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# Advances in thiosemicarbazone metal complexes as anti-lung cancer agents

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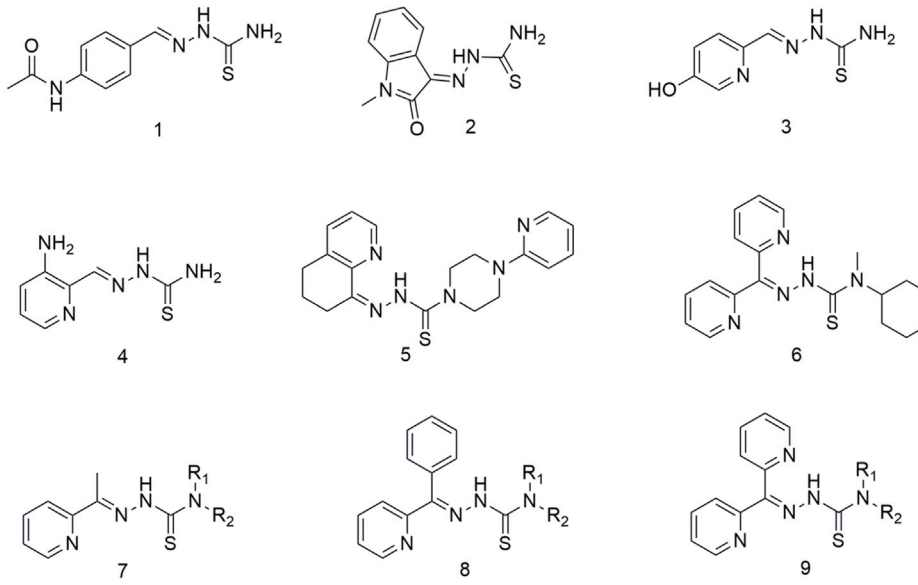
The great success of cisplatin as a chemotherapeutic agent considerably increased research efforts in inorganic biochemistry to identify more metallic drugs having the potential of treating lung cancer. Metal coordination centres, which exhibit a wide range of coordination numbers and geometries, various oxidised and reduced states and the inherent ligand properties offer pharmaceutical chemists a plethora of drug structures. Owing to the presence of C=N and C=S bonds in a thiosemicarbazone Schiff base, N and S atoms in its hybrid orbital has lone pair of electrons, which can generate metal complexes with different stabilities with most metal elements under certain conditions. Such ligands and complexes play key roles in the treatment of anti-lung cancer. Research regarding metallic anti-lung cancer has advanced considerably, but there remain several challenges. In this review, we discuss the potential of thiosemicarbazone Schiff base complexes as anti-lung cancer drugs, their anti-cancer activities and the most likely action mechanisms involving the recent families of copper, nickel, platinum, ruthenium and other complexes.

KEYWORDS

**thiosemicarbazone, schiff base, metal complexes, anti-lung cancer, chemotherapy**

## Introduction

Lung cancer is a malignant tumour that develops from the bronchial mucosa or glands of the lung, which poses the greatest threat to human health and lives, exhibiting the fastest increase rate in terms of morbidity and mortality (Brody, 2014). In 2020, ~19.3 million new cancer cases and approximately 10 million cancer-related deaths were reported worldwide (Sung et al., 2021). Lung cancer remains the leading cause of cancer-related death, engendering an estimated 1.8 million deaths (18%) with an estimated 2.2 million new cases (11.4%) in 2020 (Sung et al., 2021). The dramatic increase in the cases of lung cancer and multidrug-resistant infections has necessitated the search for novel treatment options and strategies (Tsai et al., 2018). New small-molecule anti-cancer agents Exhibit great potentials. However, the frequent occurrence of multidrug resistance (MDR) in lung cancer warrants the development of specific agents that can overcome MDR (Barrera-Rodriguez and Fuentes, 2015). Metal-based drugs are structurally stable and have unique three-dimensional configurations, which can be effectively used to treat multidrug-resistant infections (Rostan et al., 2021).




SCHEME 1
Thiosemicarbazone ligands for clinical application.

Thiosemicarbazone Schiff bases have garnered a lot of interest due to their multifunctional metal-chelating properties, inherent biological activities and structural flexibility (Matesanz et al., 2021a). It was established that thiosemicarbazides were metal chelators before the discovery of their anti-tumour activity. In addition to the presence of the C=N and C=S bonds in their own structure, which is favourable for metal coordination, it possesses a flexible thiourea structure that can introduce different substituents or functional groups (Scarim and Chin, 2022a). Especially for various heterocyclic ligands, introducing more heteroatoms enriches thiosemicarbazone's coordination mode and enhances its coordination ability (Stefani et al., 2013). They utilize the N–N–S donor system of ligands to form stable complexes with transition metals. Many studies reported that these complexes exhibited higher anti-tumour activity than ligands *in vitro* and *in vivo* and were extensively employed in the field of anti-tumour research (Summers, 2019a). Metal complexes are biologically and chemically diverse, unlike their corresponding ligands. This diversity is not only reflected in the metal and its oxidation state but also in the diversity of ligands that form coordination bonds with it and the different coordination modes (Matesanz et al., 2021a). Many literatures report that thiosemicarbazone ligands and their metal complexes have significant antitumor activity against lung cancer, liver cancer, colon cancer, breast cancer, neuroma and other tumors (Onodera et al., 2007; Lovejoy et al., 2012; Maqbool et al., 2020; Dharmasivam et al., 2022). Among many cancers, lung cancer has been the most common disease affecting human health. Thiosemicarbazone ligands have been reported to exhibit significant antitumor activity against lung tumor xenografts and have no significant toxicological effects, both intravenously and orally routes (Lovejoy et al., 2012). Therefore, it is necessary to review the research progress of thiosemicarbazone metal complexes in the treatment of lung cancer. We hope that this review guides researchers to better understand the application of metal-based agents in anti-lung cancer treatment and provide new ideas for the design and research of anti-tumour agents.

## Thiosemicarbazone ligands in a clinical trial

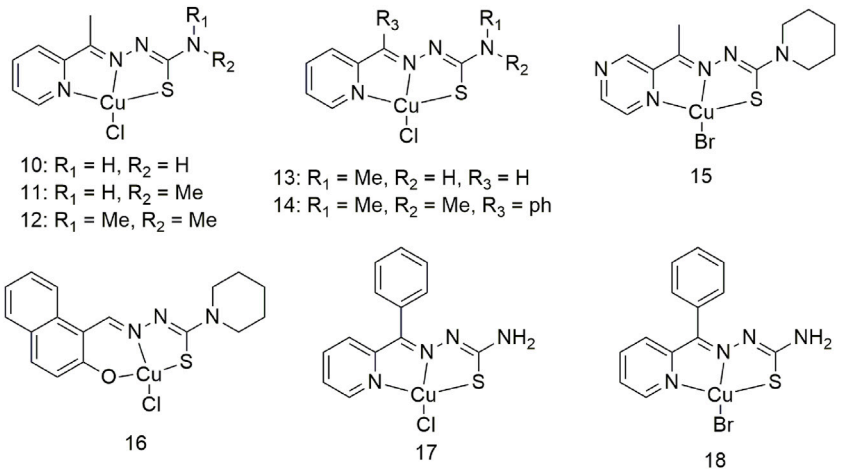
The thiosemicarbazone compounds used in clinical research include acetamidobenzaldehyde semicarbazide (**1**), N-methylisatin thiosemicarbazone (**2**), 5-hydroxy-2-methylcarbamate acylpyridine thiosemicarbazide (**3**), 3-aminopyridine-2-carbaldehyde thiosemicarbazide (**4**), COTI-2 (**5**), bis-2-pyridyl ketone 4-cyclohexyl-4-methyl-3-thiosemicarbazide (**6**) (Scheme 1) (Summers, 2019a; Matesanz et al., 2021a; Scarim and Chin, 2022a). Triapine is the most prevalent anti-cancer agent and has been included in over 20 clinical trials for treating multiple cancers, including pancreatic cancer, non-small cell lung cancer (NSCLC), leukaemia and myeloproliferative diseases (Scarim and Chin, 2022a). However, some tumour types respond poorly to Triapine, developing serious side effects, such as

