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The Ca^{2+}-ATPase Inhibition Potential of Gold(I, III) Compounds

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Abstract: The therapeutic applications of gold are well-known for many centuries. The most used gold compounds contain Au(I). Herein, we report, for the first time, the ability of four Au(I) and Au(III) complexes, namely dichloro (2-pyridinecarboxylate) Au(III) (abbreviated as **1**), chlorotrimethylphosphine Au(I) (**2**), 1,3-bis(2,6-diisopropylphenyl) imidazole-2-ylidene Au(I) chloride (**3**), and chlorotriphenylphosphine Au(I) (**4**), to affect the sarcoplasmic reticulum (SR) Ca^{2+}-ATPase activity. The tested gold compounds strongly inhibit the Ca^{2+}-ATPase activity with different effects, being Au(I) compounds **2** and **4** the strongest, with half maximal inhibitory concentration (IC_{50}) values of 0.8 and 0.9 μM, respectively. For Au(III) compound **1** and Au(I) compound **3**, higher IC_{50} values are found (4.5 μM and 16.3 μM, respectively). The type of enzymatic inhibition is also different, with gold compounds **1** and **2** showing a non-competitive inhibition regarding the native substrate MgATP, whereas for Au compounds **3** and **4**, a mixed type of inhibition is observed. Our data reveal, for the first time, Au(I) compounds with powerful inhibitory capacity towards SR Ca^{2+}ATPase function. These results also show, unprecedently, that Au (III) and Au(I) compounds can act as P-type ATPase inhibitors, unveiling a potential application of these complexes.

Keywords: Gold(I, III) compounds; Ca^{2+}-ATPase; P-type ATPases inhibitors; anticancer

1. Introduction

Gold compounds were used in medicine in ancient Egypt, Arabia, India, and China [1]. Later on, in Europe, Paracelsus (1493–1541) described the preparation of red colloidal gold by reduction of gold trichloride with an alcoholic extract of plants, followed by the addition of sugar or syrup. This preparation was named aurum potable, quinta essential auri, or oleum auri, being used in the treatment of several diseases (such as syphilis), for improving strength, prolonging life and rejuvenating. In 1618, Francisco Antonii published the first book on colloidal gold, which was preserved until nowadays. It contains information on the preparation of gold colloids and on its medical applications, including practical suggestions. In 1927, colloidal gold started to be used in rheumatoid arthritis (RA), and in 2013, Au(III) compounds were reported for the first time as Na^+/K^+-ATPase inhibitors [2,3]. Other medical applications of colloidal gold are described elsewhere [4].

Later, the well-known orally active Au(I)-phosphine-thiolate, a dicoordinate complex, also named auranofin, was developed to treat RA in clinical tests [2,5,6]. Aurothiomalate, aurothioglucose, and auro-bis(thiosulfate) are drugs with composition and structure similar to auranofin that have been extensively referred as anticancer agents [7–13].

The ligands present on gold complexes are also a subject of design. In recent years, anti-cancer-active Au(I) complexes containing multidentate *N*-donor ligands (e.g., 2,2′-terpyridine;

6′,2″-terpyridine, 2,2′-bipyridine, porphyrin), cyclometalating or dithiocarbamate ligands were reported [14]. Gold(I) complexes containing phosphine, *N*-heterocyclic carbene (NHC), thiolate, alkynyl, thiourea, triazole-peptide or other ligands were also studied for the same purpose [15]. Additionally, gold compounds have been a subject of research in terms of antibacterial, antivirus and anti-parasite activity [12,13,16,17] and in Alzheimer's disease [18]. Gold compounds, together with vanadium complexes and polyoxometalates, have been selected by some researchers as alternative anti-tumor substances with promising results in tumor growth suppression [19–23]. Moreover, some of these studies used gold nanoparticles functionalized with polyoxometalates, which are highly uptaken by cancerous cells [13].

