	7.7	>100	12.4	14.8
3e	9.7	>100	43.7	21.8
3f	8.8	>100	10.2	14.8
3g	7.9	>100	11.6	14.3
5a	9.9	>100	14.7	20.6
5b	5.2	>100	9.1	10.4
5c	12.1	>100	20.0	14.2
5d	6.0	>100	20.9	21.4
5e	21.4	>100	20.3	23.3
5f	19.6	>100	13.1	13.4
5g	22.1	45.1	14.0	12.1
5h	6.3	>100	6.6	10.1
5i	5.9	>100	8.3	11.4
7a	9.9	>100	14.2	41.2
7b	8.2	>100	4.0	21.9
7c	4.6	>100	3.3	44.9
7d	9.5	>100	9.4	14.3
7e	12.6	>100	20.2	44.7
7f	5.1	>100	4.7	8.9
7g	13.3	>100	13.7	11.0
AMP^a	>100	>100	0.81	0.91
HIST^a	2.1	>100	0.01	37.5

* Mean from 3 different assays, by a stopped flow technique (errors were in the range of $\pm 5-10\%$ of the reported values). ^a Data from Ref.²¹

The following SAR can be observed regarding the activation data of **Table 1**:

i) The cytosolic, widespread isoform hCA II was not activated by these amines except for compound **5g** which showed an activation constant of 45.1 μM .

ii) The widely abundant cytosolic isozyme hCA I was moderately activated by most of the compounds here studied. β -Aminosulfides **3a-g** showed K_{AS} spanning between 7.7 to 13.5 μM . The presence of the bromine atom on aromatic ring plays a crucial role in modulation of activity. Indeed, bromine in position 3 decreased the potency almost 2 times for compound **3d** and 1.5 time when Br is placed in position 4 (**3d**). The replacement of the sulfur with selenium or tellurium in compounds **3a**, **5a** and **7a** did not influence significantly the activity (K_{AS} of 11.8 to 9.9 μM). On the other hand, this replacement from sulfur to tellurium influenced the potency for the analogous compounds **3f**, **5h** and **7f**. Indeed, such structures showed an increased activity moving within the chalcogen series, from sulfur to tellurium (K_{AS} 8.8 to 5.1 μM). A different trend was observed for β -aminochalcogenides with isopropyl moiety (**3g**, **5i** and **7g**). Compound **3g** showed a K_A of 7.9 μM and the activity increases with the passage of selenium analog **5i** (K_A 5.9 μM), but this time, organo-tellurium compound **7g** showed a less activating effects near two time than **3g** (K_A 13.3 μM).

iii) The mitochondrial isoform hCA VA, involved in different metabolic pathway such as ureagenesis, lipogenesis and neoglucogenesis,³⁴ was also activated by β -aminochalcogenides **3a-g**, **5a-I** and **7a-g** with K_{AS} ranging between 3.3 and 43.7 μM (**Table 1**). The best activators against this isoform were compounds with *para* substitution **7b** and **7c** with a K_{AS} of 4.0 and 3.3 μM respectively. Additional substitution (**7d**) or replacement with a more bulky scaffold (**7e**) decreases the activity (K_A 9.4 and 20.2 μM respectively). On the other hand, β -aminosulfides **3a-g** showed a flat SAR against hCA VA with K_{AS} ranging in a narrow interval of 10.2 to 13.3 μM except for compound **3e** with K_A of 43.7 μM . It

should be noted that, also for this isoform, replacing the chalcogen atom within the homologous series of compounds **3f**, **5h** and **7f** results in increased potency moving from sulfur- to tellurium-containing derivatives (K_{AS} 10.2 to 4.7 μM). These compounds remained, in any case, less effective on mitochondrial hCA compared to the psychotropic amine **AMP**.

iv) The brain-associated cytosolic CA isoform, hCA VII,³⁵ is moderately activated by compounds here reported, with activation constants in the range of 8.9-44.9 μM . The less active compounds (**7a**, **7c** and **7e**) contain a tellurium atom. Contrarily, all other analog compounds such as **3a**, **5a** and **5e** showed near two time more efficacy than them (K_{AS} 20.4-23.4 versus 41.2-44.9 μM). It is interesting to note there were not differences between the activating effects of β -aminosulfides **3a-g** and β -aminoselenides **5a-i**.

2.3 Antioxidant assays.

In order to develop novel hCA activators with antioxidant properties, we also evaluated the thiol peroxidase-like properties of selected β -arylchalcogeno amines. Therefore, the activity of sulfides **3a,c**, selenides **5a,e,i** and tellurides **7a-c,f** as mimics of glutathione peroxidase (GPx) was assessed by using the dithiothreitol (DTT) oxidation test³⁶⁻³⁹ and the GSH/GR coupled assay.³⁹⁻⁴⁰ Results of this investigation are reported in **Table 2**.

Table 2. Thiol peroxidase-like activity of β -arylchalcogeno amines according to different methods.

Entry	Compound	DTT (T_{50}) ^{a,b,c}	GSH (T_{50}) ^{a,d}
1	5a	442 (± 42)	24 (± 7)
2	5e	434 (± 38)	21 (± 6)
3	5i	398 (± 54)	28 (± 6)
4	5h	402 (± 48)	32 (± 9)
5	7a	<60	<5
6	7b	<60	<5
7	7c	<60	<5
8	7f	<60	<5
9	<i>N</i> -Ts- 5i	3186 (± 198)	129 (± 16)
10	<i>N</i> -Ts- 7f	600 (± 61)	14 (± 3)

[^a] T_{50} is the time required, in seconds, to reduce the initial thiol concentration with 50% after the addition of H₂O₂; data in parenthesis are the experimental error. [^b] DTT oxidation was monitored by the mean of ¹H NMR spectroscopy; 10 mol % of organoselenide and 1 mol % of organotelluride with respect to the thiol were used. [^c] T_{50} for PhSeSePh (commonly used reference compound) was found to be 796 (± 84) seconds, according to DTT oxidation test. [^d] NADPH consumption was monitored by UV spectroscopy (340 nm); 10 mol % of organoselenide and 1 mol % of organotelluride with respect to GSH were used.

