Review

Mercury Toxicity and Detection Using Chromo-Fluorogenic Chemosensors

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Abstract: Mercury (Hg), this non-essential heavy metal released from both industrial and natural sources entered into living bodies, and cause grievous detrimental effects to the human health and ecosystem. The monitoring of Hg^{2+} excessive accumulation can be beneficial to fight against the risk associated with mercury toxicity to living systems. Therefore, there is an emergent need of novel and facile analytical approaches for the monitoring of mercury levels in various environmental, industrial, and biological samples. The chromo-fluorogenic chemosensors possess the attractive analytical parameters of low-cost, enhanced detection ability with high sensitivity, simplicity, rapid on-site monitoring ability, etc. This review was narrated to summarize the mercuric ion selective chromo-fluorogenic chemosensors reported in the year 2020. The design of sensors, mechanisms, fluorophores used, analytical performance, etc. are summarized and discussed.

Keywords: fluorescent sensors; colorimetric sensors; mercuric ions; mercury toxicity

1. Introduction

Metals like Na, K, Mg, Ca, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, and Mo are well-known to play important roles in human physiological functions. However, the excessive as well as deficiency of these essential metals in human body can cause serious detrimental effects. Similarly, several non-essential metals entered into the human body from different sources can also cause grievous toxic effects even at trace quantity. Therefore, there is an exponential growth in the development of facile and cost-effective analytical techniques for the on-site and real-time detection of both essential and non-essential metal ions [1–5]. Among the various analytical techniques, the chromogenic and fluorogenic chemosensors are extensively developed for the detection of metal ions because of their high selectivity and sensitivity, easy-to-design, low-cost, simplicity, real-time, and on-site detection ability. The chromo-fluorogenic chemosensors are designed and developed by considering three important things: (i) signaling unit, (ii) recognition unit, and (iii) mechanism (Figure 1). The signaling unit may be an organic fluorophore, chromophore, or optically active nanoparticles. When the recognition unit selectively recognize the target analyte, the mechanism based on electron/charge/energy transfer occurred in the sensor can alter the electronic properties of the signaling unit that gives detectable optical response [6].

Mercury (Hg), one of the non-essential heavy metal can cause serious toxicity to human health and ecosystem. Because of the high affinity to S-containing ligands, the accumulation of mercury in human body can affect the normal functioning of proteins and enzymes leading to the wide variety of diseases related to kidney, brain, reproductive disturbance, central nervous system, etc. [7]. Considering the toxicity, the acceptable limit of inorganic mercury in drinking water was prescribed as 2 µg L^{-1} (10 nM) by the United States Environmental Protection Agency (US EPA). Also, the inorganic mercury can be converted into organic mercury (like methylmercury) that affects the brain and cause...
other neurotoxic effects, and therefore, the intake of 1.6 µg/kg body weight per week of methylmercury was recommended by Joint FAO/WHO Expert Committee on Food Additives (JECFA) [8]. The associated toxicity even at a trace amount of mercury resulted an expedite growth in the design of novel analytical methods, including optically active chemosensors for the detection of mercuric ions. Literature survey supported the reports of several reviews on mercuric ions sensing and toxicity [9–15]. In this review, the mercury toxicity and important chelates available for mercury intoxication will be discussed first, and then the chemosensors reported in the year 2020 will be summarized. The fluorophores used for the designing of sensors, the sensing mechanisms and the detection performance will be summarized and discussed.

Figure 1. Schematic representation showing the design of a chemosensor.

2. Mercury Toxicity and Intoxication

Mercury, a silvery colored metal, liquid at room temperature, is characterized by atomic number 80 in group 12 of the periodic table of elements, standard atomic weight 200.59 g/mol. Mercury can assume the three oxidation states 0, +1, +2. It presents a high density 13.53 g/mL, and a relatively high vapor pressure (0.0017 torr at 25 °C, corresponding to a concentration of 20 mg/m³). It is monoatomic in vapor phase, and is highly soluble in polar and non-polar solvents (a mercury water solution can reach the concentration of 0.6 µg/L at 25 °C).

The use of mercury is reported since the ancient times, mainly as the pigment cinnabar. The mined amount of mercury has been almost constant over the centuries until 1500, when huge amounts were produced in Spain (Almaden) to be shipped to Spanish South America for silver extraction. A paper by Hylander and Meili [16], takes into account the trend in mercury production from this period to 2000. The discovery of gold in California in 1850 produced a jump in mercury production, as well as its use in chlor-alkali plants in the 20th century. The consumption of explosives in the war industry contributed to the large production of mercury during World Wars I and II. The increasing awareness of mercury toxicity has led in the years to its banning from different applications (amalgamation in China in 1985 and in Russia in 1990, pesticides in USA in 1993, batteries in USA in 1996), until the Protocol on Heavy Metals (cadmium, lead and mercury), signed in 1998 by different
countries, the 2005 EU Mercury Strategy, and finally the Minamata Convention on Mercury in 2013. At the fifth session of the Intergovernmental Negotiating Committee in Geneva, Switzerland, on 19 January 2013, it was agreed the Minamata Convention on Mercury, a global treaty to protect human health and the environment from the adverse effects of mercury. The major highlights of the Minamata Convention on Mercury include a ban on new mercury mines, the phase-out of existing ones, control measures on air emissions, and the international regulation of the informal sector for artisanal and small-scale gold mining [17]. Despite the above legislative controls, mercury remains one of the major toxicants in the world [18] and deserves a careful consideration about its environmental quantification, its toxic action, and the strategy for the clinical treatment of intoxication.

Mercury presents in the environment mainly in three chemical forms, i.e., elemental mercury (liquid or vapor Hg\(^0\)), inorganic mercuric compounds (Hg\(^{2+}\)), and organic mercury compounds (methylmercury, MeHg, CH\(_3\)Hg and ethyl mercury EtHg, C\(_2\)H\(_5\)Hg) [19]. Toxicity of mercury in humans can be related to any of these three forms, absorbed in different ways: inhalation, oral, and dermal. The kind and the degree of intoxication is highly specific for any of these three chemical species, as well as the symptoms and the consequences [20]. Table 1 presents the main sources of exposure of the different forms of mercury and the affected organs [21].

| Species | Occupational Exposure | Environmental Exposure | Routes of Exposure | Affected Organs |
|---------|-----------------------|------------------------|-------------------|-----------------|
| Elemental mercury, Hg | Chlor-alkali plants, gold extraction, incineration of wastes, coal burning, dental amalgam handling | Dental amalgam | Inhalation | Nervous system |
| Organic mercury, CH\(_3\)Hg\(^+\) | - | Food (Fish and seafood) | Ingestion | Nervous system |
| Inorganic mercury, Hg\(^{2+}\) | - | Medicinal uses, dermatological creams | Ingestion, transdermal | Kidneys |

The exposure of mercury by human body can be occurred via ingestion or inhalation [7]. The extensive utilization of elemental mercury in a number of industrial processes has led the involved personnel exposed to gaseous mercury. To have a quantitative evaluation of this exposure, we remember that in presence of liquid Hg\(^0\) the surrounding non-ventilated air can reach a concentration of 20 mg/m\(^3\) of monoatomic mercury vapors. Since a person inhales 15–20 m\(^3\) of air daily, a worker who stays about 8 h in a mercury saturated place inhales 5–6.6 m\(^3\) of air, i.e., 100–135 mg of Hg\(^0\). Mercury vapors are efficiently absorbed by lungs due to their high liposolubility producing severe pulmonary injuries. Elemental mercury in the lungs enters the blood flow, where a certain amount is oxidized to Hg\(^{2+}\) and excreted in urine, and some, due its high liposolubility, passes through the blood–brain barrier (BBB) and enters in the central nervous system.

Various forms of inorganic mercury in water are converted by microorganisms to methyl mercury that accumulates in fish and pass to humans through the food chain. In humans, inhaled or ingested methyl mercury is well absorbed and is found in red blood cells, liver, kidneys, and above all in the brain (including the fetal brain, since methyl mercury can cross the placental barrier), where it causes severe, cumulative, and irreversible injuries to the central nervous system. Its retention time in the human body varies from months to years, and the appearance of symptoms can be delayed for many years. Symptoms of methylmercury intoxication include mental retardation, cerebral blindness, deafness, palsy, and dysarthria, particularly in children exposed in utero. It is important to emphasize that methylmercury exposure mainly affects people whose diet includes the consumption of high amounts of fish.
Inorganic mercury compounds were largely used in the chemical industry, and were the cause of heavy occupational exposure. Inorganic salts are poorly absorbed, and kidneys represent their main target. From a chemical point of view, mercury toxicity depends primarily from the mercuric ion ability to form covalent bonds with sulfur atoms, substituting hydrogen atoms in sulphydryl groups of proteins to form mercaptides. This can deactivate a number of essential enzymes, completely altering their regular biological functions [21–23].

Chelation therapy is used for the treatment of all forms of mercury intoxication. In clinical use, chelating agents remove metal ions from the biological ligands in the organism, where they exert their toxic action, through the formation of metal complexes that are successively excreted. Characteristics of a good chelator should be great solubility in both water and lipids, resistance to biotransformation, capacity to reach the sites of metal accumulation, high stability of the complexes at the pH of body fluids, and toxicity of the formed complexes lower than that of the free metal ions [21]. Most of the chelating agents in use today are not able to cross the BBB and this limits their ability to remove the toxic metal ions from the brain. The main chelating agents used in the treatment of mercury intoxication are summarized in Figure 2.

![Figure 2. Molecular structure of the chelating agents used for mercury intoxication.](image)

The dithiol chelating agent 2,3-dimercaptopropan-1-ol (BAL) was originally synthesized for the treatment of the effects of the war gas Lewisite. It competes successfully with protein SH groups forming stable chelates with Hg2+ metal ions. For several decades after its synthesis, it was recommended for the treatment of inorganic mercury poisoning, but it presents severe adverse effects, including painful intramuscular injections, high blood pressure and tachycardia, and predisposition to redistribute the complexed toxic elements to the brain. At any rate, it is contraindicated in the treatment of alkyl-Hg intoxication. In most cases, it has been replaced by DMSA and DMPS in the treatment of metal poisoning [24].