Although the biological effects of gold compounds, either Au(I) or Au(III), are known, their molecular mechanisms are largely unknown. It is possible to define three major classes according to their mode of action towards biological targets: (1) Involving an alkylation mechanism: Coordination to biomolecules side chains, e.g., thiols, imidazole and selenols, after activation (gold compounds are prodrugs), through the release of labile ligands; e.g., auranofin; (2) As DNA intercalators: Gold complexes are capable of crossing membranes and binding noncovalently to proteins, enzymes, DNA, some examples being gold(III) porphyrins; (3) In redox reactions with biomolecules causing oxidative damage, e.g., Auoxo6 [24]. Moreover, the antitumor activity of these inorganic compounds is also attributed, at least in part, to the inhibition of certain protein functions, for example aquaporin [22,23], P-type ATPases [3] and protein tyrosine phosphatases, among others [7–13]. Some of them, like Na^+/K^+-ATPase and Ca^{2+}-ATPase, are putative pharmacological targets of a substantial number of drugs, well-known as ion pumps inhibitors [25–28]. To the best of our knowledge, only a few studies describe the use of gold compounds as Na^+/K^+-ATPase inhibitors (including [3,29]).

In this work, we describe and compare, for the first time, the effects of both gold(I) and gold(III) compounds in P-type ATPases, particularly in Ca^{2+}-ATPase function. The compounds used are shown in Figure 1, dichloro (2-pyridinecarboxylate) gold (**1**) being the only gold(III) compound, the other three are gold(I) complexes. They were successfully used as catalysts in the oxidation of alkanes and alcohols in a previous work [30].

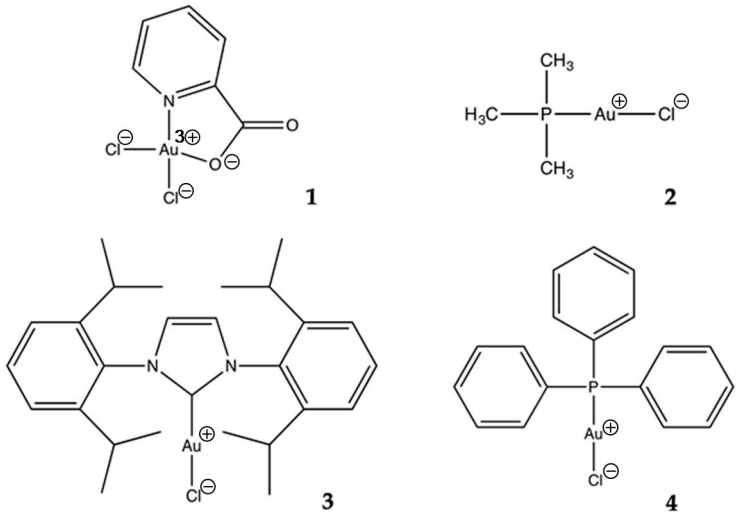

Figure 1. Gold complexes (**1**) dichloro (2-pyridinecarboxylate) gold(III); (**2**) chlorotrimethylphosphine gold(I); (**3**) 1,3-bis(2,6-diisopropylphenyl) imidazole-2-ylidene gold(I) chloride and (**4**) chlorotriphenylphosphine gold(I), showing the formal charge distribution.

The aims of the present study are: (i) to test the extent of Ca^{2+}-ATPase inhibition of gold complexes **1**–**4**; (ii) to characterize the type of Ca^{2+}-ATPase inhibition regarding the protein native substrate MgATP; (iii) to compare the inhibition capacity of the gold complexes with other metal compounds

and with well-known drugs that target P-type ATPases; (iv) to obtain a more accurate description of the targets involved in the mechanism of action of gold complexes and their possible biological effects.

2. Results

Inhibition of Ca^{2+}-ATPase by Gold Compounds

The effects of four gold compounds on the activity of the sarcoplasmic reticulum (SR) Ca^{2+}-ATPase was investigated for the first time (results shown in Figures 2 and 3). Compounds were added to the assay just a few seconds before the beginning of the experiment and the reaction time was 3 min. All the Au complexes inhibited Ca^{2+}-ATPase activity, which is expressed as percentage of the control enzyme value obtained without inhibitor, in a concentration dependent manner. The inhibitory capacity of the investigated gold compounds was evaluated by the half maximal inhibitory concentration (IC_{50}) values, meaning that the inhibitor concentration induced 50% of Ca^{2+}-ATPase inhibition of the enzyme activity. The IC_{50} values ranged from 0.8 to 16.3 μM. Gold(I) compounds **2** and **4** showed the lowest IC_{50} values: 0.8 and 0.9 μM, respectively, indicating a higher inhibition (Figure 2A,B). The gold(III) compound **1** and gold(I) compound **3** exhibited IC_{50} values in the range of 4–16 μM (Figure 3A,B). Thus, the Au(I) complexes **2** and **4** were, respectively, around 6–20 fold more powerful inhibitors of the ATPase than compounds **1** and **3** (Table 1).