hyperhemoglobinemia (Yu et al., 2006; Yu et al., 2009). Among this class of compounds, the 2-acetylpyridine thiosemicarbazone series (**7**) demonstrated significant anti-cancer activity (Onodera et al., 2007). Initial studies have shown that thiosemicarbazone can inhibit ribonucleotide reductase activity and further impede DNA synthesis, finally exerting its anti-cancer effect (Kowol et al., 2009; Yu et al., 2011). Further studies have shown that the anti-tumour activity of thiosemicarbazone is due to the joint action of multiple mechanisms (de Siqueira et al., 2019a). Thiosemicarbazone ligands have been widely reported to inhibit cellular iron uptake and affect cellular iron metabolism, and up-regulation of the downstream regulatory gene 1 of the metastasis suppressor protein N-myc (Serda et al., 2014; Malarz et al., 2018). Moreover, compound **9** causes DNA damage and inhibition of selective cellular topoisomerase IIα (Rao et al., 2009). This kind of a ligand is a good metal chelator, and the complexes formed by metal coordination exhibit redox activity and can catalyse hydrogen peroxide formation *in vivo* to generate reactive oxygen species (ROS) (Mckenzie-Nickson et al., 2016; Matesanz et al., 2021a). Through extensive structure-activity relationship studies, a new series of 2-benzoylpyridine thiosemicarbazone (**8**) incorporated 2-benzoylpyridine moieties in the structural backbone of previous arylhydrazone (Stefani et al., 2013). These compounds exhibited potent anti-lung cancer and anti-metastatic activities (Wu et al., 2006; Mahale et al., 2015). A series of di-2-pyridyl ketone thiosemicarbazone (**9**) were developed (Lovejoy et al., 2012) to improve the efficacy and safety of these potential anti-tumour drugs. This new compound exhibits markedly selective activity *in vivo* against human lung cancer (DMS-53) xenografts *via* the intravenous and oral routes (Lovejoy et al., 2012). Importantly, these analogues did not cause cardiotoxicity in tumour-bearing nude mice at high doses (Wangpu et al., 2016). To overcome MDR, di-2-pyridyl ketone thiosemicarbazone analogues target lysosomes by 'hijacking' the MDR pump p-glycoprotein (Pgp), which has potent anti-tumour and anti-metastatic activities both *in vitro* and *in vivo* (Wangpu et al., 2016). However, high doses of di-2-pyridyl ketone thiosemicarbazone may cause cardiac fibrosis, which warrants the design and synthesis of new thiosemicarbazone compounds. In second-generation thiosemicarbazone compounds wherein the terminal H of N4 was substituted with an alkyl group, compound 6 showed potential application prospects in antitumor (Lovejoy et al., 2012). Compound **6** exhibits higher anti-tumour activity *in vivo* and demonstrates considerable improvement in tolerability when administered orally or intravenously compared with first-generation thiosemicarbazone compounds (Stariat et al., 2013; Maqbool et al., 2020; Dharmasivam et al., 2022). Thiosemicarbazones combine with intracellular copper (Cu) ions to produce stable Cu complexes once they are inside the lysosome. These complexes generate ROS through the redox cycle, causing the lysosomal membrane to collapse (LMP), ultimately inducing apoptosis (Zhao et al., 2017; Gu et al., 2019).

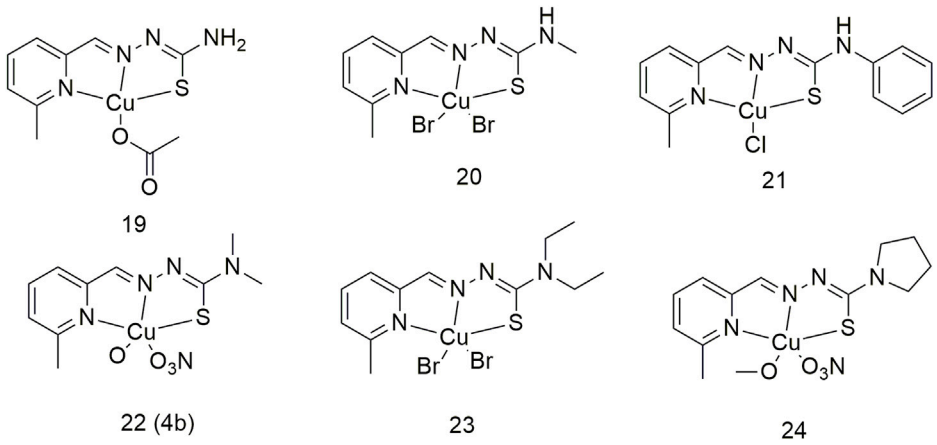
## Thiosemicarbazone copper complexes

Three Cu (II) complexes (**10–12**, Scheme 2) were synthesised and characterised to show that thiosemicarbazone Cu (II) complexes demonstrate strong anti-tumour activity (Qi et al., 2020). The anti-proliferative activity of complex 12 on the human lung cancer cell line A549 was the highest (0.20 ± 0.04 mM) of all the complexes. The lipophilicity of thiosemicarbazone ligands is closely related to the anti-proliferative activity of Cu (II) complexes on lung cancer cells Studies regarding the cellular mechanism of such Cu (II) complexes have shown that they promote apoptosis by catalysing hydrogen peroxide to generate intracellular ROS. Researchers proposed to develop two Cu (II) complexes based on the His242 residue of the IIA subdomain of the human serum albumin (HSA) carrier (**13** and **14**, Scheme 2) (Qi et al., 2016) to improve the delivery efficiency, anti-cancer activity and selectivity of anti-cancer metal preparations *in vivo*. Complex 13 binds to HSA subdomain IIA by hydrophobic interaction, while complex **14** binds to HSA by coordinating with His242. The cytotoxicity of complex 14 toward the A549 cell line (0.15 ± 0.01 μM) was significantly higher than that of cisplatin (17.36 ± 0.25 μM), and it significantly inhibited the growth of A549 tumours in nude mice. The anti-tumour activity against human large cell lung cancer cells (NCI-H460) of complexes **15** and **16** increased more than 40-fold compared with the ligands and exhibited significant pro-apoptotic activity at nanomolar concentrations (Scheme 2). Complexes **15** and **16** mediate significant antitumor activity by generating reactive oxygen species (ROS) through the redox cycle. (Scheme 2). Excess ROS can lead to the dissipation of mitochondrial membrane potential and promote the release of mitochondrial apoptotic factors. Complexes **17** and **18** also show effective oxidative cleavage of supercoiled DNA in the presence of hydrogen peroxide and exhibit good anti-tumour activity against the NCI-H460 cell line with $IC_{50}$ values in the range of 0.08–1.98 μM, which are higher than cis-Platinum (Pt) is 83 times lower (Scheme 2) (Liu et al., 2016).