The chelating agent meso-dimercaptosuccinic acid (DMSA) or simply called succimer is the water-soluble dithiol. DMSA can be administered as intravenous and oral preparations, being DMSA a hydrophilic chelator. When administered orally, about 20% is absorbed in the gut, and about 95% of the absorbed amount is bound to plasma albumin, presumably by one SH group to a cysteine residue, being the second SH group free for binding metal ions. The action of DMSA is limited to the extracellular space. It increases Hg excretion in the urine. DMSA is considered the drug of choice for the treatment of organic-Hg intoxication. Even if DMSA does not pass the BBB, it indirectly reduces the brain burden of methyl mercury presumably by changing the brain/blood equilibrium. The derivative monoisoamyl 2,3-dimercaptosuccinic acid (MiADMSA) is at the moment under evaluation. Differently from DMSA, which removes extracellularly distributed metal ions [25], MiADMSA is also able to chelate intracellularly distributed metal ions [26].
The structure of 2,3-dimercaptopropane-1-sulfonic acid (DMPS), also known as unitiol, is shown in Figure 2. DMPS is a drug produced in Germany and registered for the treatment of mercury intoxication. It is not an approved drug in the USA, unless the FDA gives a special permission. The daily dose is usually 3–10 mg DMPS/kg body weight. DMPS is believed the optimal remedy in poisoning by inorganic mercury [27], while it is less effective than DMSA for organic mercury [28]. DMPS can be administered both intravenously and orally; less than 40% of orally administered DMPS is effectively adsorbed [29]. DMPS, which is a hydrophilic chelating agent, is mainly distributed in the extracellular space, but a small fraction is found in the intracellular compartments [30]. DMPS scavenges mercury from kidneys more effectively than DMSA, and is considered the drug of choice for the treatment of acute intoxication by inorganic mercury [30,31].

The α-lipoic acid ((R)-5-(1,2-dithiolan-3-yl)pentanoic acid, LA) is the sulfur containing organic compound known as thioctic acid, presented in Figure 2. LA, essential for aerobic metabolism, is ordinarily produced in the body. Its reduced form, called dihydrolipoic acid (DHLA), contains a couple of -SH groups; it is characterized by high affinity for mercuric ion and has been recently proposed as an effective mercury chelator [31].

3. Chromo-Fluorogenic Chemosensors

Because of the potential toxicity of mercuric ions to living systems, there is an expedite growth in the design of optically activity chemosensors. In the year 2020, more than 100 Hg^{2+} selective chromo-fluorogenic chemosensors were reported, which can be classified in three different categories based on the optical responses, i.e., (i) fluorescence, (ii) colorimetric, and (iii) dual-mode chemosensors. The fluorescence chemosensors are discussed based on the fluorescence changes, i.e., turn-off, turn-on and ratiometric (Table 2). The fluorescent chemosensors are more sensitive than the colorimetric sensors with high visual effects that allowed for different bioimaging and diagnosis applications. The most dominating mechanisms for Hg^{2+} sensing are complexation-induced change in the optical properties due to electron/energy/charge transfer or the sensor possess a reactive group that undergoes Hg^{2+}-catalyzed chemical transformation. The sensing mechanisms and other experimental parameters (such as solvent medium, pH and concentration of sensors etc.) important in fabricating a suitable chemosensors are discussed.

| Sensors | Medium | λ_{exc}/λ_{em} (nm) | LOD   | Applications                                | Ref.          |
|---------|--------|--------------------|-------|--------------------------------------------|---------------|
| 1       | H_{2}O | 350/464            | 0.16 µM | Real water sample analysis [32]            |               |
| 2       | H_{2}O | 500/632            | 39.2 nM | -                                          | [33]          |
| 3       | H_{2}O | 316/416            | 0.1243 µM | Real water sample analysis [34]            |               |
| 4       | THF:H_{2}O (1:49, v/v) | 380/500 | 22.8 ppb | -                                          | [35]          |
| 5       | CH_{3}CN:H_{2}O (1:1, v/v) | 270/380 | 0.68 µM | Real water analysis and pH sensing [36]   |               |
| 6       | EtOH   | 410/485            | 172 nM | Living cell imaging [37]                  |               |
| 7       | CH_{3}CN:HEPES buffer (2.8, v/v) | 369/490 | 1 nM | Live-cell imaging [38]                    |               |
| 8       | H_{2}O | 490/574            | 312 nM | Real water sample analysis [39]            |               |
| 9       | Tris-HCl buffer | 358/445 | 4.40 µM | Real water sample analysis and live cell imaging [40] |               |
| 10      | HEPES buffer | 330/550 | 7.59 nM | Real water samples and live cell imaging [41] |               |
| 11      | CH_{3}CN:H_{2}O (6:4, v/v) | 375/485 | 1.26 nM | Test paper strip and real water analysis [42] |               |
| 12      | CH_{3}CN | 420/603 | 20 ppb | Removal from water [43]                    |               |
| 13      | DMF    | 325/463            | 17 nM | Air and real water samples [44]            |               |
| 14      | MeOH   | 280/436            | 0.01 µM | -                                          | [45]          |
| 15      | H_{2}O | 495/521            | 0.34 µM | Industrial effluents and paper strip [46]  |               |
| 16      | H_{2}O | 430/512            | 5.02 nM | Real water analysis [47]                  |               |
| Sensors | Medium | λ<sub>exc</sub>/λ<sub>em</sub> (nm) | LOD | Applications | Ref. |
|---------|--------|-----------------|-----|--------------|-----|
| 17      | MeOH:H<sub>2</sub>O (1/4, v/v) | 340/455 | 3.12 nM | Live cell imaging | [48] |
| 18      | CH<sub>3</sub>CN | 440/492 | 200 nM | Real water sample analysis | [49] |
| 19      | DMF:H<sub>2</sub>O (1:1, v/v) | 337/378 | 16 nM | - | [50] |
| 20      | H<sub>2</sub>O:DMSO (95:5, v/v) | 360/540 | 0.51 µM | - | [51] |
| 21      | H<sub>2</sub>O | 285/364, 464 | 7.41 nM | Bioimaging | [52] |
| 22      | DMSO:HEPES buffer (9:1, v/v) | 355/455 | 8.12 nM | Live cell imaging | [53] |
| 23      | H<sub>2</sub>O | 359/495 | 22 nM | Real water analysis and bioimaging | [54] |
| 24      | MeOH:H<sub>2</sub>O (4:1, v/v) | 305/387 | 0.28 ppb | Real water analysis | [55] |
| 25      | DMSO:HEPES buffer (9:1, v/v) | 315/495 | 0.645 µM | Real water analysis and live cell imaging | [56] |
| 26      | Acetonitrile:HEPES buffer (1:1, v/v) | 401/520 | 1.01 µM | Real water analysis | [57] |
| 27      | Acetonitrile:HEPES buffer (1:1, v/v) | 405/525 | 1.98 µM | - | [57] |
| 28      | THF:H<sub>2</sub>O (9:1, v/v) | 480/675 | 13.1 nM | Live cell imaging | [58] |
| 29      | H<sub>2</sub>O:DMSO (99.6: 0.4, v/v) | 495/543 | 19.2 nM | Test color strips and bio-imaging | [59] |
| 30      | CH<sub>3</sub>CN | 520/587 | 3.9 ppb | Drinking water, live cells and plant tissues | [60] |
| 31      | CH<sub>3</sub>CN:HEPES (1:9, v/v) | 470/530 | 27 nM | Live cell imaging | [61] |
| 32      | MeOH:H<sub>2</sub>O (4:1, v/v) | 360/590 | 4.8 µM | Test paper strips, bioimaging | [62] |
| 33      | DMSO:H<sub>2</sub>O mixture | 360/453 | 406 nM | Adsorption of H<sub>2</sub>S | [63] |
| 34      | DMSO:H<sub>2</sub>O (2:8, v/v) | -/600 | 30 nM | Real food samples and live cell imaging | [64] |
| 35      | HEPES buffer (pH 7.4) | 480/518 | 3 nM | Real water and biological analysis | [65] |
| 36      | EtOH:H<sub>2</sub>O mixture | 366/491 | 750 nM | Real water sample analysis | [66] |
| 37      | CH<sub>3</sub>CN:H<sub>2</sub>O (0.2:99.8, v/v) | 366/463 | 2.1 nM | - | [67] |
| 38      | DMF:buffer (8:2, v/v) | 315/529 | 2.40 µM | Live cell imaging | [68] |
| 39      | DMF:buffer (8:2, v/v) | 470/621 | 2.86 µM | Live cell imaging | [68] |
| 40      | DMF:buffer (8:2, v/v) | 470/614 | 0.22 µM | Live cell imaging | [68] |
| 41      | H<sub>2</sub>O | 480/532 | 8.619 nM | Real water analysis and live cell imaging | [69] |
| 42      | PBS buffer | 460/515 | 0.4 nM | Real water analysis and live cell Imaging | [70] |
| 43      | CH<sub>3</sub>CN:HEPES buffer (1.9, v/v) | 545/580 | - | - | [71] |
| 44      | DMSO | 520/606 | 0.13 µM | INHIBIT logic gate | [72] |
| 45      | CH<sub>3</sub>CN:HEPES buffer(2.8, v/v) | 580/691 | 1.5 nM | Living cells imaging | [73] |
| 46      | EtOH:HEPES buffer (1:1, v/v) | 590/664 | 1.87 ppb | Real water sample analysis | [74] |
| 47      | H<sub>2</sub>O | 335/574 | 3.55×10<sup>13</sup> mL<sup>-1</sup> | - | [75] |
| 48      | DMSO:H<sub>2</sub>O (7/3, v/v) | 390/504,613 | 0.2 µM | Living cells | [76] |
| 49      | CH<sub>3</sub>CN | 510/573 | 28.5 nM | - | [77] |
| 50      | DMSO:H<sub>2</sub>O (6:4, v/v) | 505/585 | 16.9 nM | - | [78] |
| 51      | CH<sub>3</sub>CN | 520/585 | 16 nM | Real water sample analysis | [79] |
| 52      | DMSO:H<sub>2</sub>O (7/3, v/v) | 500/562 | 26 nM | Real water sample analysis | [80] |
| 53      | CH<sub>3</sub>CN:H<sub>2</sub>O (7/3, v/v) | 520/604 | 0.38 µM | - | [81] |
| 54      | DMF:Tris-HCl buffer (1:1, v/v) | 562/557 | 1.61 nM | Bio-sensing and live cell imaging | [82] |
| 55      | H<sub>2</sub>O:THF (4:1, v/v) | 565/590 | 16 nM | Live cell imaging | [83] |
| 56      | DMSO:H<sub>2</sub>O (7/3, v/v) | 480/582 | 13.4 nM | Test color strips and live cell imaging | [84] |
| 57      | DMSO:H<sub>2</sub>O (7/3, v/v) | 480/578 | 15.6 nM | Test color strips and live cell imaging | [85] |
3.1. Fluorescent Chemosensors