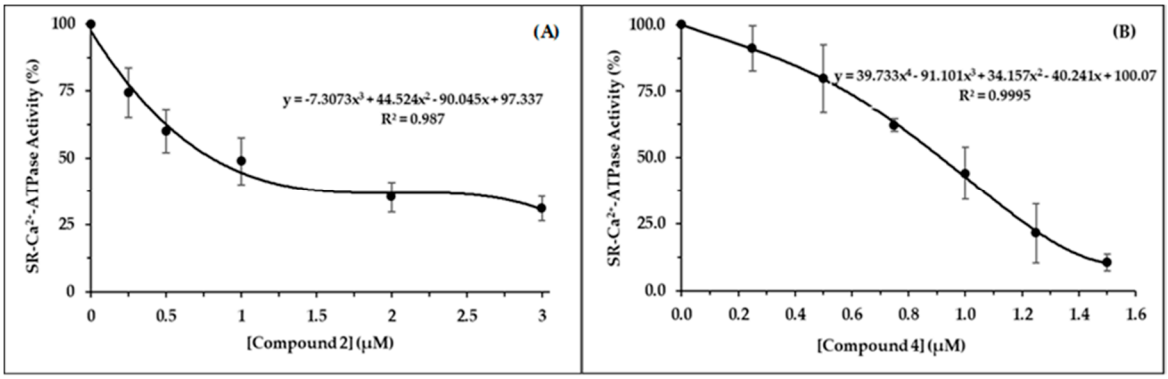

Figure 2. Inhibition of Ca^{2+}-ATPase activity by gold compounds **2** (**A**) and **4** (**B**). Ca^{2+}-ATPase was measured spectrophotometrically at 340 nm and 25 °C, using the coupled enzyme pyruvate kinase/lactate dehydrogenase assay. The experiments were initiated after the addition of 10 μg/mL calcium ATPase. Data are plotted as means ± SD. The results shown are the average of triplicate experiments carried out in distinct preparations.

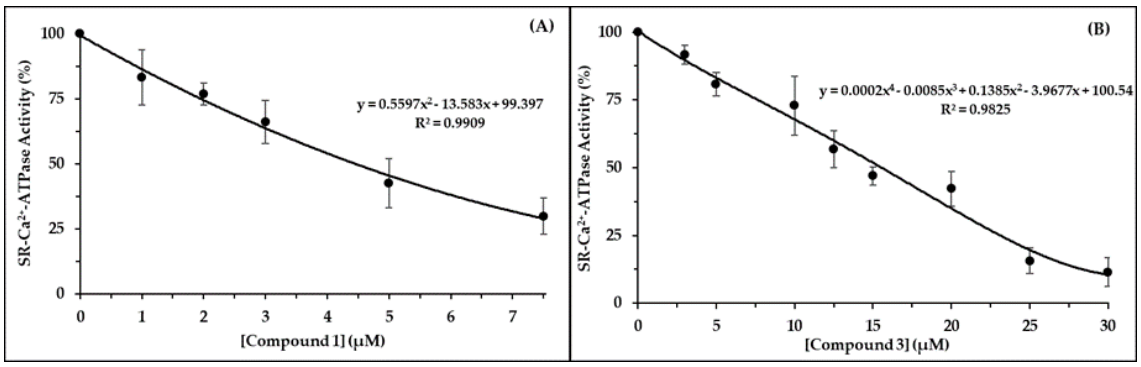

Figure 3. Inhibition of Ca^{2+}-ATPase activity by gold compounds **1** (**A**) and **3** (**B**). Experiments performed in the same conditions as Figure 2 assays.

Table 1. K_m, V_{max} and IC_{50} values of Ca^{2+}-ATPase inhibition by gold compounds.

Gold Compound		K_m (mM)	V_{max} (nM ATP·min^{-1})	Type of Inhibition	IC_{50} (μM)
1	0 (μM)	0.243 ± 0.024 [a]	67.57 ± 8.85 [a]	non-competitive	4.5 ± 0.1 [a]
	4 (μM)	0.255 ± 0.019 [a]	30.77 ± 5.34 [b]		
2	0 (μM)	0.098 ± 0.011 [b]	51.55 ± 4.38 [c]	non-competitive	0.8 ± 0.1 [b]
	1 (μM)	0.096 ± 0.012 [b]	41.67 ± 3.31 [d]		
3	0 (μM)	0.257 ± 0.021 [a]	75.76 ± 6.87 [e]	mixed	16.3 ± 0.1 [c]
	15 (μM)	0.421 ± 0.025 [c]	60.98 ± 2.98 [f]		
4	0 (μM)	0.210 ± 0.016 [d]	42.02 ± 2.78 [d]	mixed	0.9 ± 0.1 [b]
	1 (μM)	0.281 ± 0.020 [e]	22.62 ± 2.08 [g]		

[a–d] Averages with different letters in same column are significantly different ($p < 0.05$).