Whilst aminosulfides did not show significant catalytic properties ($T_{50} \gg 24$ h for DTT assay), according both tests, all tested organoselenides and organo-tellurides exhibited remarkable catalytic antioxidant activity. Particularly, as expected, β -aryltelluro amines **7a-c,f** proved to behave as excellent catalysts, being able to promote DTT and GSH oxidation in 2 minutes and in 15 seconds, respectively.³⁹ As previously reported, aminoselenides and aminotellurides bearing a free amino function behave as better catalysts with respect to their *N*-Ts protected analogues (**Table 2**, entries 3, 7 vs entries 8, 9);³⁹ this suggests important criteria in order to maintain both the hCA activator and the GPx-like properties.

3. Conclusions

We have used a fast and easy synthetic strategy to obtain ring opening reactions of strained heterocycles with chalcogen-containing nucleophiles to afford novel CAA activators. Many of them showed an interesting pattern of activation against four of the most important physiologic carbonic anhydrase isoforms such as the cytosolic enzymes hCA I, II and VII, but also, against the mitochondrial isoform hCA VA. Compounds with selenium and in particular tellurium atoms are also able to prevent the generation of ROS metabolites that cause oxidative stress and cellular damage. In this context, CAAs may be used in the memory therapy and cognitive neurodegenerative disorders where the levels of ROS are particular high, but also in the metabolic pathologies where is present a decreased function of hCA VA without increased ROS levels.

4. Experimental Part

4.1. General

Anhydrous solvents and all reagents were purchased from Sigma-Aldrich, Alfa Aesar and TCI. All reactions involving air- or moisture-sensitive compounds were performed under a nitrogen atmosphere using dried glassware and syringes techniques to transfer solutions. Nuclear magnetic resonance (^1H -NMR, ^{13}C -NMR, ^{19}F NMR, ^{77}Se -NMR, and ^{125}Te) spectra were recorded using a Bruker Advance III 400 MHz spectrometer in $\text{DMSO-}d_6$. $(\text{PhSe})_2$ was used as an external reference for ^{77}Se NMR ($\delta = 461$ ppm). Diphenyl ditelluride $(\text{PhTe})_2$ was used as an external reference for ^{125}Te NMR ($\delta = 420$ ppm). Chemical shifts are reported in parts per million (ppm) and the coupling constants (J) are expressed in Hertz (Hz). Splitting patterns are designated as follows: s, singlet; d, doublet; t, triplet; q, quadruplet; m, multiplet; brs, broad singlet; dd, double of doublets. The assignment of exchangeable protons (OH and NH) was confirmed by the addition of D_2O . Analytical thin-layer chromatography (TLC) was carried out on Merck silica gel F-254 plates. Flash chromatography purifications were performed on Merck Silica gel 60 (230-400 mesh ASTM) as the stationary phase and ethyl acetate/ n -hexane were used as eluents. Melting points (mp) were measured in open capillary tubes with a Gallenkamp MPD350.BM3.5

apparatus and are uncorrected. The solvents used in MS measures were acetone, acetonitrile (Chromasolv grade), purchased from Sigma-Aldrich (Milan - Italy), and mQ water 18 M Ω , obtained from Millipore's Simplicity system (Milan-Italy). The mass spectra were obtained using a Varian 1200L triple quadrupole system (Palo Alto, CA, USA) equipped by Electrospray Source (ESI) operating in both positive and negative ions. Stock solutions of analytes were prepared in acetone at 1.0 mg mL⁻¹ and stored at 4°C. Working solutions of each analyte were freshly prepared by diluting stock solutions in a mixture of mQ H₂O/ACN 1/1 (v/v) up to a concentration of 1.0 μ g mL⁻¹. The mass spectra of each analyte were acquired by introducing, via syringe pump at 10 μ L min⁻¹, of the its working solution. Raw-data were collected and processed by Varian Workstation Vers. 6.8 software. The HPLC was performed by using a Waters 2690 separation module coupled with a photodiode array detector (PDA Waters 996) using a Nova-Pak C18 4 μ m 3.9 mm \times 150 mm (Waters) silica-based reverse phase column. The sample was dissolved in 10% acetonitrile/H₂O and an injection volume of 45 μ L. The mobile phase (flow rate 1.0 mL/min) was a gradient of H₂O + trifluoroacetic acid (TFA) 0.1% (A) and acetonitrile + TFA 0.1% (B), with steps as follows: (A%/B%), 0– 10 min 90:10, 10–25 min gradient to 60:40, 26:28 min isocratic 20:80, 29–35 min isocratic 90:10. TFA 0.1% in water as well in acetonitrile was used as counterion. All compounds reported here were \geq 95% HPLC pure.

4.2. General Procedure for the synthesis of β -arylthio- and β -arylseleno-amines:

Potassium hydroxide (1.1 eq.) was added to a solution of thiol 1a-e or selenol **4a-g** (1.0 eq.) in DMF (2 mL) under inert atmosphere (N₂) at 0°C. Then *N*-H aziridine **2a-c** (1.2 eq.) was slowly added and the mixture was stirred at 0°C for 3 h. Afterwards the reaction was diluted with EtOAc (5 mL) and saturated *aq.* NH₄Cl (2 mL) was added. The organic phase was collected and the aqueous phase was extracted with EtOAc (2 x 5 mL). The combined organic phases were dried over Na₂SO₄ and concentrated under reduced pressure. The residue was purified by flash column chromatography to yield the desired β -arylthioamine **3** or β -arylselenoamine **5**.