3.1.1. Fluorescent Turn-Off Chemosensors

The heavy metal ions like Hg$^{2+}$ greatly influence the fluorescence of a sensor after complexation leading to the fluorescence quenching via energy or electron transfer mechanism. Ebru et al. [32] have reported the pyrazoline based fluorogenic sensor 1 for the detection of Hg$^{2+}$ in aqueous medium. Sensor 1 (Figure 3) showed a fluorescence maxima at 464 nm ($\lambda_{\text{exc}} = 350$ nm), but the fluorescence intensity was decreased upon addition of Hg$^{2+}$ with the sensitivity limit of 0.16 $\mu$M (Figure 4). The electrostatic interaction between 1 and the heavy metal ion Hg$^{2+}$ caused the fluorescence quenching. This reversibility sensor 1 formed a complex with Hg$_2$$^{2+}$ in 2:1 stoichiometry. The fluorescent turn-off sensor 2 was
introduced for the detection of Hg\(^{2+}\) in water medium [33]. Sensor 2 (Figure 3) showed an absorption band at 525 nm while fluorescence maxima at 632 nm. Prominent fluorescence quenching accompanied by 25 nm red-shift was observed upon binding with Hg\(^{2+}\) leading to the solution color turned from pink to pale violet due to the intramolecular charge transfer occurred between Hg\(^{2+}\) and the N-atoms of 2. Sensor 2 formed complex with Hg\(^{2+}\) in 1:2 stoichiometry, and the estimated LOD was reported to be 39.2 nM.

![Molecular structures of the sensors 1 to 4.](image)

The Hg\(^{2+}\) selective fluorescent turn-off sensor 3 (Figure 3) using the Zn-based metal organic framework (Zn-MOF) was prepared by reacting the ligand 5-aminoisophthalic acid with Zn\(^{2+}\) [34]. The Zn-MOF formed a 3D supramolecular network having uncoordinated carboxylic atoms and pores size of 8.2 Å. Addition of Hg\(^{2+}\), the fluorescence of Zn-MOF at 416 nm (\(\lambda_{exc} = 316 \text{ nm}\)) was quenched with a sensitivity limit of 0.1243 \(\mu\text{M}\) due to the complexation-induced inhibition of intermolecular energy transfer. In another work, the triarylamine-based covalent organic framework (COF) polymer 4 (Figure 3) was converted into nanosphere via Suzuki polymerization under mini-emulsion condition, which showed
selective fluorescence turn-off response for Hg$^{2+}$ in mixed aqueous medium. The blue-green fluorescence of 4 is quenched upon complexation of Hg$^{2+}$ with the sulfur atom. The sensor was immobilized successfully over macroporous sponge for facile detection and removal of Hg$^{2+}$ [35].

Figure 4. (a) Fluorescence spectra of 1 ($K_1 = 1$) and (b) photographs of 1 ($B = 1$) under UV in the absence and presence of 20 µM metal ions in water. Reproduced with permission from [32]. Copyright 2020 Elsevier.

The benzimidazole derived fluorescent sensor 5 (Figure 5) showed an excellent selectivity towards Hg$^{2+}$ in CH$_3$CN/H$_2$O (1:1, v/v). Sensor 5 exhibited a fluorescence emission at 380 nm when excited at 270 nm. In the fluorescence experiments, only Hg$^{2+}$ caused significant fluorescence quenching (85%) of 5 by forming a complex in 1:1 stoichiometry (Figure 6). Sensor 5 showed a LOD of 0.68 µM, and no interference with other tested metal ions. The complex [5-Hg$^{2+}$] emits in an acidic environment whereas quenched in an alkaline environment, which can also be used for pH sensing [36]. The triazole-bridged coumarin conjugated quinoline sensor 6 (Figure 5) was developed for the fluorescent turn-off detection of Hg$^{2+}$. The complexation of 6 with Hg$^{2+}$ at the tridentate coordination site created by the quinolone and triazole stimulates the unusual PET process, and caused the fluorescence quenching at 485 nm with the sensitivity limit of 172 nM. The Hg$^{2+}$ sensing ability of 6 was further studied in live U-2-OS cells [37]. Murugan et al. [38] have reported
the tetraazamacrocyclic derivative appended with the salicylaldehyde 7 (Figure 5) for the selective fluorescent turn-off sensing of Hg\(^{2+}\) in CH\(_3\)CN/HEPES buffer (2:8, v/v). The fluorescence emission of 7 at 490 nm was quenched upon complexation with Hg\(^{2+}\) in 1:1 ratio due to chelation enhancement quenching (CHEQ) effect. The quenched fluorescence is recovered with the addition of KI. The lowest limit of detection for Hg\(^{2+}\) is 1 nM. In addition, the sensor 7 showed fluorescence turn-on response for the detection of HSO\(_4^-\).

**Figure 5.** Molecular structures of the sensors 5 to 8.

The marine cyanobacterium based natural protein C-phycoerythrin 8 (Figure 5) was applied for the fluorescent turn-off detection of Hg\(^{2+}\). Sensor 8 showed an intense yellow orange fluorescence at 574 nm due to the phycoerythrobilin (PEB), a linear tetrapyrrole. Upon interaction of Hg\(^{2+}\) with the amino acid side chain and thioether bridges in the protein 8, the fluorescence is quenched. The complexation of Hg\(^{2+}\) caused indirect charge transfer that quenched the fluorescence of 8. The LOD of 8 for Hg\(^{2+}\) was estimated as 312 nM [39].

The acridine-based chemosensor 9 (Figure 7) possessing two S-donor atoms was developed for the fluorescent turn-off detection of Hg\(^{2+}\) in Tris-HCl buffer. The fluorescence of 9 at 445 nm was quenched upon complexation with Hg\(^{2+}\) in 1:1 binding stoichiometry. Sensor 9 showed a LOD of 4.40 µM, and applied for the monitoring of Hg\(^{2+}\) in real water samples and bioimaging ability in living cells [40]. Adopting the complexation-induced fluorescence quenching approach, the dansyl-peptide based sensor 10 was developed by the conjugating two serines and dansyl groups. Sensor 10 (Figure 7) exhibited sensitivity towards Hg\(^{2+}\) through fluorescence quenching at 550 nm in HEPES buffer solutions. Upon complexation of sensor 10 with Hg\(^{2+}\) in 2:1 stoichiometry, the heavy atom effect and the electron transfer caused the fluorescence quenching. With nanomolar detection limit (7.59 nM), sensor 10 was successfully applied for monitoring Hg\(^{2+}\) ions in real water samples (lake and tap water) and living LNCaP cells [41].
Figure 6. (A) Coordination mode of 5 with Hg$^{2+}$, and (B) the UV absorption (a) and fluorescence (b) spectra of sensor 5 (10 µM) in CH$_3$CN/H$_2$O (1:1, v/v, pH = 7.4) upon the addition of different cations. Reproduced with permission from [36]. Copyright 2020 Elsevier.

Figure 7. Molecular structures of the sensors 9 to 13.
Thiocarbohydrazide based Schiff base 11 (Figure 7) was introduced for the colorimetric and fluorescent sensing of Hg$^{2+}$. Sensor 11 showed AIE behaviour in a mixture of acetonitrile and water. The emission intensity was found to increase gradually with the addition of water up to 40%, and the cyan fluorescence was clearly developed from aggregates. Upon interaction of Hg$^{2+}$ with the AIE active 11 led to the color change from colorless to yellow. The quenching in fluorescence intensity was attributed to combine effect of chelation enhanced fluorescence quenching (CHEQ) and photo-induced electron transfer (PET). The limit of detection for Hg$^{2+}$ is 1.26 nM. Sensor 11 was applied for the detection of Hg$^{2+}$ by using test paper strip and in various real water samples [42].

Yanxin et al. [43] reported a covalent organic frameworks (COFs) 12 (Figure 7) with extended hydrazone-linked π-conjugation by condensing two different monomers for the detection and removal of Hg$^{2+}$ in acetonitrile. Sensor 12 showed an absorption peak at 350 nm while emission band at 603 nm due to the ESPT. Addition of Hg$^{2+}$ ion, the color change from orange to light blue with the significant fluorescence quenching due to the inhibition of the ESPT process. Limit of detection is calculated as 20 ppb without any significant interference with other ions. Moreover, sensor 12 was applied for the effective Hg$^{2+}$ removal from water. The pyrene-based COFs 13 (Figure 7) was introduced for the simultaneous detection and removal of Hg$^{2+}$ in DMF. After interaction with Hg$^{2+}$, the fluorescence of COFs 13 at 463 nm was quenched with the sensitivity limit of 17 nM, and the blue-emitting 13 turned to colorless. Fluorescence quenching of 13 is attributed to a PET process from sensor to the Hg$^{2+}$. Sensor 50 was applied for removing Hg$^{2+}$ from both air and water [44]. The microporous porphyrinic zirconium-based MOF 14 was developed by using meso-tetra(4-carboxyphenyl)porphyrin as a ligand for the detection of Hg$^{2+}$ in methanol medium [45]. Sensor 14 fluorescence at 436 nm was quenched upon addition of Hg$^{2+}$ ions with a fast response rate under <1 min and sensitivity limit of 0.01 µM. The quenching efficiency was explain by donor-acceptor (D-A) electron transfer mechanism. Also, sensor was applied for the detection of DMF.