It is worth noting that the presence of gold compounds, under the conditions used in the enzymatic assays, does not affect the basal activity of the ATPase (see Material and Methods section). This means that, for the concentrations used in the experiments, the gold compounds do not induce uncoupling of the process. This also means that vesicles containing the ATPase maintain their integrity, thus ATP hydrolysis and calcium transport are coupled, and the inhibitory capacity can be evaluated after the addition of the ionophore.

Using concentrations near the IC_{50} values displayed in Table 1, the type of inhibition of the four gold compounds was analyzed, regarding the native substrate of the ATPase, MgATP. It was observed that both Au(III) compound **1** and also Au(I) compound **2** present a non-competitive type of inhibition (Table 1, Figure 4A,B). It can be seen that the values of K_m obtained with complexes **1** and **2** are very similar to those observed for the control (in the absence of compounds), whereas V_{max} values decreased (Table 1, Figure 4). On the other hand, it can be observed that both Au(I) compounds **3** and **4** present a mixed type inhibition, once their V_{max} values decrease, compared to the control, whereas K_m values increase (Table 1, Figure 5A,B). Thus, it can be suggested that both Au(I) compounds **3** and **4** can interact with Ca^{2+}-ATPase, whether or not the enzyme has already bound to the substrate, pointing out two distinct protein binding sites for these complexes.

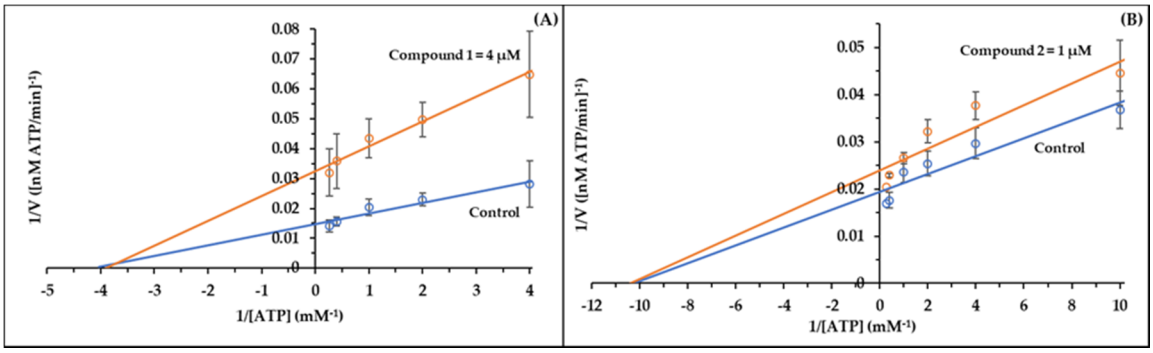

Figure 4. Lineweaver-Burk plots of Ca^{2+}-ATPase activity in the absence (blue) and in the presence (orange) of 1 or 4 μM of the gold compounds. (**A**) Gold compound **1**; (**B**) Gold compound **2**. The plots were used for determining the type of enzyme inhibition. The gold complexes presented a non-competitive type of inhibition. Data are plotted as means ± SD. The results shown are the average of triplicate experiments, carried out in distinct preparations.