By assessing the structure–activity relationship of six Cu (II) complexes (**19–24**, Scheme 3), researchers confirmed the multi-target ability of complex **22** on proteins and DNA (Wang et al., 2018). These six Cu complexes showed higher cytotoxicity against lung cancer cells (A549 and NCI-H460) than the ligands. The toxicity of complexes **19–24** toward the A549 cell line (≤13.69 ± 0.97 μM) was significantly higher than that of cisplatin (17.36 ± 0.75 μM). Importantly, in the A549 cell line, the cytotoxicity of the Cu (II) complex was enhanced by 1.0–3.0-fold upon binding to HSA. These six complexes inhibit cancer cell growth mainly by targeting DNA and apoptosis-related proteins in tumours. Forming a complex with HSA can improve the delivery efficiency of Cu complexes and impart a stronger ability to inhibit tumour growth.



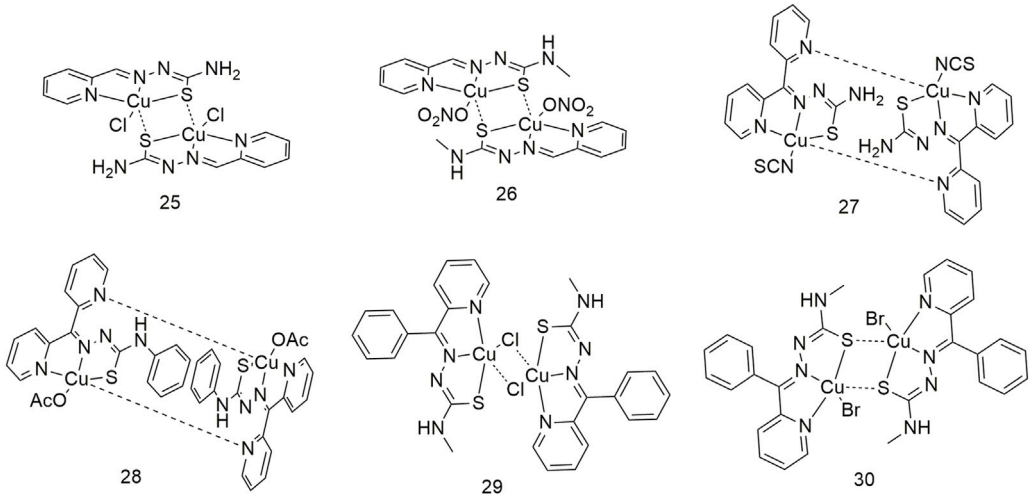

SCHEME 2
Structures of the copper (II) complexes (**10–18**) of thiosemicarbazone.



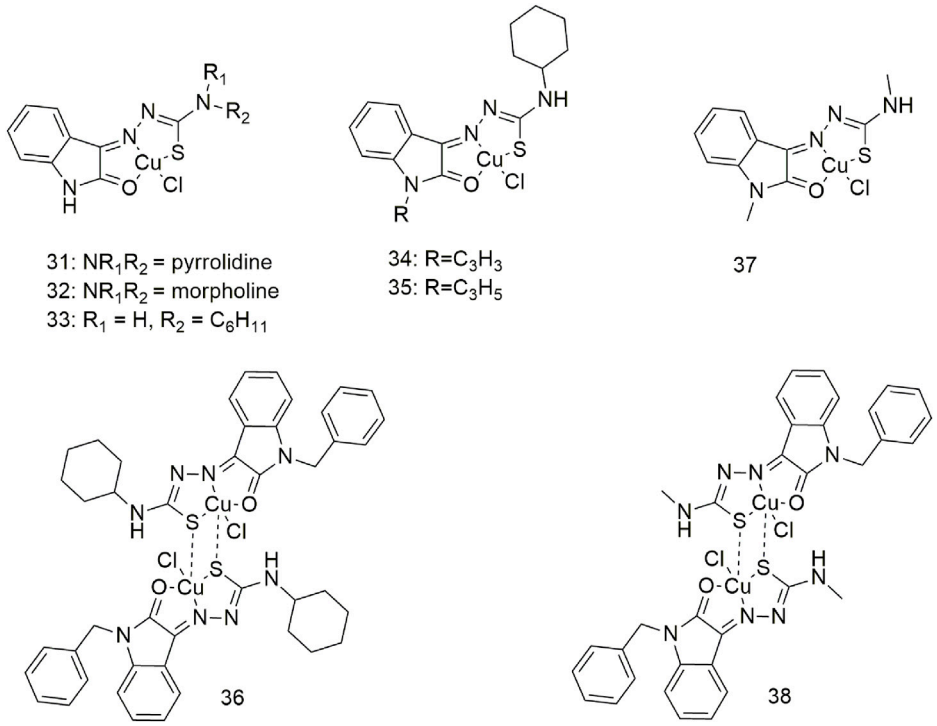

SCHEME 3
Structures of the copper (II) complexes (**19–24**) of thiosemicarbazone.

It is essential to conduct research regarding Cu complexes to produce a new generation of metallic anti-cancer drugs. Therefore, four dinuclear Cu (II) complexes (**25–28**, Scheme 4) were synthesised using Schiff base thiosemicarbazides (Qi et al., 2015). The anti-proliferative activity of these four Cu complexes on lung cancer cells (A549 and NCI-H460) was stronger than that of the ligands. Among them, the cytotoxic activity of complex **28** was the highest, with $IC_{50}$ values of 0.507 ± 0.021 μM and 0.235 ± 0.010 μM for the A549 and NCI-H460 cell lines, respectively. Binuclear structures and their cytotoxicity are also shown in complexes **29** and **30** (Scheme 4); $IC_{50}$ values are at the nanomolar level and 83 times lower than cisplatin. Their cytotoxicity is significantly more enhanced than that of the corresponding ligands (Liu et al., 2016).