Reena et al. [46] have reported a phenylalaninol-fluorescein conjugated Schiff base receptor 15 (Figure 8) for the colorimetric and fluorescent detection of Hg$^{2+}$ in pure aqueous medium. Upon gradual addition of Hg$^{2+}$, the emission at 521 nm is quenched and slightly red-shift, while the absorption showed a hypsochromic shift of 30 nm at 430 nm causing color change from green to light pink. The lowest limit of detection for Hg$^{2+}$ is 0.34 µM. The job’s plot supported the 1:1 binding stoichiometry between 15 and Hg$^{2+}$. Sensor 15 is applicable for Hg$^{2+}$ detection in industrial effluents and paper strip visualization with the irreversible mode. The curcumin and β-cyclodextrin inclusion complex 16 (Figure 8) was applied for the chromo-fluorogenic sensing of Hg$^{2+}$ in aqueous medium. The supramolecular system 16 complexed with Hg$^{2+}$ after deprotonation of aliphatic hydroxy group caused apparent color change from yellow to colorless [47]. The absorbance of 16 at 482 nm was quenched and blue-shifted to 379 nm. Also, the fluorescence emission at 512 nm was significantly diminished and blue-shifted to 502 nm. With the fluorescence change, the concentration of Hg$^{2+}$ can be detected down to 5.02 µM, and applied to quantify Hg$^{2+}$ concentration in real water samples. In another work, the fluorescent turn-off sensor 17 was developed for the detection of Hg$^{2+}$ in MeOH/H$_2$O (1/4, v/v) solvent [48]. The emission at 455 nm is red-shifted to 485 nm and quenched with the addition of Hg$^{2+}$ due to the chelation enhancement quenching effect (CHEQ). Without responding with other ions, sensor 17 can be applied to detect Hg$^{2+}$ down to 3.12 nM. In addition, the absorption band of 17 at 292 nm was quenched whereas the band at 337 nm red-shifted to 355 nm. The absorbance and fluorescence changes occurred in 17 after the addition of Hg$^{2+}$ were recovered with the addition of iodide ions.

Ashwani et al. [49] reported an anthrapyridone-based receptor 18 (Figure 8) for the sensing of Hg$^{2+}$ and Cu$^{2+}$ in CH$_3$CN. The fluorescence emission at 492 nm of sensor 18 was quenched in presence of Hg$^{2+}$ with the LOD of 200 nM. In UV–vis absorption study, the absorption peak of 18 at 445 nm was quenched and a red-shifted absorption band appeared at 630 nm leading to the color change from yellow-green to green. Similar spectral changes
of 18 were also observed with Cu$^{2+}$. The quinoline-based benzimidazole derivative 19 (Figure 8) was developed for the dual-mode chemosensing of Hg$^{2+}$ in DMF/H$_2$O. Sensor 19 formed gel in DMF and converted to sol with the addition of Hg$^{2+}$, and also the fluorescence at 378 nm was quenched. This gel-sol transition based sensor showed the 16 nM LOD for Hg$^{2+}$. Sensor 19 also showed naked-eye detectable color change for Cu$^{2+}$ from white to dark pink in the gel state.

![Molecular structures of the sensors 15 to 22.](image)

The multi-analytes selective bis-thiosemicarbazone based receptor 20 (Figure 8) was developed for the detection of Hg$^{2+}$, Zn$^{2+}$ and Cd$^{2+}$ in H$_2$O:DMSO (95:5 v/v) [51]. Sensor 20 showed an intense emission at 540 nm, while addition of Hg$^{2+}$, the fluorescence was quenched and red-shifted to 578 nm with the color changed from yellow to reddish-brown ($\lambda_{ex} = 360$ nm). Color and fluorescence changes were attributed by intra-ligand fluorescence and influence of coordination of Hg$^{2+}$ to the receptor. The LOD for Hg$^{2+}$ was estimated as 0.51 $\mu$M. With Zn$^{2+}$ and Cd$^{2+}$, the fluorescence of 20 was blue-shifted and enhanced respectively at 488 and 470 nm. The tryptophan-based polymer 21 (Figure 8) was reported for the detection of Hg$^{2+}$ and Cu$^{2+}$ [52]. Sensor 21 showed dual emissions at 364 nm and 464 nm with the yellow colored fluorescence in aqueous medium at physiological pH. Upon complexation with Hg$^{2+}$/Cu$^{2+}$, sensor 21 showed significant quenching at 464 nm without any change at 364 nm. The quenching was attributed by PET process from the tryptophan donor to the pyridine acceptor unit. With 21, the concentration of Hg$^{2+}$ and Cu$^{2+}$ can be detected down to 7.41 nM and 4.94 nM, respective, and applied for the bioimaging of intracellular Hg$^{2+}$/Cu$^{2+}$ in live CP3 cells. The isocoumarin based sensor 22 (Figure 8) was
developed for the fluorescent turn-off sensing of Hg\(^{2+}\) and Fe\(^{3+}\) DMSO/HEPES buffer solution (9/1, v/v, pH 7.0) [53]. After complexation of Hg\(^{2+}\)/Fe\(^{3+}\) with 22 in 1:2 stoichiometry, significant fluorescence quenching was observed at 455 nm. Sensor 22 showed the sensitivity limit of 8.12 nM and 5.51 nm for Hg\(^{2+}\) and Fe\(^{3+}\), respectively. Sensor 22 was applied to imaging intracellular Hg\(^{2+}\)/Fe\(^{3+}\) in live HepG2 cells.

3.1.2. Fluorescent Turn-On and Ratiometric Chemosensors

The fluorescent turn-on and ratiometric sensors are more advantageous than fluorescent turn-off sensors for biological applications because of the facile measuring of low-concentration contrast in compared to a ‘dark’ background, reduction in the false positive signals and enhancement in the sensitivity. Therefore, more numbers of Hg\(^{2+}\) selective fluorescent turn-on and ratiometric sensors were reported and applied for the monitoring of Hg\(^{2+}\) in real samples and bioimaging intracellular Hg\(^{2+}\) in live cells. The micellar based Hg\(^{2+}\) selective fluorescent turn-on sensor 23 (Figure 9) was developed by organizing the fluorophore 10-methylacridinium perchlorate, sulfur-containing ligand N,N-bis(2-hydroxyethylthio-1-ethyl)dodecylamine and the surfactant sodium dodecyl sulfate (SDS) [54]. Under micellar condition, the ligand decorated with the SDS formed complex with Hg\(^{2+}\) that enhanced the fluorescence of 10-methylacridinium due to the inhibition of PET from the ligand to the excited fluorophore. The turn-on fluorescence is observed at \(\lambda_{em} = 495\) nm (\(\lambda_{exc} = 359\) nm) with the limit of detection of 22 nM Hg\(^{2+}\). The pyridyl-based sensor 24 (Figure 9) containing multiple binding sites was developed for fluorescent turn-on sensing of Hg\(^{2+}\) in aqueous solution with a limit of detection of 0.28 ppb [55]. The broad fluorescence emission spectrum of sensor 24 with maxima at 387 nm showed about 5-fold emission enhancement upon addition of Hg\(^{2+}\) due to the complexation-induced inhibition of PET and C=N isomerization. The applicability of sensor 24 was assessed in real water samples. The ESIPT and PET based sensor 25 (Figure 9) was developed for the detection of Hg\(^{2+}\) in DMF/HEPES solution (1:1, v/v) medium [56]. After interaction with Hg\(^{2+}\), sensor 25 showed fluorescence enhancement at 495 nm with 180 nm Stokes shift. The complex formation between 25 and Hg\(^{2+}\) in 1:1 stoichiometry inhibited both the ESIPT and PET processes (Figure 10), which caused significant fluorescence enhancement. The limit of detection of 25 was \(6.45 \times 10^{-7}\) M Hg\(^{2+}\) and applied in environmental and biological samples for Hg\(^{2+}\) quantification. In addition, the in situ generated complex of 25 with Hg\(^{2+}\) was applied as a secondary sensor for the detection of S\(^{2-}\).

Two polystyrene solid-phase sensors 26 and 27 (Figure 9) were synthesized with different lengths of the linker [57]. The fluorescence intensity was determined with an excitation wavelength of 401 nm for 26 and 405 nm for 27. These naphthalimide-piperazine-pyridine based sensors 26 and 27 showed fluorescence enhancement at 520 nm and 525 nm upon the incremental addition of Hg\(^{2+}\) in HEPES buffer (pH 7.2), respectively. The detection mechanism involving the Hg\(^{2+}\) sensing is chelation-induced inhibition of PET. Sensor 26 showed a higher fluorescence response than 27 with the LOD of 1.01 µM Hg\(^{2+}\). Also, sensor 26 was successfully applied to monitor Hg\(^{2+}\) in tap water and lake water.