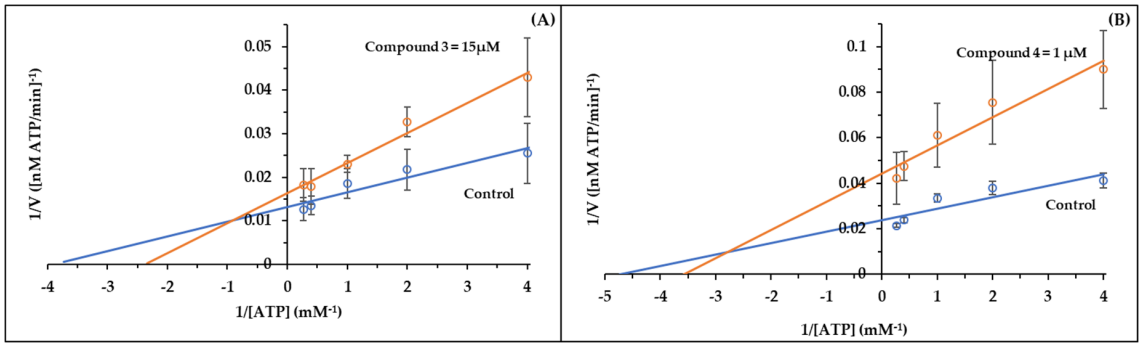

Figure 5. Lineweaver-Burk plots of Ca^{2+}-ATPase activity in the absence (blue) and in the presence (orange) of 1 or 15 μM of the gold compounds (**A**) Gold compound **3**; (**B**) Gold compound **4**. The plots were used for determining the type of enzyme inhibition. The gold complexes presented a mixed type of inhibition. Data are plotted as means ± SD. The results shown are the average of triplicate experiments carried out in distinct preparations.

Moreover, compounds were incubated for 30 min with and without the protein and the inhibitory effects of the ATPase were the same as without any incubation, suggesting that the Au compounds are stable, and the effects observed can be attributed their addition to the medium.

3. Discussion

We studied the effects of the four gold compounds mentioned above (Figure 1, Table 2 of Materials and Methods section) on the activity of sarcoplasmic reticulum (SR) Ca^{2+}-ATPase. It is worth noting that the gold compounds solutions prepared in DMSO are colorless and the UV-Vis spectra obtained after 1 min is the same as the measured after 30 min, showing their stability.

Table 2. Gold compounds used in the present study (structures can be found in Figure 1).

Formula	Abbreviation	Net Charge	MW (g/mol)	CAS Number
$C_6H_4NAuCl_2O_2$	1	+3	389.97	88215-41-2
C_3H_9PAuCl	2	+1	308.50	15278-97-4
$C_{27}H_{36}AuClN_2$	3	+1	621.01	852445-83-1
$C_{18}H_{15}PAuCl$	4	+1	494.71	14243-64-2

Gold(I) compounds **2** and **4** strongly inhibit Ca^{2+}-ATPase, showing IC_{50} values of 0.8 and 0.9 μM. Using exactly the same experimental conditions as those of the present study, similar Ca^{2+}-ATPase IC_{50} values of inhibition were previously determined for polyoxotungstates (POTs), such as P_2W_{18} (0.6 μM) and for Se_2W_{29} (IC_{50} = 0.3 μM), as well as for polyoxovanadates (POVs), such as PV_{14} (IC_{50} = 1 μM) [31,32]. However, a lower capacity was observed for some POTs, such as TeW_6 (IC_{50} = 200 μM) [31]. Inhibitory power (IC_{50}) ranging from 1 to 35 μM was reported for decaniobate $[Nb_{10}O_{28}]^{6-}$ (IC_{50} = 35 μM) and for Keggin-based polyoxotungstates (POTs), such as mono-substituted CoW_{11} (IC_{50} = 4 μM), tri-lacunary SiW_9 (IC_{50} = 16 μM) and AsW_9 (20 μM), lacunary Dawson type P_2W_{12} (11 μM), and also for As_2W_{19} (28 μM) [31]. Note that the P-type ATPases have long been known to be inhibited by metal complexes although the structural details of its inhibitory mechanism remain unresolved [27,29,31–35]. Regarding the Ca^{2+}-ATPase, it was previously determined that polyoxovanadates, such as decavanadate (V_{10}) (IC_{50} = 15 μM), are more powerful Ca^{2+}-ATPase inhibitors than monomeric vanadate (V_1) (IC_{50} = 80 μM). Herein, we show that gold(I) compounds **2** and **4** are stronger inhibitors of the calcium pump (IC_{50} < 1 μM), whereas gold(III) compound **1** and gold(I) compound **3**, although with less capacity, can be also considered as complexes with high affinity for the ATPase (IC_{50} < 16 μM).