Three complexes (**31–33**, Scheme 5) with broad-spectrum anti-tumour activity are formed when Cu (II) coordinates with 5-methoxyisatin thiosemicarbazone ligands with different N-terminal substituents; the $IC_{50}$ value of complex **31** for the A549 cell line is 17.88 ± 0.16 μM (Aneesrahman et al., 2019). Complexes **34–36** act as an intercalating agent toward DNA and as a binding agent toward bovine serum albumin (BSA), (Scheme 5) (Mathiyan et al., 2016). The *in vitro* cytotoxicity study of complex **36** showed good anti-proliferative activity against the A549 cell line (Mathiyan

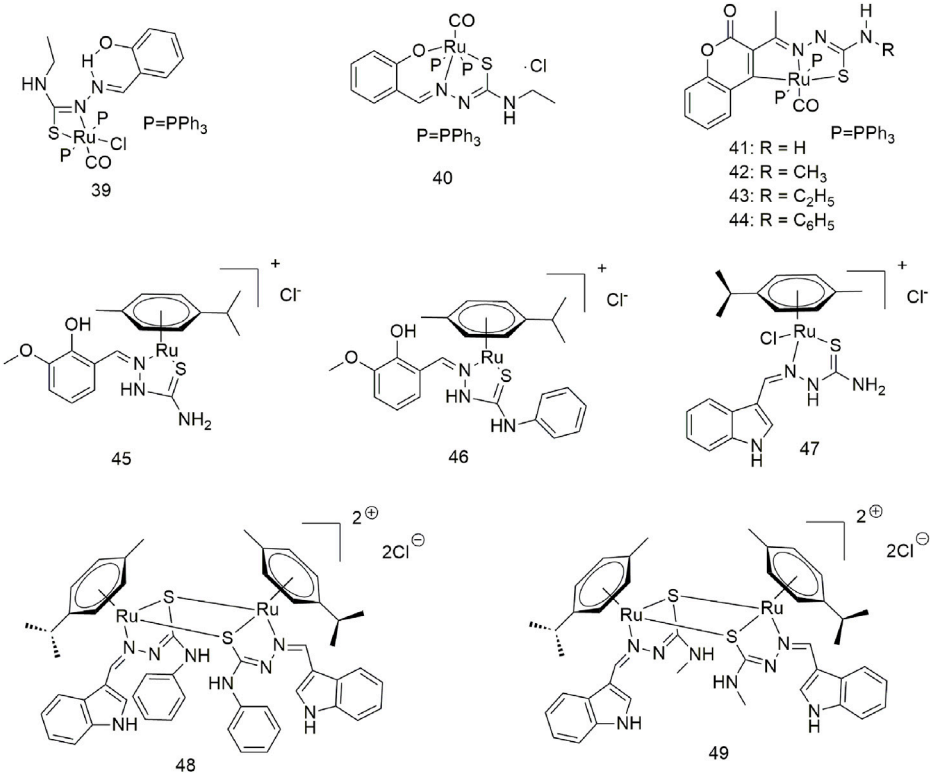



SCHEME 4
Structures of copper (II) complexes (**25–30**) of thiosemicarbazone.




SCHEME 5
Structures of the copper (II) complexes (**31–38**) of thiosemicarbazone.

et al., 2016). Molecular docking studies have shown that complexes **37** and **38** formed hydrogen bonds and hydrophobic interactions with the tyrosinase kinase receptor (Scheme 5) (Haribabu et al., 2021). These complexes can generate morphological changes in the A549 cell line based on fluorescence microscopy and show



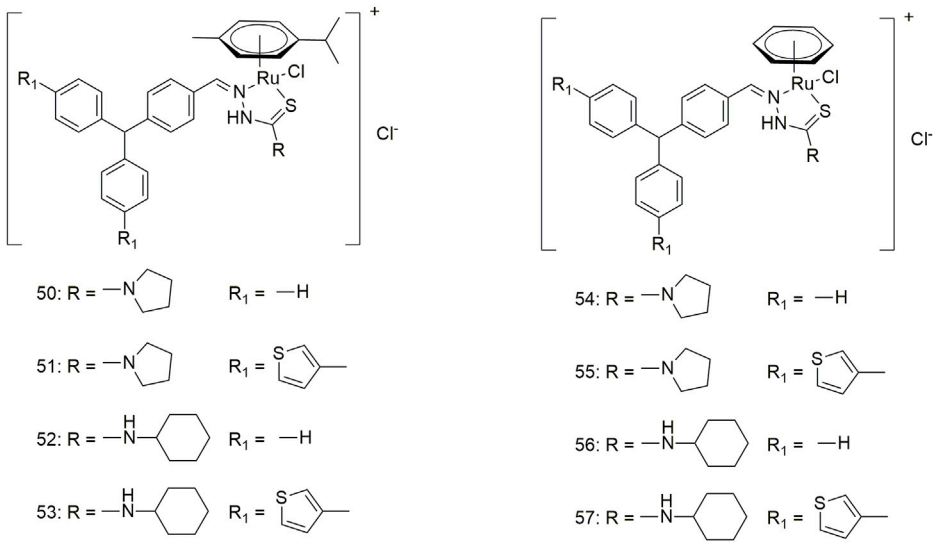

SCHEME 6
Structures of ruthenium complexes (**39–49**) of thiosemicarbazone.

cytotoxic activity against the A549 cell line with $IC_{50}$ values of 59.50 and 29.51 μM, respectively.

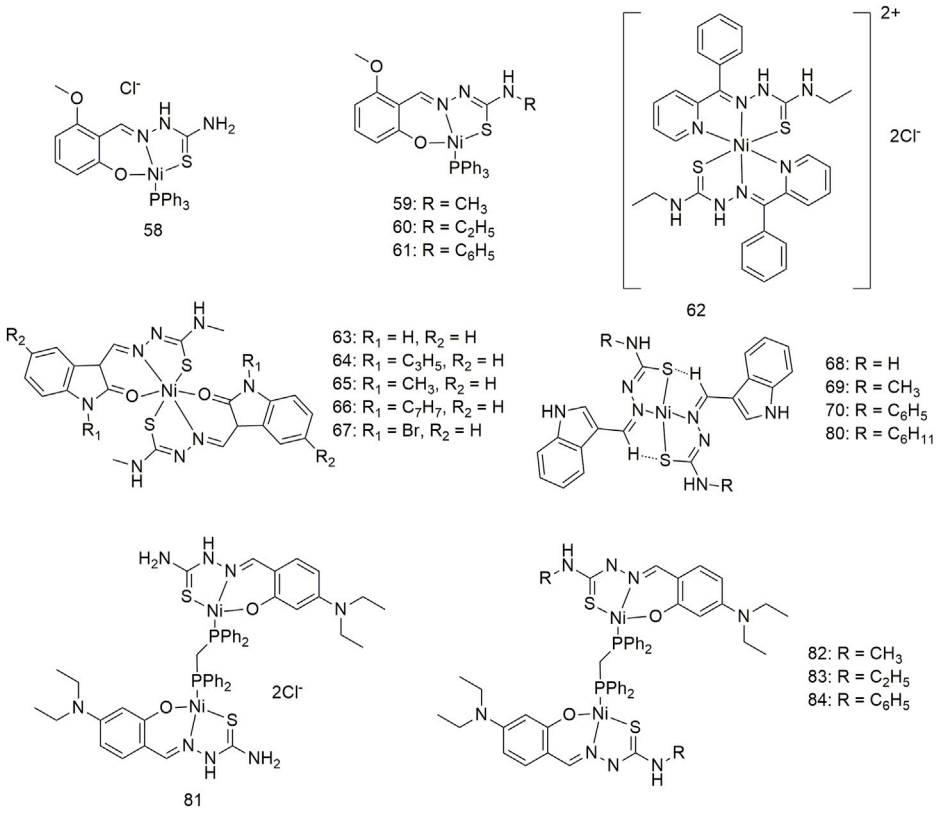
## Thiosemicarbazone ruthenium complexes