The near-infrared (NIR) fluorescent receptor 28 (Figure 11) containing a donor-acceptor structure was reported for the detection of Hg\(^{2+}\) in THF-H\(_2\)O (9:1, v/v), where the triphenylamine-benzothiadiazole acts as a fluorophore and the rhodanine-3-acetic acid as metal ion recognition unit [58]. The complexation of Hg\(^{2+}\) with 28 in 1:1 stoichiometry at the S and O donor atoms of rhodanine-3-acetic acid enhanced the electron-donating ability and blocked the intermolecular charge transfer process, which caused significant fluorescence enhancement at 675 nm with the sensitivity limit of 13.1 nM. This low cytotoxic sensor 28 was applied for the imaging of intracellular Hg\(^{2+}\) in live A549 cells and zebrafish larvae. The chemosensor 29 (Figure 11) based on the piperazine derivative was developed for the detection of Hg\(^{2+}\) ion in mixed aqueous DMSO. In UV–vis spectral analysis, sensor 29 showed a strong ICT band at 495 nm that significantly blue-shifted (13 nm), while the emission band at 543 nm was greatly enhanced in the presence of Hg\(^{2+}\) with LOD of 19.2 nM. The sensing mechanism was explained by the aspect that after addition of
Hg$^{2+}$, the electron donating ability of aniline group was reduced that suppressed the PET process in sensor resulting fluorescent turn-on detection of Hg$^{2+}$ (Figure 12A) along with the naked-eyes colorimetric change from orange to yellow. Sensor 29 was also applied for the potential application in paper strip visualization and bioimaging (Figure 12B) [59]. Another PET based sensor 30 (Figure 11) containing NBD fluorophore and thiophene ionophore was applied for the fluorescent turn-on sensing of Hg$^{2+}$. Sensor 30 showed weak emission at 587 nm in CH$_3$CN:H$_2$O (4:6 v/v), while the fluorescence enhanced by 50 folds upon complexation with Hg$^{2+}$ in 1:1 binding stoichiometry. The fluorescence enhancement was observed due to complexation-induced inhibition of PET. The limit of detection of 3.9 ppb Hg$^{2+}$ was estimated for sensor 30, and was applied to detect Hg$^{2+}$ in drinking water, live cells and plant tissues [60].

Figure 9. Molecular structures of the sensors 23 to 27.

Xiaobo et al. [61] recently introduced a bismacrocyclic polyamine-based chemosensor 31 (Figure 11) containing two 4-nitro-1,2,3-benzoxa-diazole molecules for the selective detection of Hg$^{2+}$ in CH$_3$CN/HEPES (1:9, v/v). Sensor 31 showed a fluorescence enhancement at 530 nm upon addition of Hg$^{2+}$ with the sensitivity limit of 27 nM. The binding stoichiometry of 31-Hg$^{2+}$ complex was 1:1 determined by Job’s plot and ES-MS. Sensor 31 was applied to monitor exogenous Hg$^{2+}$ in living HeLa cells. Furthermore, the complex 31-Hg$^{2+}$ was applied for the detection of glutathione (GSH) in FBS and human serum.

Madhusmita et al. [62] reported a dual-mode sensor 32 (Figure 11) containing a styrylpyridinium dye for the detection of Hg$^{2+}$ in mixed methanol-H$_2$O (4:1, v/v) medium. Orange color solution of 32 turned to colorless under daylight, whereas started emitting yellow-color under UV light after adding of Hg$^{2+}$. The weak fluorescence band of 32 at 590 nm showed a remarkable blue-shift and enhancement at 566 nm with the LOD of 4.8 µM. Complexation of 32 with Hg$^{2+}$ in 1:1 binding stoichiometry inhibited the PET and increase the conformational rigidity that caused the chelation enhanced fluorescence
(CHEF) at 590 nm. Sensor 32 was applied for Hg$^{2+}$ detection in test paper strips, bioimaging in *E. coli* DH5-α cells and mimicking INHIBIT molecular logic gate.

**Figure 10.** (A) Proposed binding mode and responding mechanism of 25 with Hg$^{2+}$. (B) Confocal fluorescence images of HeLa cells incubated by 25 (a, d, and g), 25-Hg$^{2+}$ (b, e, and h) and 25-Hg$^{2+}$ + S$^{2-}$ (c, f, and i), respectively. Reproduced with permission from [56]. Copyright 2020 Elsevier.

Guilin et al. [63] reported a terpyridine-based probe 33 (Figure 11) for detection of Hg$^{2+}$ in aqueous solution. Probe showed the aggregation-induced emission (AIE) property in mixed DMSO/H$_2$O mixture. In acidic medium (pH = 2), significant fluorescent enhancement was noticed in presence of Hg$^{2+}$ with the red-shifting from 453 nm to 521 nm. The fluorescence enhancement is due to the complexation of 33 with Hg$^{2+}$ in 2:1 ratio followed by coordination-triggered self-assembly of 33. Additionally, probe displayed highly efficient removal of Hg$^{2+}$ ions from solution by rapid precipitation. In another approach, the triphenylamine (TPA) based NIR fluorescent sensor 34 was developed for the sensing of Hg$^{2+}$. Sensor 34 (Figure 11) showed AIE properties with the red-emitting fluorescence at 639 nm in DMSO/H$_2$O (1:99 v/v) mixed media. Sensor 34 is weakly emissive in 80% H$_2$O-DMSO mixed solvent, but with the addition of Hg$^{2+}$ caused significant fluorescence enhancement with the spectral shift from 600 nm to 639 nm. The strong fluorescence appeared due to the Hg$^{2+}$-directed aggregation of 34 with the sensitivity limit of 30 nM. Sensor 34 was applied for the bio-imaging in HepG-2 cells [64].
Hai-Ling et al. reported a Cu(II)-based three-dimensional zwitterionic MOF and then functionalized with carboxyfluorescein labeled thymine-rich (T-rich) DNA for the sequential detection of Hg$^{2+}$ and biothiols. The non-fluorescent hybrid MOF showed fluorescence enhancement at 518 nm due to the formation of hairpin-like T-Hg$^{2+}$-T structure with the sensitivity limit of 3 nM. The formation of rigid complex, the MOF is separated that recovered the fluorescence of dye. This MOF based sensing approach was applied on the environmental water and serum samples for Hg$^{2+}$ and homocysteine recovery [65].

Inal et al. [66] reported a salicylaldehyde derived sensor (Figure 13) for the determination of Hg$^{2+}$, Zn$^{2+}$, and Cd$^{2+}$ in ethanol-aqueous medium. The formation of -Hg$^{2+}$, -Zn$^{2+}$, and -Cd$^{2+}$ complexes resulted significant fluorescence enhancement at 491, 452, and 474 nm, respectively. Sensor formed complexes with Hg$^{2+}$ and Zn$^{2+}$ in 2:1 ratio whereas with Cd$^{2+}$ in 1:1 ratio. Sensor showed LOD of 270, 750, and 600 nM towards Zn$^{2+}$, Hg$^{2+}$, and Cd$^{2+}$, respectively. In another work, the quinolone-based sensor (Figure 13) was introduced for the detection of Hg$^{2+}$ with the fluorescence method. In presence of Hg$^{2+}$, the weakly emissive at 463 nm undergoes large fluorescence enhance-
ment at 490 nm, and the fluorescent color changed from faint blue to green [67]. The sensing mechanism was attributed to complex formation between 37 and Hg\(^{2+}\) which inhibited the PET and the excited-state intramolecular proton-transfer (ESIPT). The detection limit of Hg\(^{2+}\) is 2.1 nM. The binding stoichiometry between 37 and Hg\(^{2+}\) is 1:1 performed by job’s plot. Sensor 37 gives the reversible response with the addition of NaBH\(_4\). In addition, sensor 37 showed selective changes in the presence of Cu\(^{2+}\).

![Figure 12](image-url)

**Figure 12.** (A) Proposed binding mode and responding mechanism of 29 with Hg\(^{2+}\). (B) Fluorescence images of three-day-old zebrafish incubated with 29 (20 µM) at 28 °C for 30 min (a–c), subsequent treatment with Hg\(^{2+}\) (40 µM) for 30 min (d–f) and further incubation with I\(^-\) (40 µM) for 30 min (g–i). \(\lambda_{ex} = 488\) nm; yellow channel: \(\lambda_{em} = 500-600\) nm; scale bar: 500 µm. Reproduced with permission from [59]. Copyright 2020 Elsevier.
Xiao et al. [68] reported the boron dipyrromethene (BODIPY) based monomeric 38 and polymeric sensors (39 and 40) for the detection of Hg\textsuperscript{2+} in DMF/buffer (8:2, pH = 7.0) (Figure 13). Emission band of 38 at 529 nm was enhanced after addition of Hg\textsuperscript{2+} due to the complexation-induced inhibition of PET with a sensitivity limit of 2.40 µM, and also the color of sensor solution turned from orange to orange-green. Similar to 38, the polymeric sensors 39 and 40 also showed high selectivity towards Hg\textsuperscript{2+}, and their fluorescence enhanced respectively at 621 and 614 nm. The LOD of sensors 39 and 40 to Hg\textsuperscript{2+} was estimated as 2.86 µM and 0.22 µM, respectively. The polymeric sensors 39 and 40 also showed the colorimetric response from colorless to pink after Hg\textsuperscript{2+} addition. These low cytotoxicity sensors 38–40 showed good cell permeability and applied successfully to monitor intracellular Hg\textsuperscript{2+} in live A549 cells and zebrafish.

Rhodamines are extensively applied for the designing of fluorescent turn-on sensors, where the colorless and non-fluorescent ring-closed spirolactam form turned to ring-opened form upon interaction with target analyte that caused significant fluorescent enhancement and color change from colorless to an intense color. The complexation-promoted ring-opening of rhodamines is widely used for the designing of many Hg\textsuperscript{2+} selective fluorescent turn-on sensors. With some exceptions, majority of the sensors discussed here showed dual-mode chromo-fluorogenic response but the fluorescence changes of the sensors are discussed because of their high sensitivity than the UV–vis method with possible appli-
cation in bioimaging. The rhodamine B based fluorescent organic nanoparticles (FONs) 41 (Figure 14) was prepared via the reprecipitation technique. Upon addition of Hg\(^{2+}\), the fluorescence of 41 was enhanced at 532 nm (\(\lambda_{\text{exc}} = 480\) nm) due to the chelation-enhanced fluorescence (CHEF) phenomenon that open the spirolactum ring. Sensor 41 with the LOD of 8.619 nM was successfully applied to quantify Hg\(^{2+}\) in environmental samples (tap and river water) and for intracellular Hg\(^{2+}\) imaging [69]. With the spirolactum ring opening mechanism, several Hg\(^{2+}\) selective fluorescent turn-on sensors were reported. The polyacrylamide-fluorescein based sensor 42 was reported for the fluorescent turn-on sensing of Hg\(^{2+}\) (\(\lambda_{\text{em}} = 515\) nm, \(\lambda_{\text{ex}} = 460\) nm) in PBS buffer (pH = 7.0) [70]. Sensor 42 (Figure 14) showed turn-on fluorescent response to Hg\(^{2+}\) due to the complexation-induced opening of spirolactum ring with the detection limit of 0.4 nM. Sensor 42 showed biological compatibility and cell permeability and successfully applied for turn-on fluorescent determination of Hg\(^{2+}\) both in aqueous samples (lake and tap water) and living cells.