1-(Phenylthio)propan-2-amine (3a):

Following the general procedure, thiophenol **1a** (22 mg, 0.2 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-(phenylthio)propan-2-amine **3a** (17 mg, 51%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.23 (3H, d, $J = 6.3$ Hz), 2.49 (2H, bs, NH_2), 2.84 (1H, dd, $J = 7.9, 13.1$ Hz, $\text{CH}_a\text{H}_b\text{S}$), 3.07 (1H, dd, $J = 4.7, 13.1$ Hz, $\text{CH}_a\text{H}_b\text{S}$), 3.09-3.17 (1H, m, CHNH_2), 7.19-7.23 (1H, m), 7.28-7.32 (2H, m), 7.38-7.41 (2H, m). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.4, 43.6, 46.2, 126.3, 129.0, 129.8, 135.9. All recorded spectroscopic data matched those reported in the literature.⁴¹

1-((2-Bromophenyl)thio)propan-2-amine (3b)

Following the general procedure, 2-bromobenzenethiol **1b** (38 mg, 0.2 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-((2-bromophenyl)thio)propan-2-amine **3b** (21 mg, 43%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.23 (3H, d, $J = 6.3$ Hz), 2.23 (2H, bs, NH_2), 2.83 (1H, dd, $J = 7.9, 12.9$ Hz, $\text{CH}_a\text{H}_b\text{S}$), 3.05 (1H, dd, $J = 4.7, 12.9$ Hz, $\text{CH}_a\text{H}_b\text{S}$), 3.11-3.19 (1H, m, CHNH_2), 7.04 (1H, td, $J = 1.6, 7.9$ Hz), 7.23-7.28 (1H, m), 7.28, 7.33 (1H, m), 7.55 (1H, dd, $J = 1.0, 7.9$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.8, 43.0, 45.9, 124.1, 126.9, 127.7, 128.9, 133.0, 137.4. MS (ESI, positive): 246.2 $[M+H]^+$.

1-((3-Bromophenyl)thio)propan-2-amine (3c)

Following the general procedure, 3-bromobenzenethiol **1c** (47 mg, 0.25 mmol) and 2-methylaziridine **2a** (17 mg, 0.30 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-((3-bromophenyl)thio)propan-2-amine **3c** (33 mg, 54%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.18 (3H, d, $J = 6.3$ Hz), 1.86 (2H, bs, NH_2), 2.78 (1H, dd, $J = 7.9$, 13.0 Hz, $\text{CH}_a\text{H}_b\text{S}$), 3.02 (1H, dd, $J = 4.7$, 13.0, $\text{CH}_a\text{H}_b\text{S}$), 3.06-3.14 (1H, m, CHNH_2), 7.13 (1H, t, $J = 7.9$ Hz), 7.25- 7.31 (2H, m), 7.48 (1H, apt, $J = 1.8$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.8, 43.6, 46.0, 122.7, 127.5, 128.9, 130.1, 131.4, 138.7. MS (ESI, positive): 246.6 $[\text{M}+\text{H}]^+$.

1-((4-Bromophenyl)thio)propan-2-amine (3d)

Following the general procedure, 4-bromobenzenethiol **1d** (38 mg, 0.2 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-((4-bromophenyl)thio)propan-2-amine **3d** (30 mg, 61%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.13 (3H, d, $J = 6.3$ Hz), 1.78 (2H, bs, NH_2), 2.72 (1H, dd, $J = 8.0$, 13.1, $\text{CH}_a\text{H}_b\text{S}$), 2.96 (1H, dd, $J = 4.6$, 13.1, $\text{CH}_a\text{H}_b\text{S}$), 2.99-3.08 (1H, m, CHNH_2), 7.18 (2H, apt, $J = 8.5$ Hz), 7.35 (2H, apt, $J = 8.5$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.8, 43.9, 45.9, 119.8, 130.89, 131.84, 135.7. MS (ESI, positive): 246.3 $[\text{M}+\text{H}]^+$.

1-(Pyrimidin-2-ylthio)propan-2-amine (3e)

Following the general procedure, pyrimidine-2-thiol **1e** (23 mg, 0.2 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-(pyrimidin-2-ylthio)propan-2-amine **3e** (14 mg, 41%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.25 (3H, d, $J = 6.3$ Hz), 2.09 (2H, bs, NH_2), 3.09 (1H, dd, $J = 6.7$, 12.9 Hz, $\text{CH}_a\text{H}_b\text{S}$), 3.33 (1H, dd, $J = 4.9$, 12.9 Hz, $\text{CH}_a\text{H}_b\text{S}$), 3.25-3.32 (1H, m, CHNH_2), 6.96 (1H, t, $J = 4.8$ Hz), 8.50 (2H, d, $J = 4.8$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.5, 40.1, 46.8, 116.6, 157.2, 127.2. MS (ESI, positive): 169.9 $[\text{M}+\text{H}]^+$.

(S)-1-Phenyl-3-(phenylthio)propan-2-amine (3f)

Following the general procedure, thiophenol **1a** (22 mg, 0.2 mmol) and (*S*)-2-benzylaziridine **2b** (32 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:1), (*S*)-1-phenyl-3-(phenylthio)propan-2-amine **3f** (27 mg, 55%). All recorded spectroscopic data matched those reported in the literature.⁴²

(*S*)-3-Methyl-1-(phenylthio)butan-2-amine (3g)

Following the general procedure, thiophenol **1a** (28 mg, 0.25 mmol) and (*S*)-2-isopropylaziridine **2c** (26 mg, 0.30 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:1), (*S*)-3-methyl-1-(phenylthio)butan-2-amine **3g** (28 mg, 57%). All recorded spectroscopic data matched those reported in the literature.⁴¹

1-(Phenylselanyl)propan-2-amine (5a)

Following the general procedure, benzeneselenol **4a** (31 mg, 0.2 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-(phenylselanyl)propan-2-amine **5a** (21 mg, 48%).