Salicylaldehyde-4-ethylthiosemicarbazide was reacted with [RuHCl(CO) $(PPh_3)_3$] to produce complexes **39** and **40** (Scheme 6) (Kalaivani et al., 2014). Complexes **39** (20 ± 1.10 μM) and **40** (17 ± 0.93 μM) showed higher $IC_{50}$ values for the A549 cell line compared with standard cisplatin (25 ± 1.11 μM). Complexes **39** and **40** are absorbed intracellularly and bind to the protein lysozyme, releasing CO and generating cytotoxicity. Four new divalent organoRu complexes (**41–44**, Scheme 6) were synthesised by modifying different groups at the N4 position of 3-acetylcoumarin-4-substituted thiosemicarbazide (Kalaiarasi et al., 2018). These complexes are more active than cisplatin and nontoxic to the human normal keratinocyte cell line HaCaT. The activity of complex **43** was optimal due to the N-terminal modification of thiourea with a more electron-donating ethyl group. Anna Gatti et al. reported the structures of two semi-sandwich aromatic Ru (II) thiosemicarbazide complexes (**45** and **46**, Scheme 6) with remarkable anti-tumour proliferative activity (Gatti et al., 2018). It forms mononuclear (**47**) and dinuclear complexes (**48** and **49**) with $[RuCl_2(p\text{-cymene})]_2$ in different reactions using indole thiosemicarbazide as a ligand, which is water soluble (Scheme 6) (Haribabu et al., 2018; Haribabu et al., 2021). These complexes exhibit significant cytotoxic activity against the A549 cell line *via* apoptosis, but not normal cells (L929). Most likely due to its higher DNA/protein binding affinity, binuclear Ru ($\eta^6$-p-cymene) complex shows better activity than Mono-Ru ($\eta^6$-p-cymene) complex. Complex 49 ($IC_{50}$ = 11.5 μM) was significantly more active on the A549 cell line than homogeneous cisplatin ($IC_{50}$ = 21.3 μM).

Eight triarylamine-thiosemicarbazides Ru (II)-arene semi-sandwich complexes (**50–57**, Scheme 7) have been reported to bind DNA/HSA, generating significant cytotoxic activity (Muralisankar et al., 2019). The anti-tumour activity of these Ru (II) complexes on the A549 cell line was significantly different. The $IC_{50}$ values of complexes **50**, **52** and **57** were greater than 100 μM, while complex **54** showed optimal anti-tumour activity (7.24 ± 5.4 μM). Furthermore, the treatment of the A549 cell line with complex **54** (10 μM) for 24 h produced significant changes in cell morphology, nuclear condensation and




SCHEME 7
Structures of ruthenium complexes (**50–57**) of thiosemicarbazone.




SCHEME 8
Structures of nickel (II) complexes (**58–84**) of thiosemicarbazone.

cell shrinkage, suggesting that cell death occurs *via* apoptosis under the action of this complex.

## Thiosemicarbazone nickel (II) complexes

Complexes **58–61** bind to calf thymus (CT)-DNA in an intercalated manner and interact with BSA in a statically quenched manner (Scheme 8) (Kalaivani et al., 2014a). Complexes **58–61** show significant anti-proliferative activity ($IC_{50}$, 27–30 μM) in the case of the A549 cell line under certain experimental conditions. The anti-tumour mechanisms of these complexes may be ROS-super-generation and lipid peroxidation, resulting in decreased mitochondrial membrane potential, caspase-3 activation and DNA fragmentation. Therefore, complexes **58–61** can cause apoptosis in the A549 cell line in a mitochondria-mediated manner and inhibit the migration and metastasis of lung cancer cells. Additionally, complex **62** interacts with CT-DNA and does not require any external force to cleave the DNA (Scheme 8) (Mathiyan et al., 2016). Complex 62 can bind to BSA and change the secondary structure of the protein. Complex **62** exhibits significant activity against the A549 cell line with an $IC_{50}$ value of 80.10 μM and is less toxic to the L929 cell line. Complexes **63–67** can interact with CT-DNA and BSA, cleave DNA without external factors and change the secondary structure of proteins (Scheme 8) (Haribabu et al., 2015). These five complexes have significant activity ($IC_{50}$ < 30 μM) against the A549 cell line, with complex 64 showing an $IC_{50}$ value of less than 0.1 μM. Complexes **68–80** show high antioxidant activity and antihemolytic activity against 2-2-diphenyl-1-picrylhydrazyl (DPPH) free radicals (Haribabu et al., 2017). The four complexes shows excellent anti-tumour activity (Scheme 8) ($IC_{50}$, 45.1–57.2 μM) against lung cancer (A549) and low toxicity ($IC_{50}$ > 600 μM) against mouse embryonic fibroblasts (L929). Intercalation interactions of four binuclear Ni (II) complexes (**81–84**) with DNA were confirmed by ethylene bromide shift studies and DNA viscosity measurements (Scheme 8) (Kalaiarasi et al., 2020). The interaction mechanism of complexes **81–84** with BSA is static. The four binuclear Ni (II) complexes exhibited significant cytotoxicity against the A549 cell line with lower $IC_{50}$ values (4.97–6.44 μM) than the standard metal drug cisplatin ($IC_{50}$, 31.08 ± 0.79 μM).

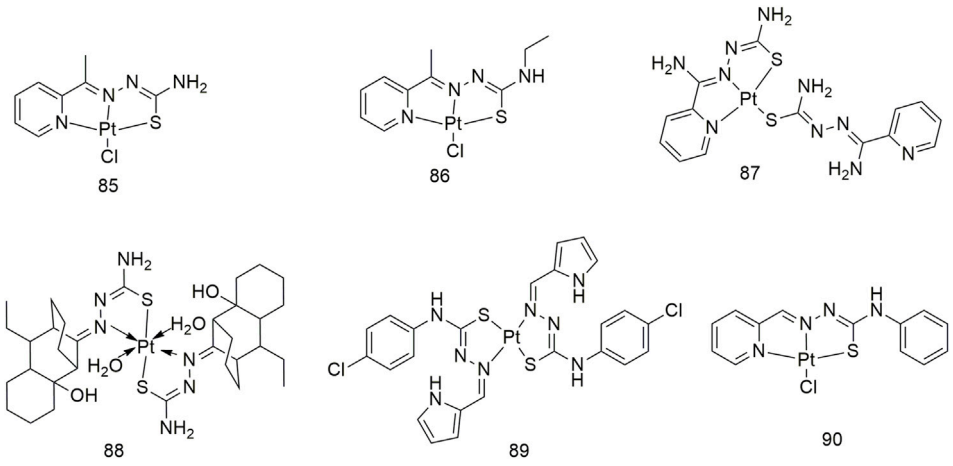
## Thiosemicarbazone platinum (II), gold (I) and silver (I) complexes

Cisplatin has become a widely used anti-cancer drug in clinics. Current research shows the action mechanism of involves the hydrolysis of cisplatin in the human body, removal of two chlorine atoms and binding to two purines on the genomic DNA and twisting the DNA double helix structure, thereby inhibiting DNA replication and transcription and ultimately causing cancer cell apoptosis (Pahontu et al., 2016a). The quenching mechanism of HSA by three thiosemicarbazide Pt (II) complexes (**85–87**) may be a static binding mode and impact the microenvironment of tryptophan residues in HSA (Scheme 9) (Lin et al., 2017). These three complexes show significant anti-proliferative activity ($IC_{50}$, 2.8–9.6 μM) against the NCI-H460 cell line. Among them, the $IC_{50}$ value of complex **87** toward the NCI-H460 cell line was 2.8 ± 1.1 μM, which was significantly higher than that of cisplatin ($IC_{50}$, 5.2 ± 2.2 μM). After 48 h of exposure, complex **88** reduced the proliferation rate of NCI-H1573 lung adenocarcinoma cells by 16.46% (Scheme 9) (Pahontu et al., 2016a). Complex **89** is a Pt (II) complex with α-n heterocyclic thiosemicarbazide as ligand, with an $IC_{50}$ value of 79 ± 4 μM for the A549 cell line after 72 h of incubation (Scheme 9) (Matesanz et al., 2018). The $IC_{50}$ values of complex **90** against the A549 cell line were 146.6, 101.7 and 69.28, respectively, after 24, 48 and 72 h of treatment (Scheme 9) (Salehi et al., 2022).