Kaijie et al. [71] reported the rhodamine-based sensor 43 (Figure 14) for the selective detection of Hg\(^{2+}\) in CH\(_3\)CN-HEPES buffer (1:9, \(v/v\)). After addition of Hg\(^{2+}\), sensor 43 showed fluorescence enhancement at 580 nm due to the coordination between 43 and Hg\(^{2+}\) in 2:1 binding stoichiometry followed by opening of rhodamine spirolactam ring. In addition, the sensor 43 showed colorimetric response for the detection of Cu\(^{2+}\). Heng et al. [72] reported a diarylethene and triazole-linked rhodamine B based sensor 44
(Figure 14) for the recognition of Hg\textsuperscript{2+} in DMSO (\(\lambda_{\text{ex}} = 520\) nm, \(\lambda_{\text{em}} = 606\) nm). Sensor 44 showed a 88-fold fluorescence enhancement at 606 nm with the addition of Hg\textsuperscript{2+} due to the complexation-induced opening of the rhodamine-spirolactam ring. This sensor showed a detection limit of 0.13 \(\mu\text{M}\) and applied to mimic the INHIBIT logic gate by taking Hg\textsuperscript{2+} and TFA as two molecular inputs.

Jin et al. reported a near-infrared fluorescent sensor 45 (Figure 15) for the selective detection of Hg\textsuperscript{2+} in HEPES buffer (10 mM, pH = 7.4, containing 20% CH\textsubscript{3}CN), where the thiosemicarbazide moiety served as a recognition site [73]. Sensor 45 showed the potential response towards Hg\textsuperscript{2+} by absorption and fluorescence spectra with the detection limit as low as 1.5 nM with fast response times (3 min). Sensor 45 showed fluorescence enhancement at 691 nm with the large Stokes shift (78 nm), while in the absorption spectra, sensor gives the intense absorption at 613 nm after binding with Hg\textsuperscript{2+} in 1:1 stoichiometry leading to the color change from colorless to dark blue was observed. Sensor 45 also applied as an efficient organelle-targeting sensor for Hg\textsuperscript{2+} in mitochondria of living cells imaging. In another work, the same sensor 45 was applied for the sensing of Hg\textsuperscript{2+} in in HEPES buffer solution (10 mM, pH 7.4, containing 50% EtOH) [74]. Sensor 45 showed the specific fluorescence enhancement at 664 nm (\(\lambda_{\text{ex}} = 590\) nm) with large Stokes-shift after addition of Hg\textsuperscript{2+} with the fluorescent color change from colorless to deep red. Sensor also showed the colorimetric response with changing the color from colorless from dark blue with naked-eyes. The determined LOD for Hg\textsuperscript{2+} is 1.87 ppb. Mechanism of colorimetric and fluorescence response were explained by desulfurization-cyclization reaction promoted by mercury ions, resulting in the formation of spirolactum ring-opening products. Sensor 45 was also applied in different real water samples.

Asif et al. [75] reported a water soluble \(p\)-sulphonatocalix[4]arene derived sensor 46 appended with a rhodamine dye for the detection of Hg\textsuperscript{2+} in aqueous medium (Figure 15). Sensor gives spectral changes after addition of Hg\textsuperscript{2+} with ‘turn-on’ fluorescent response at 574 nm with the specific color change from colorless to pink. The detection limit for Hg\textsuperscript{2+} sensing was 3.55 \(\times\) \(10^{-13}\) mL\(^{-1}\). The sensing mechanism was explained by inhibition of PET and the fluorescence enhanced due to chelation-enhanced fluorescence (CHEF) after forming a complex 46-Hg\textsuperscript{2+} in 1:1 binding stoichiometric. Zifan et al. [76] reported a sensor 47 (Figure 15) containing conjugated dyad quinolone-benzothiazole and rhodamine for the ratiometric detection of Hg\textsuperscript{2+} in DMF-H\textsubscript{2}O (7/3, \(v/v\)). Upon excitation at 390 nm, the emission band at 504 nm was decreased with the addition of Hg\textsuperscript{2+}, while the rhodamine emission intensity at 613 nm was gradually increased due to the FRET with the sensitivity limit of 0.2 \(\mu\text{M}\) (Figure 16). In this process, the conjugated dyad serve as a donor and the rhodamine as an acceptor. Sensor 47 was also successfully applied in living cells. In addition, significant color change from colorless to pink attributed by Hg\textsuperscript{2+} induced by the opening of spirolactum ring.

Saswati et al. [77] reported a rhodamine coupled copillar[5]arene sensor 48 (Figure 17) for the selective sensing of Hg\textsuperscript{2+} ions in CH\textsubscript{3}CN medium. Sensor 48 was non-fluorescent, while showed strong fluorescence enhancement at 573 nm with the addition of Hg\textsuperscript{2+}, and the color changed from colorless to pink due to the complexation-induced spirolactam ring opening mechanism. Sensor 48 formed a complex with Hg\textsuperscript{2+} in 1:1 stoichiometry, and can be applied to detect Hg\textsuperscript{2+} down to 28.5 nM. Jian-Peng et al. [78] reported a supramolecular sensor 49 via host–guest inclusion complexation between the host rhodamine hydrazone functionalized pillar[5]arene and the guest bis-pyridinium derivative (Figure 17). Sensor 49 showed both chromogenic and fluorogenic for the detection of Hg\textsuperscript{2+} in DMSO/H\textsubscript{2}O (6:4, \(v/v\)). The absorbance and fluorescence respective at 562 and 585 nm was enhanced with the addition of Hg\textsuperscript{2+} due to the complexation-induced opening of spirolactam ring. The estimated LOD with the UV–vis and fluorescence methods were 4.07 \(\times\) \(10^{-7}\) M and 1.69 \(\times\) \(10^{-8}\) M, respectively. In addition, the inductively coupled plasma data supported the ability of sensor to remove Hg\textsuperscript{2+}.

Jiwen et al. [79] reported a rhodamine B based sensor 50 (Figure 17) for the detection of Hg\textsuperscript{2+}. Sensor was synthesized by combining rhodamine B fluorophore with the thiophene-
triazole unit as an ionophore. With the addition of Hg$^{2+}$, sensor 50 showed significant fluorescence enhancement at 585 nm and also the absorbance increased at 560 nm that turned the colorless solution of 50 into red. With high sensitivity, the fluorescence enhancement of 50 can be applied to detect Hg$^{2+}$ down to 16 nM. Sagar et al. [80] reported the sensor 51 (Figure 17) containing two rhodamine units linked with 2,6-pyridinedicarboxaldehyde for the selective detection of Hg$^{2+}$ in DMSO:H$_2$O (1:1; v/v). After addition of Hg$^{2+}$, the absorption band at 530 nm enhanced significantly with a visual color change from colorless to pink. While in emission spectra, new fluorescence band appeared at 562 nm due to the conversion of closed form of spirolactum ring of rhodamine to its ring opened form on both side of pyridine ring. The detection limit was obtained as 26 nM. Sensor 51 was applied for the real water samples for practical application.

Figure 15. Molecular structures of the sensors 45 to 47.
Figure 16. (A) Fluorescence responses of 47 (1 in image represents sensor 47) (10 μM) in the presence of various metal ions; (B) Fluorescence intensity at 613 nm of 47 (red bars: 47 with other metal ions, green bars: 47 with other metal ions and Hg^{2+}, λ_{ex} = 390 nm). (C) The color image of 47 (10 μM) with other metal ions. (D) The fluorescence photo of 47 (10 μM) upon addition Hg^{2+} (20 eq.) in the existence of other metal ions (20 eq.). (E) The FRET process of 47 detecting Hg^{2+}. Reproduced with permission from [76]. Copyright 2020 Elsevier.
Zixiang et al. [81] reported a rhodamine 6G based sensor 52 (Figure 18) for the selective detection of Hg\(^{2+}\) in DMSO/H\(_2\)O (7/3, v/v). The non-fluorescent sensor showed a significant fluorescence enhancement at 581 nm after addition of Hg\(^{2+}\). In UV–vis spectra, the absorption band at 538 nm become stronger after addition of Hg\(^{2+}\) with the colorimetric response from colorless to red. The hydrogel of 52 was also prepared and applied for the reversible sensing of Hg\(^{2+}\). The sensing mechanism was described by the blocking of PET process upon complexation in 1:1 stoichiometry and the rigidity of the sensor promotes to the chelation-enhanced fluorescence (CHEF) effect. The detection limit of probe for Hg\(^{2+}\) detection is 14.9 nM. Yuesong et al. [82] have reported a novel rhodamine–naphthalene derivative 53 (Figure 18) for the Hg\(^{2+}\) detection in CH\(_3\)CN-H\(_2\)O (7/3, v/v). Absorption and emission peak of 53 enhanced respectively at 554 and 604 nm upon addition of Hg\(^{2+}\). Sensor 53 is non-fluorescent due to the spirolactam structure of rhodamine moiety; however, spirolactam ring was opened in presence of Hg\(^{2+}\), and give the colorimetric and fluorescent response (Figure 19). The LOD of sensor 53 for Hg\(^{2+}\) detection are 0.12 µM and 0.38 µM by using absorption and emission analysis, respectively. Sensor 53 was also applied for the test strips and biosensing applications. Guohua et al. [83] have reported a triazole-rhodamine conjugate 54 (Figure 18) for the selective detection of Hg\(^{2+}\) in DMF/H\(_2\)O (1:1, v/v, Tris-HCl buffer, pH = 7.4). Free rhodamine sensor showed no fluorescence. However, the com-

**Figure 17.** Molecular structures of the sensors 48 to 51.
plexation of 54 with Hg\textsuperscript{2+} in 2:1 stoichiometry, the emission enhanced at 557 nm with the color changed from colorless to pink due to the spirolactam ring opening mechanism. The calculated LOD is 1.61 nM. UV–vis spectra also support the same process by exhibiting absorbance band at 563 nm. The Hg\textsuperscript{2+} detection by sensor 54 was applied for fluorescence imaging in HeLa cells. In another approach, Zhao et al. \cite{84} reported a ferrocenyl containing rhodamine B based sensor 55 (Figure 18) for the detection of Hg\textsuperscript{2+} in H\textsubscript{2}O/THF (4:1, v/v). Sensor 55 showed fluorescence off-on response at 590 nm with the addition of Hg\textsuperscript{2+} due to the formation of a complex in 1:1 stoichiometry. Sensor 55 showed a low detection limit of 16 nM and fast response time (<3 min). Mechanism of Hg\textsuperscript{2+} detection is attributed by desulfurization annulation that triggers the spirolactam ring-opening. Sensor 55 was applied for monitoring of intracellular Hg\textsuperscript{2+} ions in living cells.