¹H NMR (400 MHz, CDCl₃): δ (ppm) 1.19 (3H, dd, $J = 6.3, 12.4$ Hz), 2.45 (2H, bs, NH₂), 2.84 (1H, dd, $J = 7.7, 12.2$ Hz, CH_aH_bSe), 3.05 (1H, dd, $J = 4.8, 12.2$ Hz, CH_aH_bSe), 3.08-3.16 (1H, m, CHNH₂), 2.24-2.30 (3H, m), 7.52-7.56 (2H, m). ¹³C NMR (100 MHz, CDCl₃): δ (ppm) 23.1, 38.7, 46.7, 127.0, 129.1, 130.0, 132.8. ⁷⁷Se NMR(76 MHz, CDCl₃): δ (ppm) 254.1. MS (ESI, positive): 216.5 [*M*+H]⁺.

1-(*p*-Tolylselanyl)propan-2-amine (5b)

Following the general procedure, 4-methylbenzeneselenol **4b** (43 mg, 0.25 mmol) and 2-methylaziridine **2a** (17 mg, 0.30 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-(*p*-tolylselanyl)propan-2-amine **5b** (30 mg, 53%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.18 (3H, d, $J = 6.3$ Hz), 2.31 (3H, s), 2.48 (2H, bs, NH_2), 2.79 (1H, dd, $J = 7.9, 12.4$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 2.99 (1H, dd, $J = 4.8, 12.4$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 3.04-3.12 (1H, m, CHNH_2), 7.07 (2H, apd, $J = 8.0$ Hz), 7.42 (2H, apd, $J = 8.0$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 21.1, 23.0, 38.8, 46.7, 125.9, 133.4, 137.2. ^{77}Se NMR (76 MHz, CDCl_3): δ (ppm) 246.7. MS (ESI, positive): 230.2 $[M+\text{H}]^+$.

1-(*o*-Tolylselanyl)propan-2-amine (5c)

Following the general procedure, 2-methylbenzeneselenol **4c** (34 mg, 0.20 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-(*o*-tolylselanyl)propan-2-amine **5c** (31 mg, 67%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.20 (3H, d, $J = 6.3$ Hz), 2.08 (2H, bs, NH_2), 2.42 (3H, s), 2.80 (1H, dd, $J = 7.9, 12.2$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 3.01 (1H, dd, $J = 4.8, 12.2$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 3.07-3.15 (1H, m, CHNH_2), 7.09 (1H, td, $J = 1.7, 7.4$ Hz), 7.14 (1H, td, $J = 1.3, 7.4$ Hz), 7.14-7.19 (1H, m), 7.44 (1H, apd, $J = 7.6$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.4, 23.3, 37.6, 46.7, 126.5, 126.8, 130.0, 131.0, 131.8, 139.5. ^{77}Se NMR (76 MHz, CDCl_3): δ (ppm) 206.7. MS (ESI, positive): 229.8 $[M+\text{H}]^+$.

1-((2,6-Dimethylphenyl)selanyl)propan-2-amine (5d)

Following the general procedure, 2,6-dimethylbenzeneselenol **4d** (37 mg, 0.20 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-((2,6-dimethylphenyl)selanyl)propan-2-amine **5d** (20 mg, 42%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.21 (3H, d, $J = 6.4$ Hz), 2.58 (6H, s), 2.70 (1H, dd, $J = 7.3, 12.0$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 2.79 (1H, dd, $J = 5.5, 12.0$ Hz, $\text{CH}_a\text{H}_b\text{Se}$) 2.92-3.08 (3H, m, CHNH_2 partially overlapped con NH_2), 7.07-7.14 (3H, m). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.4, 24.7, 47.6, 127.7, 128.4, 136.5, 143.0. MS (ESI, positive): 244.1 $[M+\text{H}]^+$.

1-((4-Methoxyphenyl)selanyl)propan-2-amine (5e)

Following the general procedure, 4-methoxybenzeneselenol **4e** (56 mg, 0.30 mmol) and 2-methylaziridine **2a** (21 mg, 0.36 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-((4-methoxyphenyl)selanyl)propan-2-amine **5e** (40 mg, 55%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.17 (3H, d, $J = 6.3$ Hz), 2.40 (2H, bs, NH_2), 2.73 (1H, dd, $J = 8.0, 12.3$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 2.93 (1H, dd, $J = 4.7, 12.3$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 3.0-3.08 (1H, m, CHNH_2), 3.78 (3H, s, CH_3O), 6.80 (2H, apd, $J = 8.6$ Hz), 7.48 (2H, apd, $J = 8.6$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.9, 39.6, 46.6, 55.3, 114.8, 119.6, 135.7, 159.3. MS (ESI, positive): 245.8 $[M+H]^+$.