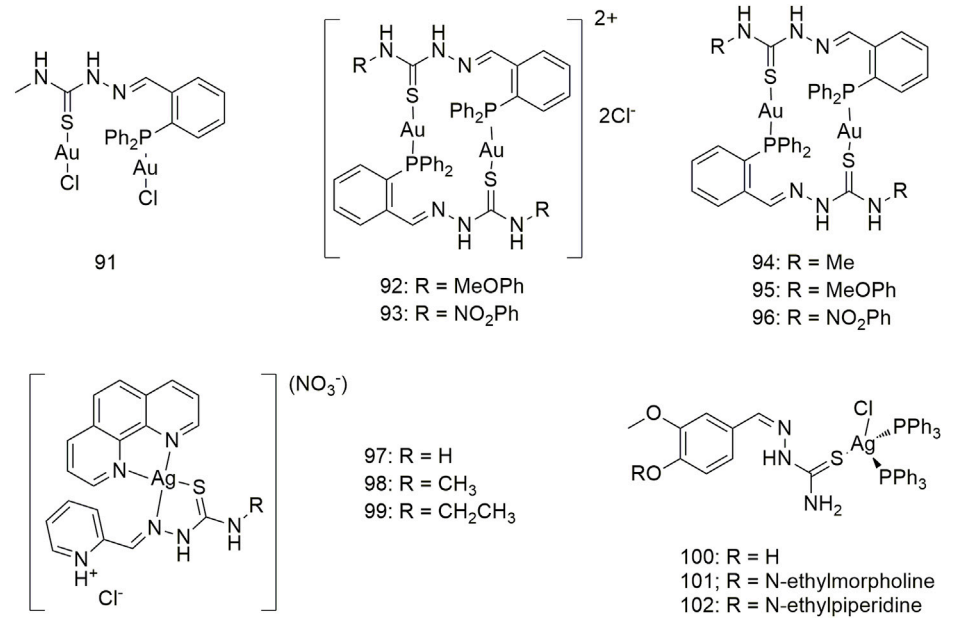
Six phosphothiosemicarbazide gold (Au) (I) binuclear complexes (**91–96**, Scheme 10) were prepared using synthetic methods. In terms of *in vitro* cytotoxic activity, the cytotoxic of these Au (I) complexes against the NCI-H460 cell line and MRC5 (normal human lung fibroblasts) was investigated and their $IC_{50}$ values were compared with that of cisplatin (Lessa et al., 2011). Complexes **91** and **94** strongly inhibited thioredoxin reductase activity. Compounds **97–99** not only exhibited good *in vitro* anti-proliferative activity against the A549 cell line with $IC_{50}$ values of 1.49–2.64 μM but also showed less cytotoxicity toward human breast non-tumour cells (MCF-10A) (Silva et al., 2020). The complexes **100–102** showed obvious cytotoxic activity against the A549 cell line, and the $IC_{50}$ values of 48 h incubation were 7.48 ± 0.21, 8.15 ± 1.21 and 6.46 ± 0.51 μM, respectively, which were higher than the cell activity of cisplatin ($IC_{50}$, 23.36 ± 0.42 μM) (Scheme 10).

## Other classes of thiosemicarbazone metal complexes

Complexes **103** and **104** are two novel pyrazolylthiourea Pd (II) complexes that bind to CT DNA and cleave supercoiled DNA (pUC19) (Scheme 11) (Haribabu et al., 2020). Complexes **103** and **104** show cytotoxic $IC_{50}$ values of 130.3 and 117.2 μM for the A549 cell line, respectively, *in vitro* and are less toxic to normal human lung (IMR90) cells ($IC_{50}$ > 133 μM). Three kinds of clamp-type Pd (II) complexes (**105–107**, Scheme 11) were synthesised by reacting $PdCl_2$ with thiourea ligands, which can bind to CT-DNA and BSA. Spectroscopic evidence shows an intercalation pattern between DNA and Pd (II) complexes.

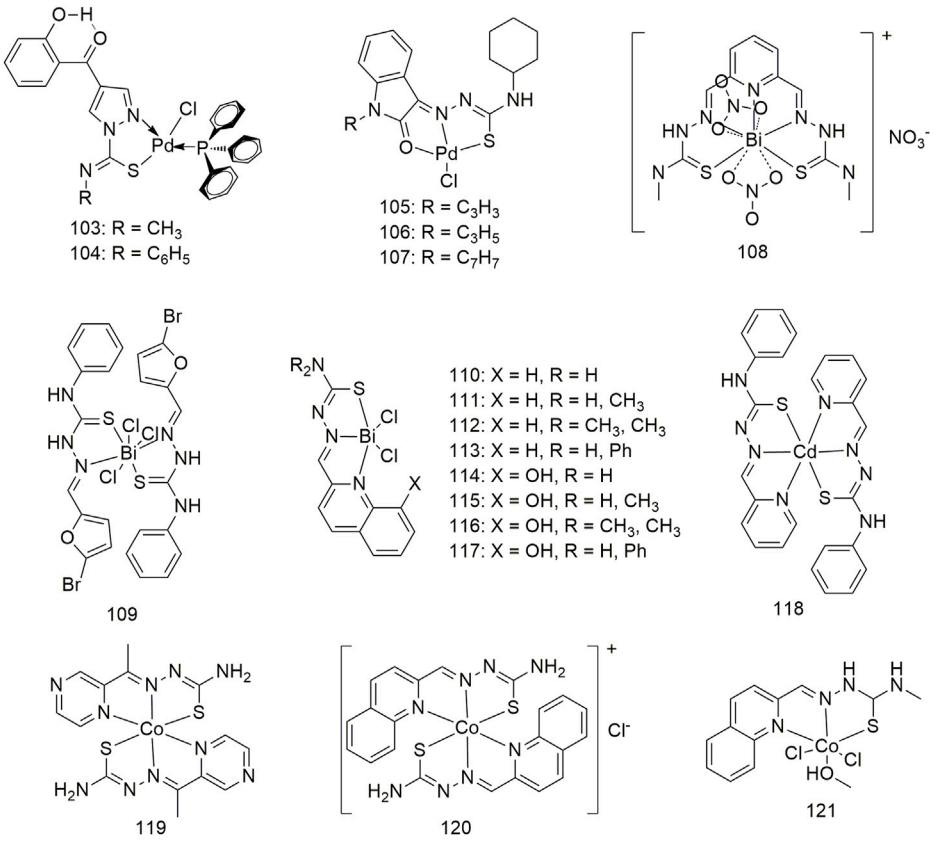



SCHEME 9
Structures of platinum (II) complexes (**85–90**) of thiosemicarbazone.