Both decaniobate (Nb_{10}) and decavanadate (V_{10}) showed to be Ca^{2+}-ATPase non-competitive inhibitors regarding the natural ligand MgATP [34]. The same type of inhibition (non-competitive) was observed in this work for gold(III) compound **1** and gold(I) compound **2**, as they did not bind to ATP binding site. On the other hand, the gold(I) compounds **3** and **4** presented a mixed type inhibition, as previously observed for POTs and POVs, such as P_2W_{18} and PV_{14} [31,32]. This observation suggests that these gold(I) compounds can interact with the Ca^{2+}-ATPase, whether or not the enzyme has already bound substrate, and point out to the existence of two distinct protein binding sites for these types of gold compounds, one of them probably being the ATP binding site.

The non-competitive type of inhibition observed for gold compounds **1**–**4** is in agreement with previous studies dealing with Na^+/K^+-ATPase. For example, it was previously described that gold(III) complexes $H[AuCl_4]$, $[Au(DMSO)_2Cl_2]Cl$, and $[Au(bipy)Cl_2]Cl$ (bipy = 2,2′-bipyridine), inhibited the enzymatic activity of a commercially available Na^+/K^+-ATPase, with IC_{50} values of, respectively, 0.7, 5.5, and 39.8 μM [3]. It was also reported that this inhibition can be prevented using –SH donating ligands, such as L-Cys and glutathione. Moreover, a non-competitive mode of the interaction was referred for all compounds [3]. In contrast to Au salts, no ionic Au is present in solution when those complexes are used. Those organic gold compounds tend to undergo ligand exchange reactions with protein thiols, which may be responsible for the inhibitory properties. In our study, the gold(I) compound **2** (that presents higher inhibitory potential for the Ca^{2+}-ATPase) showed a non-competitive type of inhibition, suggesting that it does not bind to the ATP binding site. However, gold(I) compound **4**, showed a similar inhibitory potential, but with a mixed type of inhibition.

It is worth mentioning that within a physiological pH range (required for optimal activity of Ca^{2+}-ATPases and in the majority of biological investigations), the gold compounds are not expected to undergo partial hydrolysis as it occurs for instance with polyoxometalates studies, where the chemical species inducing the effects should be ascertain [36]. However, in the experiments referred to above, dealing with the determination of the type of inhibition and IC_{50} values, it was observed that the incubation of the gold(I) or gold(III) compounds in the medium did not affect the inhibition of the enzyme, suggesting that the compounds are stable under the experimental conditions used. Still, although further investigations are needed, it can be assumed that the intact gold compounds are responsible for the effects observed in the Ca^{2+}-ATPase, as already referred.

It was suggested by Bondžić et al. that gold(III) compounds, i.e., $[Au(bipy)(OH)_2][PF_6]$, bipy = 2,2′-bipyridine; $[Au(py^{dmb}\text{-}H)(CH_3COO)_2]$, py^{dmb}-H = deprotonated 6-(1,1-dimethylbenzyl)-pyridine and $[Au(bipy^{dmb}\text{-}H)(OH)][PF_6]$, $bipy^{c}$-H = deprotonated 6-(1,1-dimethylbenzyl)-2,2′-bipyridine, can bind to the E1 conformation of the Na^+/K^+-ATPase [29]. The different binding modes of these gold(III) complexes to the enzyme were explained based on their distinctive structural features. In contrast to gold(III), vanadate only binds to the E2 conformation of the Ca^{2+}-ATPase (another P-type ATPase), whereas decavanadate (V_{10}) strongly binds to either E1 or E2 conformations, being phosphorylated or not [34]. However, to the majority of the inorganic compounds, the protein conformations and binding sites are still to be determined [33,34]. For some drugs, such as thapsigargin (TG) and cyclopiazonic acid (CPA), the mechanisms of action and ATPases binding sites are clearly established [28].

In fact, several drugs are known as ionic pump inhibitors, such as ouabain, omeprazole or thapsigargin. For the Ca^{2+}-ATPase, TG (IC_{50} = 0.001–0.029 μM), CPA (IC_{50} = 0.1–0.2 μM), macrocyclic lactones (IC_{50} = 66–72 μM), curcuminoides (IC_{50} = 7–17 μM) and celecoxib (IC_{50} = 35 μM) are examples of well-known inhibitors [37–41]. These organic inhibitors of the Ca^{2+}-ATPases, are used for several disease treatments, such as heart failure, psychosis, malaria and also as anaesthetics, tumour inhibitors, antibiotics and insulin mimetic agents [37–41], presenting inhibitory capacity not so different from the gold compounds described in the present study. Herein, we aimed to compare the inhibitory capacity of complexes **1**–**4** with oxometalates, metal complexes, polyoxometalates (POTs and POVs), and distinguish between gold(I) and gold(III) compounds (IC_{50} = 0.8–16.3 μM). Thus, it was described that several inorganic compounds and complexes present Ca^{2+}-ATPases IC_{50} inhibition values globally not so different from the above well-known Ca^{2+}-ATPases drugs inhibitors, for example decavanadate