![Molecular structures of the sensors 52 to 55](image)

**Figure 18.** Molecular structures of the sensors 52 to 55.

Zhong et al. \cite{85,86} have reported three rhodamine 6G derivative 56–58 (Figure 20) for the fluorescent turn-on sensing of Hg\textsuperscript{2+} in DMSO/H\textsubscript{2}O (7.3, v/v). Because of the complexation-induced spirolactam ring opening, the weakly emissive 56, 57, and 58 showed significant fluorescence enhancement respectively at 582, 578, and 560 nm. The sensors 56, 57, and 58 showed the LOD of 13.4, 15.6, and 16.1 nM respectively for Hg\textsuperscript{2+}. In addition, the absorption of 56, 57, and 58 enhanced respectively at 536, 537, and 534 nm leading to the naked-eye detectable color change from colorless to pink. Recently, Wei et al. \cite{87} applied the fluorescent sensor 59 (Figure 20) encapsulated in the hydrogel microsphere for the detection of Hg\textsuperscript{2+} by using a microfluidic device. The non-fluorescent sensor 59 showed fluorescence enhancement with the color change to red upon addition of Hg\textsuperscript{2+}. Sensor 59 showed a reversible response with EDTA and KI. The limit of detection for Hg\textsuperscript{2+} is 120 nM. Hydrogel microsphere probe was also applied for the detection of Hg\textsuperscript{2+} in real water samples.

Xuliang et al. \cite{88} developed a dansyl-peptide based Hg\textsuperscript{2+} selective sensor (60, dansyl-Glu-Cys-Glu-Trp-NH\textsubscript{2}). Sensor 60 showed two emission maxima at 337 and 550 nm due to the tryptophan and dansyl fluorophores. Addition of Hg\textsuperscript{2+} caused chelation-induced fluorescence enhancement at 550 with a blue-shift to 505 nm, and the FRET from tryptophan (donor) to dansyl (acceptor) caused quenching at 337 nm. Sensor 60 can be applied to detect Hg\textsuperscript{2+} down to 23.0 nM. Sequentially, the in situ generated 60-Hg\textsuperscript{2+} complex was applied for the sensing of biothiols. The lanthanide-complexes of Tb\textsuperscript{3+} are also applied for the fluorescent sensing of Hg\textsuperscript{2+}. For example, the ratiometric sensor 61 (Figure 21) based on
the lanthanide coordination polymers (CPs) between Tb\(^{3+}\), guanine monophosphate (GMP) and luminol was developed for the detection of Hg\(^{2+}\) [89]. The addition of Hg\(^{2+}\) leads to the decrease of Tb\(^{3+}\) luminescence at 548 nm due to the higher coordination between Hg\(^{2+}\) and GMP, which inhibits energy transfer from GMP to Tb\(^{3+}\). While, the fluorescence of luminol at 430 nm increased due to the aggregation-induced emission phenomenon. The ratiometric response of 61 for Hg\(^{2+}\) can be detected down to 1.3 nM. Sensor 61 was successfully used for the determination of Hg\(^{2+}\) in tap water.

Peng et al. [90] reported a dansyl based sensor 62 (Figure 21) for the fluorescence turn-on detection of Hg\(^{2+}\) in HEPES buffer. The emission of 62 at 545 nm was weak based on a single dansyl group. The formation of a complex 62-Hg\(^{2+}\) in 2:1 stoichiometry resulted conformational adjustment that reduced the distance between two dansyl groups and formed the dansyl dimer (Figure 22). The monomer-excimer mechanism resulted significant fluorescence enhancement at 515 nm with the sensitivity limit of 22.65 nM. Muzey et al. [91] reported a naphthalimide-sulfamethizole conjugated sensor 63 (Figure 21) for the ratiometric detection of Hg\(^{2+}\) in DMSO/HEPES medium (1:99, v/v). The strong fluorescence from the monomeric form of 63 at 390 nm was quenched and a new band appeared at 483 nm due to the complexation-induced formation of excimer. Sensor 63
formed a complex with Hg$^{2+}$ in 2:1 ratio that bring the naphthalimide close together to form the excimer. Sensor 63 showed LOD of 14.7 nM, and capable to quantify Hg$^{2+}$ concentration in real water samples. Tapashree et al. [92] reported a pyrene-hydroxyquinoline conjugated azine based Schiff base 64 (Figure 21) for the selective detection of Hg$^{2+}$ in ethanol-H$_2$O (9:1 v/v) medium. The absorption bands of 64 at 240 and 290 nm were enhanced with the appearance of a new band at 450 nm upon addition of Hg$^{2+}$. The color also changed from lemon yellow to golden yellow attributed to deprotonation (–OH) upon coordination with Hg$^{2+}$ in 1:1 stoichiometry. The monomeric and excimer emission bands centered at 385 and 447 nm of 64 were enhanced due to CHEF effect that inhibited the C=N isomerization and suppressed the PET. Sensor 64 showed the LOD of 0.22 µM. The monomer-excimer based fluorescent sensor 65 (Figure 21) was reported for the selective detection of Hg$^{2+}$ in CH$_3$CN/DMSO (99:1) [93]. The pyrene appended calix[4]arene sensor 65 showed monomer emission at 395 nm. Upon complexation with Hg$^{2+}$ in 2:1 ratio, the excimer emission enhanced at 472 nm with the sensitivity limit of 8.11 nM. Sensor 65 also showed fluorescence response towards Ag$^+$. Using Hg$^{2+}$ and Ag$^+$ as two chemical inputs, the changes in the fluorescence of 65 was studied to mimic the INHIBITION and IMPLICATION logic gates.

**Figure 20.** Molecular structures of the sensors 56 to 59.

### 3.1.3. Reaction-Based Chemosensors

Chemodosimeter, an irreversible reaction based detection approach, where the probe contains a reactive site that interacts with the target analyte and formed a new product that emits differently from the original probe molecule. In compared to the reversible fluorescent sensors, the reaction-based fluorescent sensors showed better selectivity and specificity due to the structural changes occurred upon chemical reactions with the target analyte. The 7-hydroxycoumarin-derived carbonothioate-based sensor 66 (Figure 23) was designed and synthesized by Xiwei et al. [94] for the detection of Hg$^{2+}$ at the maximum emission wavelength 455 nm in water medium. Upon interaction with Hg$^{2+}$, sensor 66 showed a large fluorescence enhancement because of the strengthening of the intramolecular charge transfer (ICT) due to the Hg$^{2+}$ directed hydrolysis of 66 to form HgS and phenol. The sensor 66 showed a LOD of 7.9 nM. Sensor 66 was successfully applied to detect Hg$^{2+}$ in different water samples (river water), living cells and in zebrafish. With similar
sensing mechanism, the multi-analytes selective chemodosimeter 67 (Figure 23) based on naphthalene fluorophore was developed for the rapid detection of Hg\(^{2+}\), hydrazine and H\(_2\)S in C\(_2\)H\(_5\)OH \cite{95}. This probe possesses multiple reactive groups phenyl thiobenzoate, carbon-carbon double bond α,β-unsaturated ketone for the detection of Hg\(^{2+}\), H\(_2\)S and hydrazine, respectively (Figure 24). With Hg\(^{2+}\), the phenyl thiobenzoate detached from 67 and formed HgS and phenol that resulted in the fluorescence quenching at 580 nm. The Hg\(^{2+}\) sensitivity limit of 67 is 1.10 µM, and applied for the detection of Hg\(^{2+}\) in paper test strips and environmental water samples (seawater, tap water, and mineral water).

In another work, the naphthalene derived probe 68 (Figure 23) was introduced for the selective chemodosimetric detection of Hg\(^{2+}\) in DMSO/H\(_2\)O (1:3, v/v) \cite{96}. After addition of Hg\(^{2+}\), the fluorescence intensity enhanced at 444 nm and 644 nm due to desulfurization reaction of 68 leading to the formation of HgS and phenol. The probe 68 showed a LOD of 48.79 nM for Hg\(^{2+}\), and applied successfully for quantifying Hg\(^{2+}\) in various environmental and beverages samples. In addition, probe 68 was applied for the detection of hydrazine.

Figure 21. Molecular structures of the sensors 61 to 65.
**Figure 22.** Proposed principle of 62 for detecting Hg$^{2+}$. Reproduced with permission from [90]. Copyright 2020 Elsevier.