SCHEME 10
Structures of gold (I) (**90–96**) and silver (I) (**97–102**) complexes of thiosemicarbazone.

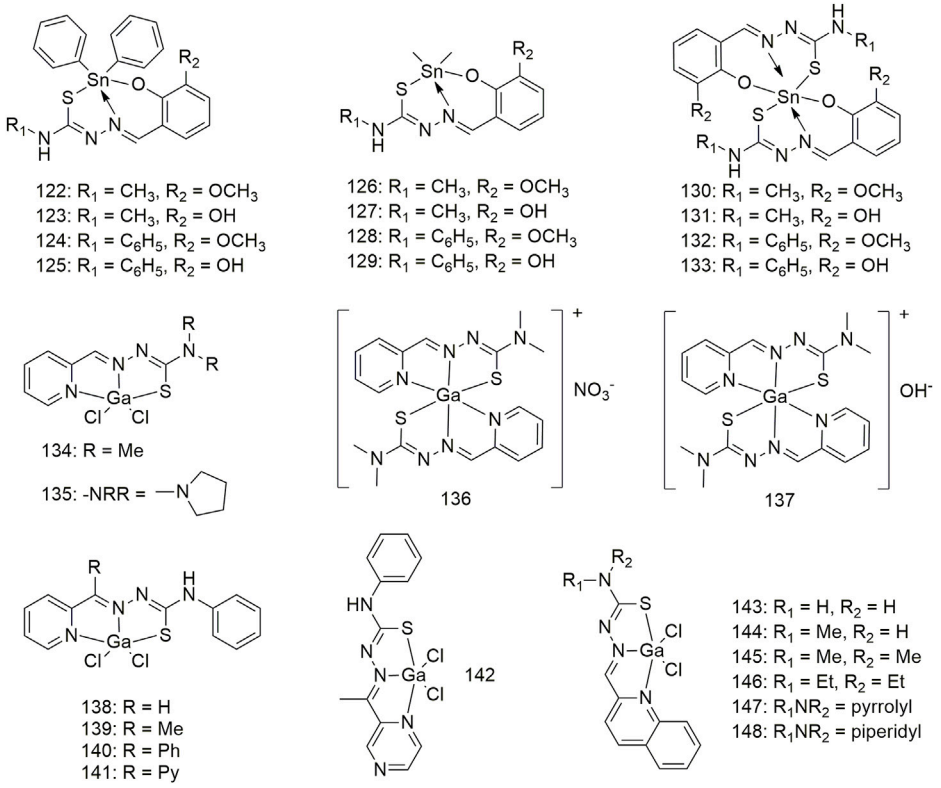
The binding mode is further confirmed by CD spectroscopy, suggesting that the binding occurs in a non-grooved mode. Changes in the protein secondary structure by the complex were confirmed by simultaneous fluorescence spectroscopy studies. Spectral evidence indicates that the complex shows good binding properties to proteins. Complex **105–107** cleaved pUC19 plasmid DNA without external reagents. These complexes exhibit significant *in vitro* cytotoxicity against the A549 cell line, suggesting that they can kill cancer cells even at low concentrations. The $IC_{50}$ of complex **107** toward the A549 cell line is 22.92 μM. Complex **108** significantly inhibits the colony formation and migration abilities of the A549 cell line and can induce apoptosis (Scheme 11) (Ouyang et al., 2017). The anti-cancer




SCHEME 11
Structures of palladium (II) (**103–107**), bismuth (III) (**108–117**), chromium (III) (**10–18**) and cobalt (II) (**119–121**)complexes of thiosemicarbazone.

activity of complex 108 is much higher than that of its parent ligand, and it generates no obvious toxicity toward non-cancerous human lung fibroblasts (HLF). *In vivo* experiments proved that complex **108** shows an obvious inhibitory effect on the growth of the A549-xenografted tumour in tumour-bearing mice and has no adverse effects on the mouse body weight and liver. Complex **109** effectively inhibits the proliferation and migration of the A549 and H460 cell lines and induces cell apoptosis (Scheme 11) (Ruizhuo et al., 2016). Complex **109** has $IC_{50}$ values of 16.41 ± 0.93 μM and 20.04 ± 1.28 μM for the A549 and H460 cell lines, respectively, and demonstrates low cytotoxicity against normal HLF ($IC_{50}$, 117.16 ± 5.96 μM). These results suggest that thiosemicarbazone complexes with bismuth (Bi) (III) may be an interesting and effective anti-lung cancer drug candidate. The coordination of Bi complexes **110–117** is a thiosemicarbazone containing a quinoline group (Scheme 11) (Scaccaglia et al., 2022). Among them, complexes **113** and **114** show significant inhibitory activities against the A549 cell line with $IC_{50}$ values of 5.05 ± 1.79 μM and 46.96 ± 16.66 μM, respectively. Complex **111** exhibits significant inhibitory activity against the A549 cell line ($IC_{50}$, 14.0 ± 1.1 μM) (Abyar et al., 2019). This complex exerts inhibitory activity on the A549 cell line in a cell cycle-dependent manner and induces caspase-mediated apoptosis and caspase-independent cell death. The anti-tumour activity of complex **119** ($IC_{50}$, 18.9 μM) on the A549 cell line was significantly higher than that of the ligand ($IC_{50}$, 265.3 μM) (Ming et al., 2015). Through groove binding, two cobalt (II) complexes (**120** and **121**, Scheme 11) bind to DNA, and the binding capacity of complex **121** is stronger than that of complex **120**. Complex **121** shows higher anti-tumour (A549) activity than complex **120**. Although the inhibitory activity of complex **121** on A549 cells was inferior to that of cisplatin, its inhibitory efficacy on A549/CDDP in drug-resistant cells was higher than that of cisplatin (Xiaorui et al., 2013).

Complexes **122–133** were derived from four thiosemicarbazone Schiff bases (Scheme 12). Except for complex **124**, which has no experimental data, and complex **127**, which is inert, other complexes demonstrate significant anti-lung cancer (H460) activity with $IC_{50}$ values less than 3.2 μM (Yusof et al., 2020). Two types of gallium (Ga) (III) complexes (**134–137**, Scheme 12) with ligand/Ga (III) ratios of

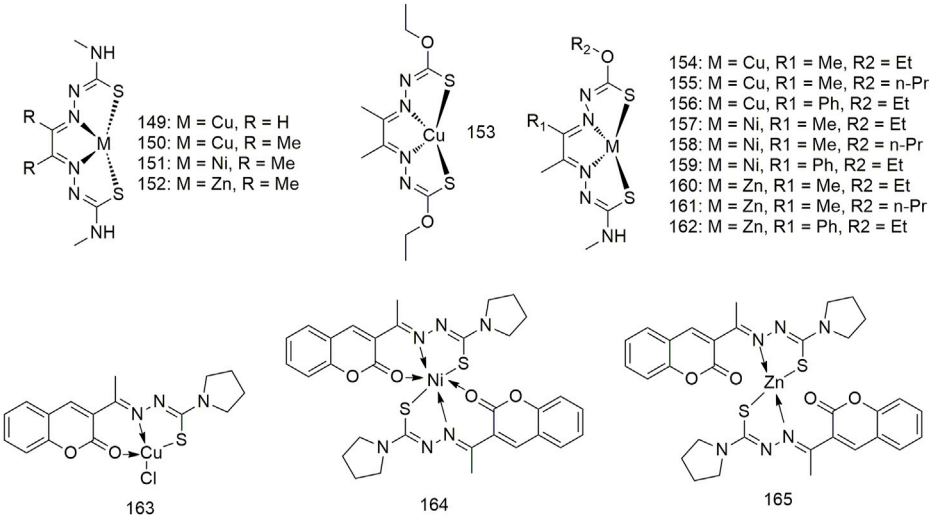



SCHEME 12
Structures of tin (II) (**122–133**) and gallium (III) (**134–148**) complexes of thiosemicarbazone.