1-((4-Fluorophenyl)selanyl)propan-2-amine (5f)

Following the general procedure, 4-fluorobenzeneselenol **4f** (35 mg, 0.20 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-((4-fluorophenyl)selanyl)propan-2-amine **5f** (33 mg, 71%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.17 (3H, d, $J = 6.3$ Hz), 2.24 (2H, bs, NH_2), 2.78 (1H, dd, $J = 7.9, 12.3$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 2.97 (1H, dd, $J = 4.8, 12.3$ Hz, $\text{CH}_a\text{H}_b\text{Se}$), 3.01-3.1 (1H, m, CHNH_2), 6.95 (2H, apt, $J = 8.7$ Hz), 7.50 (2H, apdd, $J = 8.7$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 23.1, 39.5, 46.6, 116.3 (d, $^2J_{\text{C-F}} = 21.5$ Hz), 124.2 ($^4J_{\text{C-F}} = 3.4$ Hz), 135.4 ($^3J_{\text{C-F}} = 7.9$ Hz), 162.4 ($^1J_{\text{C-F}} = 247.1$ Hz). ^{77}Se NMR (76 MHz, CDCl_3): δ (ppm) 251.0. ^{19}F NMR (376 MHz, CDCl_3): δ (ppm) -114.6 (tt, $J = 5.4, 8.7$ Hz). MS (ESI, positive): 234.4 $[M+H]^+$.

4-((2-Aminopropyl)selanyl)-*N,N*-dimethylaniline (5g)

Following the general procedure, 4-(dimethylamino)benzeneselenol **4g** (40 mg, 0.20 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 4-((2-aminopropyl)selanyl)-N,N-dimethylaniline **5g** (32 mg, 62%).

¹H NMR (400 MHz, CDCl₃): δ (ppm) 1.18 (3H, d, $J = 6.4$ Hz), 2.64 (2H, bs, NH₂), 2.69 (1H, dd, $J = 8.2, 12.4$ Hz, CH_aH_bSe), 2.90 (1H, dd, $J = 4.7, 12.4$ Hz, CH_aH_bSe), 2.94 (6H, s, (CH₃)₂N), 3.01-3.09 (1H, m, CHNH₂), 6.61 (2H, apd, $J = 8.8$ Hz), 7.43 (2H, apd, $J = 8.8$ Hz). ¹³C NMR (100 MHz, CDCl₃): δ (ppm) 22.6, 39.4, 40.3, 46.7, 133.1, 113.9, 136.0, 150.2. MS (ESI, positive): 258.8 [M+H]⁺.

(S)-1-Phenyl-3-(phenylselanyl)propan-2-amine (5h)

Following the general procedure, benzeneselenol **4a** (31 mg, 0.2 mmol) and (*S*)-2-benzylaziridine **2b** (32 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:1), (*S*)-1-phenyl-3-(phenylselanyl)propan-2-amine **5h** (37 mg, 64%). All recorded spectroscopic data matched those reported in the literature.⁴²

(S)-3-Methyl-1-(phenylselanyl)butan-2-amine (5i)

Following the general procedure, benzeneselenol **4a** (39 mg, 0.25 mmol) and (*S*)-2-isopropylaziridine **2c** (26 mg, 0.30 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:1), (*S*)-3-methyl-1-(phenylselanyl)butan-2-amine **5i** (37 mg, 61%). All recorded spectroscopic data matched those reported in the literature.⁴³

4.3. General Procedure for the synthesis of β -aryltelluro amines

Sodium borohydride (3.0 eq.) was added to a suspension of diaryl ditelluride **6a-e** (1.0 eq.) in dry EtOH (2 mL) under inert atmosphere (N₂) at 0°C. Then *N*-H aziridine **2a-c** (2.4 eq.) was slowly added and the mixture was allowed to warm to room temperature and then stirred 2 h. Afterwards the reaction was

diluted with EtOAc (5 mL) and saturated *aq.* NH₄Cl (2 mL) was added. The organic phase was collected and the aqueous phase was extracted with EtOAc (2 x 5 mL). The combined organic phases were dried over Na₂SO₄ and concentrated under reduced pressure. The residue was purified by flash column chromatography to yield the desired β -aryltelluroamine **7**.

1-(Phenyltellanyl)propan-2-amine (**7a**)

Following the general procedure, 1,2-diphenylditellane **6a** (41 mg, 0.1 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-(phenyltellanyl)propan-2-amine **7a** (28 mg, 53%).

¹H NMR (400 MHz, CDCl₃): δ (ppm) 1.23 (3H, d, $J = 6.2$ Hz), 2.31 (2H, bs, NH₂), 2.95 (1H, dd, $J = 7.0, 11.8$ Hz, CH_aH_bTe), 3.11 (1H, dd, $J = 5.2, 11.8$ Hz, CH_aH_bTe), 3.14-3.21 (1H, m, CHNH₂), 7.2-7.24 (2H, m), 7.28-7.32 (1H, m), 7.75-7.78 (2H, m). ¹³C NMR (100 MHz, CDCl₃): δ (ppm) 21.7, 24.3, 47.8, 111.6, 127.7, 129.2, 138.4. ¹²⁵Te NMR (126 MHz, CDCl₃): δ (ppm) 390.5. MS (ESI, positive): 266.1 [$M+H$]⁺.

1-(*p*-Tolytellanyl)propan-2-amine (**7b**)

Following the general procedure, 1,2-di-*p*-tolylditellane **6b** (44 mg, 0.1 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-(*p*-tolytellanyl)propan-2-amine **7b** (36 mg, 65%).

¹H NMR (400 MHz, CDCl₃): δ (ppm) 1.19 (3H, d, $J = 6.2$ Hz), 2.15 (2H, bs, NH₂), 2.33 (3H, s), 2.87 (1H, dd, $J = 7.1, 11.9$ Hz, CH_aH_bTe), 3.04 (1H, dd, $J = 5.1, 11.9$ Hz, CH_aH_bTe), 3.08-3.15 (1H, m, CHNH₂), 7.01 (2H, apd, $J = 7.9$ Hz), 7.64 (2H, apd, $J = 7.09$ Hz). ¹³C NMR (400 MHz, CDCl₃): δ (ppm) 21.2, 22.0, 24.4, 47.7, 107.3, 130.2, 137.7, 138.8. ¹²⁵Te NMR (126 MHz, CDCl₃): δ (ppm) 377.1. MS (ESI, positive): 280.4 [$M+H$]⁺.