(IC_{50} = 15 μM), polyoxotungstates (IC_{50} = 0.3–200 μM), polyoxovanadates (IC_{50} =1 μM), and vanadium complexes such as BMOV (IC_{50} = 40 μM), among others [31–35]. In summary, both gold(I) and gold(III) compounds **1**–**4** are strong inhibitors of the SR Ca^{2+}-ATPase, pointing out this enzyme as a putative target for gold compounds with anticancer activity, described as promising agents for the future in medicinal chemistry [1–18,23,29,42–44].

4. Materials and Methods

4.1. Gold Compounds

Au(I) and Au(III) complexes were used (Table 2). Their chemical structures can be found in Figure 1. Dichloro(2-pyridinecarboxylate)gold(III) (**1**) was purchased from Aldrich. The Au(I) complexes chlorotrimethylphosphinegold(I) (**2**), 1,3-bis(2,6-diisopropylphenyl)imidazol-2-ylidenegold(I) chloride (**3**) and chlorotriphenylphosphinegold(I) (**4**) (Figure 1, Table 2), were purchased from Strem Chemicals (Newburyport, MS, USA).

Stock solutions of the gold complexes (10 mM) were freshly prepared by dissolving the solid compounds in dimethyl sulfoxide (DMSO) and keeping the solution at room temperature before use. Wherever adequate, the freshly prepared solutions were diluted to 1 and/or 0.1 mM final concentrations before use in the enzymatic assays.

4.2. Preparation of Sarcoplasmic Reticulum Ca^{2-} ATPase Vesicles

Sarcoplasmic reticulum (SR) Ca^{2+}-ATPase-1 (SERCA-1) was prepared from rabbit skeletal muscles as described elsewhere [34]. The vesicles were suspended in 0.1 M KCl, 10 mM HEPES (pH 7.0), diluted 1:1 with 2.0 M sucrose and frozen in liquid nitrogen for storage at −80 °C. As analyzed by SDS-PAGE (Bio-Rad Laboratories, Inc., Hercules, CA, USA), Ca^{2+}-ATPase constituted at least 70% of the total protein.

4.3. Effects of Gold Compounds in the ATP Hydrolysis by the SR Ca^{2+}-ATPase

Ca^{2+}-ATPase activities were measured spectrophotometrically (Shimadzu, UV 2401-PC, Shimadzu Corporation, Kyoto, Japan) at 22 °C, using the coupled enzyme pyruvate kinase/lactate dehydrogenase assay, as described elsewhere [21,31,32,34,35]. The reaction medium contained: 25 mM HEPES (pH 7.0), 100 mM KCl, 5 mM $MgCl_2$, 50 μM $CaCl_2$, and 2.5 mM ATP. For the coupled enzyme assay 0.42 mM phosphoenolpyruvate, 0.25 mM NADH, 18 IU lactate dehydrogenase and 7.5 IU pyruvate kinase were added to the medium. The experiments were initiated by the addition of 10 μg/mL calcium ATPase, and the absorbance at 340 nm was recorded during about 1 min (basal activity). Without stopping the reaction, 4% (*w*/*w*) of calcium ionophore A23187 were added to the cuvette and the enzyme kinetics of ATP hydrolysis was followed for more 2 min. Wherever adequate, freshly prepared gold compounds solutions were added to the medium immediately prior to Ca^{2+}-ATPase addition. Finally, the ATPase activities were measured, taken into consideration the slope of the decrease of the absorbance (at 340 nm) per minute, in the absence (100%) or in the presence of the inhibitor. All experiments were performed at least in triplicate. The inhibitory power of the investigated gold compounds was evaluated determining IC_{50} values, meaning the gold compound concentration inducing 50% inhibition of Ca^{2+}-ATPase enzyme activity. The IC_{50} values were determined according to the equations shown in the graphics obtained after the best fitting of the experimental points.

4.4. Statistical Analysis

Calculations of IC_{50} were performed using Microsoft Office 365 Excel (2019) (Microsoft, Redmond, WA, USA). All values shown are presented as averages and standard deviations of measurements taken from triplicate measurements, using three distinct and independent Ca^{2+}-ATPase preparations. The statistical significance of the data was assessed using the Student's *t*-test. Differences from controls were considered significant, at $p < 0.05$.