2:1 and 1:1 exhibit significant anti-proliferative activity against the NCI-H460 cell line, and their $IC_{50}$ values are between 0.72 and 0.43 μM. The anti-proliferative activity of Ga (III) complexes with a metal/ligand ratio of 1:1 (complex **135**) was significantly higher than that of the 1:2 (complex **137**) (Qi et al., 2017). Complexes 135 and 137 significantly promote the release of cytochrome C from mitochondria and increase the activities of caspase-3 and -9 in the NCI-H460 cell line. Both types of Ga (III) complexes inhibited the G1/S transition better than ligands alone. Five 1:1 ligand/Ga (III) complexes (**138–142**, Scheme 12) exhibit significant anti-proliferative activity against lung cancer (A549) with $IC_{50}$ values between 0.37 and 2.32 μM. The structure–activity relationship results showed that modifying the lipophilic groups present on the ligands significantly improves its anti-proliferative activity, and this biological activity was further enhanced after coordination with Ga. The anti-cancer (A549) activities ($IC_{50}$, 0.46–1.41 μM) of the Ga (III) complexes **143–148** all exceeded those of the corresponding metal-free ligands (Cao et al., 2019). These Ga (III) complexes are non-toxic to normal hepatocytes and exhibit strong selectivity for tumour cells. The anti-tumour (A549) mechanism of Ga (III) complexes produces intracellular ROS, disrupts mitochondrial membrane potential and ultimately promotes apoptosis.

## Copper, nickel and zinc complexes of the same ligand

A series of hybrid ligands derived from 4-methyl-3-thiosemicarbazide and hydrazinecarbothioic acid O-alkyl ester ligands chelate with Cu, Ni and zinc (Zn) to form complexes **149–162** (Scheme 13) (Andres et al., 2020). Cu derivatives **154** and **156** exhibit strong anti-cancer selectivity, having $GI_{50}$ values of less than 100 nM against the A549 lung adenocarcinoma cells, and at least 20-fold lower $GI_{50}$ activity against IMR90 (>2.0 μM) non-malignant lung fibroblasts. While Ni complexes exhibit much lower anti-tumour activity (>5.3 μM) compared with Cu complexes, Zn complexes show weaker activity (0.055–7.6 μM). Zn and Ni complexes demonstrate little cancer selectivity. The complexes Cu (II) (**163**), Ni (II) (**164**) and Zn (II) (**165**) interacts with CT-DNA *via* intercalation binding as well as with BSA (Rahman et al., 2017). The activity of these complexes on the A549 cell line




SCHEME 13
Structures of copper (II), nickel (II) and zinc (II) complexes of thiosemicarbazone.

was lower than in the other cell lines, in which complex **165** exhibits moderate cytotoxicity against the A549 cell line with an $IC_{50}$ value of 200 μg/ml.

## Discussion

Recently, the increase in the number of patients with lung cancer and the emergence of tumour resistance, the demand for metal compounds that treat cancer continues to grow, not only because cancer is difficult to cure but also because metal-based compounds with remarkable antitumor activity. Metal-based compounds show high cytotoxicity *in vitro*. Furthermore, the substitution of ligands and modification of existing chemical structures have produced a variety of metal-based compounds, some of which show higher tumour cytotoxicity, low toxicity toward non-tumour cells and better pharmacokinetics.

Metal complexes, such as Cu, Zn, Au, silver (Ag), Pt and Ru, exhibit higher anti-tumour activity than metal-free ligands. Cancer cells have a greater demand for Cu than normal cells, and Cu metabolism is strongly linked to angiogenesis and metastasis (Santini et al., 2014; Shobha et al., 2018; Kannappan et al., 2021). Therefore, since the 1970s, various ligands that form complexes with Cu, such as thiosemicarbazides, imidazoles and phosphines, have been proposed as potential anti-cancer drugs. Furthermore, Cu salts and complexes have been shown to inhibit tumour cell proliferation (Santini et al., 2014). In fact, these Cu complexes allow Cu to undergo redox cycling between the reduced monovalent and oxidised divalent states under the action of cellular oxidants and reductants (Balsa et al., 2021). Cu complexes show redox activity, engender intracellular ROS overload and promote apoptosis through the mitochondrial apoptotic pathway. Many ruthenium (Ru) complexes have been synthesised and evaluated as possible cancer therapeutics over the past two decades (Gatti et al., 2018). The activity of Ru (III) compounds depends on *in vivo* reduction to more reactive Ru (II) substances, which has led to increased interest in organometallic Ru (II) arene complexes in octahedral configurations, where aromatics stabilise Ru in Ru oxidation states (Haribabu et al., 2018). Some semi-sandwiched Ru (II) arene complexes exhibit notable anti-cancer activity *in vitro* and *in vivo* (Gatti et al., 2018). The activity of Ru complex cells is significantly higher than that of cisplatin and the former shows tumour selectivity. This may be due to the good DNA/protein binding affinity of the Ru complex. Ni (II) complexes exhibit obvious inhibitory activity on the A549 cell line and less toxicity on the L929 cell line. Ni (II) complexes interact with CT-DNA and BSA, cleave DNA on the absence of any external factors and change the secondary structure of protein. Nickel can form rich molecular geometries, ensuring the formation of complexes with improved properties (Bisceglie et al., 2019). This facilitates improved drug properties without any increase in cellular resistance or negative side effects from the drugs. Thiosemicarbazone Pt (II), Au (I) and Ag (I) complexes were significantly more active on lung cancer cells than cisplatin. Owing to their similar structural characteristics and coordination chemistry with Pt, palladium (Pd) complexes have been thoroughly investigated for anti-cancer activity similar to that of their Pt analogues. These metal complexes have complicated and diverse action mechanisms and anti-tumour activities, which compensate for the deficiencies in the existing drugs. However, these studies are only at the basic research stage. Overall, a

rational design of anti-tumour drugs based on other metal classes is yet to be achieved, and there is a lack of unique metal complexes. Although thiosemicarbazone-based metal complexes have been used in the laboratory and have exhibited good outcomes on lung cancer cells and tumour-bearing nude mice, achieving a clear distinction between therapeutic and toxic doses is a major challenge.

## Author contributions

X-GB, YZ, and JQ contributed to conceptualizing, writing the original draft manuscript, and reviewing and editing the manuscript.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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