1-((4-Methoxyphenyl)tellanyl)propan-2-amine (7c)

Following the general procedure, 1,2-bis(4-methoxyphenyl)ditellane **6c** (47 mg, 0.1 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-((4-methoxyphenyl)tellanyl)propan-2-amine **7c** (41 mg, 70%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.21 (3H, d, $J = 6.3$ Hz), 2.86 (1H, dd, $J = 7.1, 12.0$ Hz, $\text{CH}_a\text{H}_b\text{Te}$), 2.92 (2H, bs, NH_2), 2.99 (1H, dd, $J = 5.5, 12.0$ Hz, $\text{CH}_a\text{H}_b\text{Te}$), 3.08-3.16 (1H, m, CHNH_2), 3.78 (3H, s, CH_3O), 6.75 (2H, apd, $J = 8.7$ Hz), 7.69 (2H, apd, $J = 8.7$ Hz). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 21.3, 23.9, 47.9, 55.1, 100.3, 115.2, 141.0, 159.8. ^{125}Te NMR (126 MHz, CDCl_3): δ (ppm) 379.8. MS (ESI, positive): 295.9 $[M+\text{H}]^+$.

1-((3,4,5-Trimethoxyphenyl)tellanyl)propan-2-amine (7d)

Following the general procedure, 1,2-bis(3,4,5-trimethoxyphenyl)ditellane **6d** (59 mg, 0.1 mmol) and 2-methylaziridine **2a** (14 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:2), 1-((3,4,5-trimethoxyphenyl)tellanyl)propan-2-amine **7d** (43 mg, 61%).

^1H NMR (400 MHz, CDCl_3): δ (ppm) 1.22 (3H, t, $J = 6.2$ Hz), 2.19 (2H, bs, NH_2), 2.93 (1H, dd, $J = 7.1, 11.8$ Hz, $\text{CH}_a\text{H}_b\text{Te}$), 3.09 (1H, dd, $J = 5.2, 11.8$ Hz, $\text{CH}_a\text{H}_b\text{Te}$), 3.13-3.21 (1H, m, CHNH_2), 3.83 (3H, s), 3.85 (6H, s) 6.97 (2H, s). ^{13}C NMR (100 MHz, CDCl_3): δ (ppm) 22.5, 24.5, 47.8, 56.3, 60.9, 104.2, 116.4, 138.4, 153.4. ^{125}Te NMR (126 MHz, CDCl_3) δ (ppm): 434.6. MS (ESI, positive): 356.4 $[M+\text{H}]^+$.

(S)-1-Phenyl-3-(phenyltellanyl)propan-2-amine (7f)

Following the general procedure, 1,2-diphenylditellane **6a** (41 mg, 0.1 mmol) and (*S*)-2-benzylaziridine **2b** (32 mg, 0.24 mmol) gave, after purification by flash column chromatography (petroleum

ether:EtOAc 1:1), (*S*)-1-phenyl-3-(phenyltellanyl)propan-2-amine **7f** (39 mg, 57%). All recorded spectroscopic data matched those reported in the literature.²⁶

(*S*)-3-Methyl-1-(phenyltellanyl)butan-2-amine (7g)

Following the general procedure, benzeneselenol **4a** (41 mg, 0.1 mmol) and (*S*)-2-isopropylaziridine **2c** (26 mg, 0.30 mmol) gave, after purification by flash column chromatography (petroleum ether:EtOAc 1:1), (*S*)-3-methyl-1-(phenyltellanyl)butan-2-amine **7g** (35 mg, 60%). All recorded spectroscopic data matched those reported in the literature.³⁹

4.4 Carbonic Anhydrase enzyme activation assay

An Sx.18Mv-R Applied Photophysics (Oxford, UK) stopped-flow instrument has been used to assay the catalytic activity of various CA isozymes for CO₂ hydration reaction.³³ Phenol red (at a concentration of 0.2 mM) was used as indicator, working at the absorbance maximum of 557 nm, with 10 mM HEPES (pH 7.5) as buffer and 0.1 M Na₂SO₄ (for maintaining constant ionic strength), following the CA-catalyzed CO₂ hydration reaction for a period of 10 s at 25 °C. The CO₂ concentrations ranged from 1.7 to 17 mM for the determination of the kinetic parameters and activation constants. For each activator at least six traces of the initial 5–10% of the reaction have been used for determining the initial velocity. The uncatalyzed rates were determined in the same manner and subtracted from the total observed rates. Stock solutions of activators (10 mM) were prepared in distilled-deionized water and dilutions up to 1 nM were done thereafter with the assay buffer. Activator and enzyme solutions were pre-incubated together for 15 min (standard assay at room temperature) prior to assay, in order to allow for the formation of the E–A complex. The activation constant (K_A), defined similarly with the inhibition constant K_I , can be obtained by considering the classical Michaelis–Menten equation (Eq. (1)), which has been fitted by non-linear least squares by using

PRISM 3:

$$V = \frac{V_{\max}}{\{1 + K_M/[S] (1 + [A]_f/K_A)\}} \quad (1)$$

Where $[A]_f$ is the free concentration of activator. Working at substrate concentrations considerably lower than K_M ($[S] \ll K_M$), and considering that $[A]_f$ can be represented in the form of the total concentration of the enzyme ($[E]_t$) and activator ($[A]_t$), the obtained competitive steady-state equation for determining the activation constant is given by Eq. (2):

$$V = \frac{V_0 K_A}{\left\{ K_A + ([A]_t - 0.5 \{ ([A]_t + [E]_t + K_A) - ([A]_t + [E]_t + K_A)^2 - 4[A]_t [E]_t \}^{1/2} \right\}} \quad (2)$$

Where V_0 represents the initial velocity of the enzyme-catalyzed reaction in the absence of activator.