5. Conclusions

Sarcoplasmic reticulum Ca^{2+}-ATPase activity was inhibited by both Au(I) or Au(III) compounds. Particularly significant inhibition values were found for the Au(I) compounds **2** and **4** ($IC_{50} < 1$ μM), similar to those previously described for POTs. A non-competitive type of inhibition was found for the smaller gold(I) compound **2**, which was the most powerful ATPase inhibitor, even superior to the well-known inorganic P-type ATPase non-competitive inhibitors, such as decavanadate ($IC_{50} = 15$ μM). A similar inhibiting potential, but with a mixed type of inhibition, was found for gold(I) compound **4**, suggesting that the different complexes have different modes of interaction with the Ca^{2+}-ATPase, however maintaining the high affinity for the enzyme. Although more investigations are needed to establish structure-activity relationships, both Au(I) or Au(III) compounds can be considered as responsible for the observed Ca^{2+}-ATPase inhibitory effect, showing good inhibitory capacity, suggesting this enzyme to be a potential gold target for future medicinal inorganic chemistry (Figure 6).

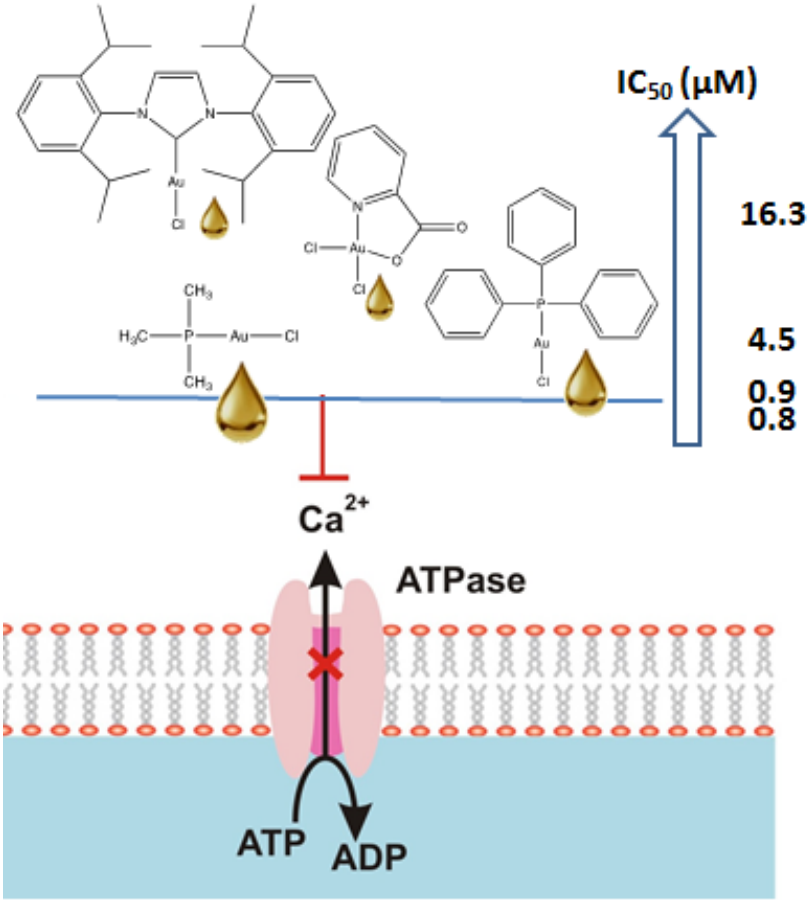

Figure 6. Gold compounds interacting with P-type ATPases, a putative target for gold anticancer activities.

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Conflicts of Interest: The authors declare no conflict of interest.

Abbreviations

ATPase	Adenosine triphosphatase
Auoxo6	[(6,6′-dimethyl-2,2′-bipyridine)$_2$Au$_2$(μ-O)$_2$][PF$_6$]$_2$
CPA	Cyclopyazonic
DNA	Deoxyribonucleic acid
DMSO	Dimethyl sulfoxide
IC_{50}	Half maximal inhibitory concentration
POTs	Polyoxotungstates
POVs	Polyoxovanadates
RA	Rheumatoid arthritis
SR	Sarcoplasmic reticulum
TG	Thapsigargin

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