The enzymes were recombinant ones, prepared in-house as reported earlier.²⁸⁻³¹

References

1. Supuran C.T., Structure and function of carbonic anhydrases. *Biochem J.* 2016; 14, 2023-2032.
2. Supuran C.T., Carbonic anhydrases: novel therapeutic applications for inhibitors and activators. *Nat Rev Drug Discov.* 2008, 2, 168-181.
3. Alterio V., Di Fiore A., D'Ambrosio K., Supuran C.T., De Simone G. Multiple binding modes of inhibitors to carbonic anhydrases: how to design specific drugs targeting 15 different isoforms? *Chem Rev.* 2012, 8, 4421-4468.
4. Supuran C.T., Carbonic anhydrases: from biomedical applications of the inhibitors and activators to biotechnological use for CO₂ capture. *J Enzyme Inhib Med Chem.* 2013, 2, 229-230.
5. Supuran C.T. How many carbonic anhydrase inhibition mechanisms exist? *J. Enzyme Inhib. Med. Chem.* 2016, 3, 345–360.
6. Berrino E., Supuran C.T., Novel approaches for designing drugs that interfere with pH regulation, *Expert Opin Drug Discov.* 2019, 3, 231-248.
7. Scozzafava A., Supuran C.T. Glaucoma and the applications of carbonic anhydrase inhibitors. *Subcell Biochem.* 2014, 75, 349-359.

8. Supuran C.T. Advances in structure-based drug discovery of carbonic anhydrase inhibitors. *Expert Opin Drug Discov.* 2017, 1, 61-88.
9. Aggarwal M., Kondeti B., McKenna R., Anticonvulsant/antiepileptic carbonic anhydrase inhibitors: a patent review. *Expert Opin Ther Pat.* 2013, 6, 717-724.
10. Supuran C.T. Carbonic anhydrase inhibition and the management of neuropathic pain. *Expert Rev Neurother.* 2016, 8, 961-968.
11. Angeli A., Di Cesare Mannelli L., Lucarini E., Peat T.S., Ghelardini C., Supuran C.T. Design, synthesis and X-ray crystallography of selenides bearing benzenesulfonamide moiety with neuropathic pain modulating effects. *Eur J Med Chem.* 2018;154, 210-219.
12. Di Cesare Mannelli L., Micheli L., Carta F., Cozzi A., Ghelardini C., Supuran C.T., Carbonic anhydrase inhibition for the management of cerebral ischemia: in vivo evaluation of sulfonamide and coumarin inhibitors. *J Enzyme Inhib Med Chem.* 2016, 6, 894-899.
13. Supuran C.T., Winum J.Y. Carbonic anhydrase IX inhibitors in cancer therapy: an update. *Future Med Chem.* 2015, 11, 1407-1414.
14. Supuran C.T. Carbonic anhydrase activators. *Future Med Chem.* 2018, 5, 561-573.
15. Briganti F, Mangani S, Orioli P, Scozzafava A, Vernaglion G, Supuran CT. Carbonic anhydrase activators: X-ray crystallographic and spectroscopic investigations for the interaction of isozymes I and II with histamine. *Biochemistry.* 1997, 34 10384-10392.
16. Akocak S, Lolak N, Vullo D, Durgun M, Supuran CT. Synthesis and biological evaluation of histamine Schiff bases as carbonic anhydrase I, II, IV, VII, and IX activators. *J Enzyme Inhib Med Chem.* 2017, 1, 1305-1312.
17. Mollica A, Macedonio G, Stefanucci A, Carradori S, Akdemir A, Angeli A, Supuran CT. Five- and Six-Membered Nitrogen-Containing Compounds as Selective Carbonic Anhydrase Activators. *Molecules.* 2017, 12, E2178.
18. Angeli A, Chiaramonte N, Manetti D, Romanelli MN, Supuran CT. Investigation of piperazines as human carbonic anhydrase I, II, IV and VII activators. *J Enzyme Inhib Med Chem.* 2018, 1, 303-308.
19. Stefanucci A, Angeli A, Dimmito MP, Luisi G, Del Prete S, Capasso C, Donald WA, Mollica A, Supuran CT. Activation of β - and γ -carbonic anhydrases from pathogenic bacteria with tripeptides. *J Enzyme Inhib Med Chem.* 2018, 1, 945-950.

20. Le Duc Y, Liesandru E, Vullo D, Barboiu M, Supuran CT. Carbonic anhydrases activation with 3-amino-1H-1,2,4-triazole-1-carboxamides: Discovery of subnanomolar isoform II activators. *Bioorg Med Chem.* 2017, 5, 1681-1686.
21. Angeli A, Vaiano F, Mari F, Bertol E, Supuran CT. Psychoactive substances belonging to the amphetamine class potently activate brain carbonic anhydrase isoforms VA, VB, VII, and XII. *J Enzyme Inhib Med Chem.* 2017, 1, 1253-1259.
22. Tanini, D.; Capperucci, A.; Degl'Innocenti, A. Bis-(trimethylsilyl)selenide in the Selective Synthesis of β -Hydroxy, β -Mercapto, and β -Amino Diorganyl Diselenides and Selenides Through Ring Opening of Strained Heterocycles. *Eur. J. Org. Chem.* 2015, 357–369.
23. Capperucci, A.; Tanini, D.; Borgogni, C.; Degl'Innocenti, A. Thiosilane- and organoselenosilane-mediated novel access to 3,7- disubstituted-1,2,5- trithiepanes and -1,2,5- dithiaselenepanes. *Heteroat. Chem.* 2014, 25, 678–683.
24. Tanini, D.; Grechi, A.; Dei, S.; Teodori, E.; Capperucci, A. An easy one-step procedure for the synthesis of novel β -functionalised tellurides. *Tetrahedron* 2017, 73, 5646–5653.
25. Capperucci, A.; Tanini, D. Silicon-assisted synthesis and functionalization of sulfurated and selenated compounds. *Phosphorus, Sulfur Silicon Relat. Elem.* 2015, 190, 1320–1338.
26. Braga, A. L.; Schwab, R. S.; Alberto, E. E.; Salman, S. M.; Vargas, J.; Azeredo, J. B. Ring opening of unprotected aziridines by zinc selenolates in a biphasic system. *Tetrahedron Letters*, 2009, 50, 2309-2311.
27. Silva, P. C.; Borges, E. L.; Lima, D. L.; Jacob, R. G.; Lenardão, E. J.; Perin, G.; Silva M. S. A simple and non-conventional method for the synthesis of selected β -arylalkylchalcogeno substituted alcohols, amines and carboxylic acids. *Arkivoc*, 2016, 5, 376-389.
28. Angeli, A.; Tanini, D.; Capperucci, A.; Malevolti, G.; Turco, F.; Ferraroni, M.; Supuran, C. T. Synthesis of different thio-scaffolds bearing sulfonamide with subnanomolar carbonic anhydrase II and IX inhibitory properties and X-ray investigations for their inhibitory mechanism. *Bioorg. Chem.* 2018, 81, 642-648.
29. Angeli, A.; Tanini, D.; Capperucci, A.; Supuran, C. T. Synthesis of novel selenides bearing benzenesulfonamide moieties as carbonic anhydrase I, II, IV, VII, and IX inhibitors. *ACS Med. Chem. Lett.* 2017, 8, 1213–1217.
30. Angeli, A.; Tanini, D.; Capperucci, A.; Supuran, C. T. First evaluation of organotellurium derivatives as carbonic anhydrase I, II, IV, VII and IX inhibitors. *Bioorg. Chem.* 2018, 76, 268-272.

31. Angeli, A.; Tanini, D.; Nocentini, A.; Capperucci, A.; Ferraroni, M.; Gratteri, P.; Supuran, C. T. Selenols: a new class of carbonic anhydrase inhibitors. *Chem. Commun.*, 2019, 55, 648-651.
32. Tanini, D.; Tiberi, C.; Gellini, C.; Salvi, P. R.; Capperucci A. A Straightforward access to stable β -functionalized alkyl selenols. *Adv. Synth. Catal.* 2018, 360, 3367 – 3375.
33. Khalifah R. G. The carbon dioxide hydration activity of carbonic anhydrase. I. Stop flow kinetic studies on the native human isoenzymes B and C. *J. Biol. Chem.* 1971, 246, 2561.
34. Supuran CT. Carbonic anhydrase inhibitors as emerging drugs for the treatment of obesity. *Expert Opin Emerg Drugs.* 2012, 1, 11-5.
35. Ruusuvoori E, Li H, Huttu K, Palva JM, Smirnov S, Rivera C, Kaila K, Voipio J. Carbonic anhydrase isoform VII acts as a molecular switch in the development of synchronous gamma-frequency firing of hippocampal CA1 pyramidal cells. *J Neurosci.* 2004, 11, 2699-707.
36. Iwaoka, M.; Kumakura, F. Applications of water-soluble selenides and selenoxides to protein chemistry. *Phosphorus, Sulfur Silicon Relat. Elem.* 2008, 183, 1009–1017.
37. Tanini, D.; D’Esopo, V.; Chen, D.; Barchielli, G.; Capperucci, A. Novel sulfur and selenium-containing antioxidants: Synthesis and evaluation of their GPx-like activity. *Phosphorus, Sulfur Silicon Relat. Elem.* 2017, 192, 166–168.
38. Menichetti, S.; Capperucci, A.; Tanini, D.; Braga, A. L.; Botteselle, G. V.; Viglianisi, C. A one-pot access to benzo[b][1,4]-selenazines from 2-aminoaryl diselenides. *Eur. J. Org. Chem.* 2016, 3097–3102.
39. Tanini, D.; Grechi, A.; Ricci, L.; Dei, S.; Teodori, E.; Capperucci, A. Novel functionalized organotellurides with enhanced thiol peroxidase catalytic activity. *New J. Chem.*, 2018, 42, 6077-6083.
40. Kumakura, F.; Mishra, B.; Priyadarsini, K. I.; Iwaoka, M. A watersoluble cyclic selenide with enhanced glutathione peroxidase-like catalytic activities. *Eur. J. Org. Chem.* 2010, 3, 440–445.
41. Ishibashi, H.; Uegaki, M.; Sakai M.; Takeda, Y. Base-promoted aminoethylation of thiols with 2-oxazolidinones: a simple synthesis of 2-aminoethyl sulfides. *Tetrahedron*, 2001, 57, 2115-2120.
42. Salman, S. M.; Narayanaperumal, S.; Schwab, R. S.; Bender, C. R.; Rodrigues O. E. D.; Dornelles, L. CuO nano particles and [bmim]BF₄: an application towards the synthesis of chiral β -seleno amino derivatives via ring opening reaction of aziridines with diorganyl diselenides. *RSC Adv.*, 2012, 2, 8478-8482.

43. Zielińska-Błajet, M.; Siedlecka R.; Skarzewski, J. Chiral phenylselenenyl derivatives of pyrrolidine and Cinchona alkaloids: nitrogen–selenium donating ligands in palladium-catalyzed asymmetric allylic alkylation. *Tetrahedron: Asymmetry*, 2007, 18, 131.