**Figure 23.** Molecular structures of the sensors 66 to 73.
Zhixiu et al. [97] synthesized a coumarin based sensor 69 (Figure 23) with the conjugation of thiourea for turn-on detection of Hg$^{2+}$ in EtOH/H$_2$O (2:8, v/v) over a broad pH range of 1-11. Addition of Hg$^{2+}$ to the sensor 69 solution induced a hypsochromic shift of the UV-vis absorption band at 360 nm to 340 nm. Additionally, a gradual enhancement in the fluorescence emission intensity was observed at 475 nm with the detection limit of $1.46 \times 10^{-7}$ M. The sensing mechanism of probe 69 for fluorescent detection of Hg$^{2+}$ ion is proposed as weakly fluorescent probe 69 readily binds with Hg$^{2+}$ ion due to the strong interaction between sulphur atom and thiophilic Hg$^{2+}$ ion. Then, a desulfurization and cyclization process occurred to form strong fluorescence. Sensor 69 was applied in real waste water sample for detecting Hg$^{2+}$. The coumarin-based ratiometric fluorescent probe 70 (Figure 23) was reported for the sensing of Hg$^{2+}$ ion in ethanol and HEPES buffer (1:9, v/v) medium [98]. Probe 70 was synthesized by the catalytic reaction between coumarin-red dye and DL-dithiothreitol. Probe showed two characteristic emission band at 495 nm and 600 nm, while addition of Hg$^{2+}$, the band at 495 nm was disappeared and the second band at 600 nm was significantly enhanced. The detection limit of probe for Hg$^{2+}$ is 1.6 nM. The DL-dithiothreitol moiety of 70 serve as the recognition receptor for Hg$^{2+}$. After recognition, the DL-dithiothreitol moiety detached from 70 and formed the $\alpha,\beta$-unsaturated ketone. This chemodosimeter based probe was applied for the detection of Hg$^{2+}$ ion in real water sample, and in living cells and zebrafish.

The fluorescent probe 71 (Figure 23) was developed by the reaction of 6-hydroxy-2-naphthaldehyde and dimethylcarbamothioic for the detection of Hg$^{2+}$ in aqueous medium [99]. After addition of Hg$^{2+}$, the fluorescence of 71 was enhanced at 443 nm due to the Hg$^{2+}$-catalyzed desulfurization reaction to form HgS and 6-hydroxy-2-naphthaldehyde. The LOD of 71 for Hg$^{2+}$ was calculated to be 39.28 nM. Probe was applied for the detection of Hg$^{2+}$ in test paper strips and in real water samples. Probe 71 also showed selective fluorescence response for the detection of H$_2$S. The chemodosimetric probe 72 (Figure 23) based on perylene diimide dye was designed for the detection of Hg$^{2+}$ in THF-H$_2$O (1:9, v/v) [100]. Highly fluorescent 72 showed a significant fluorescence quenching at 667 nm upon interaction with Hg$^{2+}$, where the butynoxy group serve as the reactive site for Hg$^{2+}$ detection. The LOD of 60 nM and 33 nM was determined by UV-vis and emission methods, respectively. Sensor were successfully applied detection of Hg$^{2+}$ in blood serum and urine and bioimaging in MG-63 cells. The thioxothiazolidin-coumarin based chemodosimeter 73 (Figure 23) was introduced for the selective sensing of Hg$^{2+}$ in HEPES-DMSO (99/1, v/v) [101]. After interaction with Hg$^{2+}$, the emission band at 630 nm was blued-shifted
and enhanced significantly at 580 nm. Remarkable changes were obtained in absorption at 530 nm with decreasing intensity and appearance of a new band at 485 nm, and also the color changed from dark to light pink. The detection limit of Hg$^{2+}$ was estimated to be 15.6 µM and 15.1 µM by absorbance and fluorescence methods, respectively. Sensing of Hg$^{2+}$ by sensor 73 is based on the desulfurization reaction of thiocarbonyl to carbonyl, which was supported by $^{13}$C NMR and HRMS-ESI analyses. Probe 73 was applied for detecting Hg$^{2+}$ in living cells by bioimaging experiment.

The phenothiazine-derived chemodosimetric probe 74 (Figure 25) containing dithioacetal unit was applied for the detection of Hg$^{2+}$ [102]. Upon interaction with Hg$^{2+}$, the UV–vis absorption band of 74 at 320 nm was shifted to 390 nm with the appearance of yellow color, while the weakly fluorescent 74 showed significant fluorescence enhancement with a remarkable red-shift from 455 nm to 610 nm due to intramolecular charge transfer process (ICT). With high sensitivity, the fluorescence change of 74 was applied to detect Hg$^{2+}$ down to 21.2 nM. Sensor 74 was applied successfully to detect Hg$^{2+}$ in drinking water and live cells. With the similar sensing mechanisms, the Hg$^{2+}$ selective fluorescent probes 75–79 were reported. The AIE active chemodosimeter 75 (Figure 25) was reported for the fluorescent turn-on sensing of Hg$^{2+}$ in PBS buffer (10 mM, pH 7.4, containing 1% DMSO). Sensor 75 showed a weak emission at 475 nm. With the Hg$^{2+}$ directed hydrolysis of 75 at the dithioacetal unit to aldehyde enhanced the ICT effect and also restricted the intramolecular rotations that amplified the fluorescence at 495 nm based on AIE effect. The detection limit of sensor 75 for Hg$^{2+}$ is 36 nM. Sensor 75 was applied for the detection of Hg$^{2+}$ in real water sample and in living cell imaging [103].

Zhonglong et al. [104] reported a camphor-based fluorescent turn-on probe 76 (Figure 25) for the detection of Hg$^{2+}$ with large Stokes shift of 153 nm in 99% PBS buffer medium. Probe 76 exhibited a maximum absorption at 322 nm that reduced remarkably while a new band emerged at about 355 nm after adding Hg$^{2+}$. With high sensitivity, the fluorescence of 76 was enhanced at 518 nm upon interaction with Hg$^{2+}$. The 1,3-dithiane unit of the 76 can be deprotected into a formyl group under the function of Hg$^{2+}$, thus the probe 76 is transformed into compound 77-CHO. The detection limit of 76 is 19.3 nM for Hg$^{2+}$. Sensor was applied in cell imaging experiment and to quantify Hg$^{2+}$ in environmental water samples (tap, distilled, and lake water). The bithiophene-based sensor 77 (Figure 25) was reported for the ultra-rapid detection of Hg$^{2+}$ in aqueous medium with the fluorescent color changed from colorless to blue under UV light irradiation [105]. Significant enhancement in emission of 77 at 470 nm was observed due to the Hg$^{2+}$ induced desulfurization reaction that strengthened the ICT upon conversion of bithiophene moiety in to aldehyde group (Figure 26). With 77, the Hg$^{2+}$ concentration can be detected down to 19 nM. Sensor 77 also showed selective changes in the absorption by quenching of absorbance at 334 nm and appearance of a new band at 370 nm after the addition of Hg$^{2+}$. The fluorescence changes of 77 was applied to quantify Hg$^{2+}$ detection in water, seafood as well as human urine samples. In addition, Sensor 77 was applied for Hg$^{2+}$ detection by developing test paper strips and performing bio-imaging in HeLa cells.

Meiju et al. [106] reported a fluorescent sensor 78 (Figure 25) based on the naphthalimide derivative for the detection of Hg$^{2+}$ in PBS buffer. In UV–vis absorption, the sensor 78 showed blue-shift from 461 to 417 nm with Hg$^{2+}$. The fluorescence emission enhanced at 510 nm with increasing concentration of Hg$^{2+}$. In the presence of Hg$^{2+}$, the sensor 78 reacted specifically with the mercury ion to produce an aldehyde and emitted strong fluorescence, and the yellow color of the solution turned to light green. The detection limit for Hg$^{2+}$ was found to be 40 nM. The sensor 78 was successfully applied to the living cell imaging to detect Hg$^{2+}$ in PC-12 cells. The tetraphenylenethylen (TPE) derivative 79 (Figure 25) reported by Long et al. showed aggregation-induced emission features in THF/water mixtures. TPE derivatives maintain AIE activities after grafting on fibers, however, the strong fluorescence emission at 477 nm was gradually weakened after Hg$^{2+}$ addition due to Hg$^{2+}$-initiated cleavage of dithioacetal moieties. The LOD reached as low as 20 nM Hg$^{2+}$. In addition, the electrospun fibrous strips with crafted TPE and
dithioacetal moieties are designed for the detection of trace Hg\textsuperscript{2+} with the visual change of color strip from green to blue [107].

Figure 25. Molecular structures of the sensors 74 to 79.
Figure 26. (A) Proposed sensing mechanism of \(77 (2TS = 77)\) for \(\text{Hg}^{2+}\). (B) Fluorescence spectra of \(77 (10 \mu\text{M})\) after addition of 20 \(\mu\text{M}\) of various tested ions in 100% aqueous solution; Inset: fluorimetric responses of \(77 (10 \mu\text{M})\) in 100% aqueous solution after the addition of various tested ions. Reproduced with permission from [105]. Copyright 2020 Elsevier.

Abani et al. [108] reported a trinuclear \(\text{Zn(II)}/\text{Cd(II)}\) Schiff base complexes 80 and 81 for the detection of \(\text{Hg}^{2+}\) in aqueous medium via chemodosimetric approach (Figure 27). Two absorption maxima at 388 nm and 390 nm were red-shifted and the colorless solution turned to distinct yellow. Complexes showed emission maxima at \(\lambda_{\text{em}} \sim 461\) nm (80) and 464 nm (81) were red-shifted to 475 and 472 nm, respectively and undergo considerable decrease in fluorescence intensity. The LOD estimated for complexes 80 and 81 to detect \(\text{Hg}^{2+}\) were 1.11 and 1.89 \(\mu\text{M}\), respectively. The most probable chemodosimetric mechanism explained via the cleavage of the imine bond through hydrolysis. Results were confirmed by different spectroscopic techniques including \(^1\text{H}\) NMR titration.
Chunqing et al. [109] reported the monomeric BODIPY based Schiff bases 82 and 83, and the polymeric derivative 84 for the fluorescent turn-on sensing of Hg\textsuperscript{2+} and Fe\textsuperscript{3+} in DMF/H\textsubscript{2}O (1:1, v/v) (Figure 27). Both the selective metal ions hydrolysed the imine linkage and formed the original BODIPY aldehyde. The emission and absorption peak of the probes were blue-shifted and enhanced. The emission of 82, 83, and 84 at 549, 550, and 559 nm was blue-shifted and enhanced respectively at 523, 529, and 528 nm. The probes 82, 83, and 84 showed the LOD of 0.21, 0.63, and 0.19 µM respectively for Hg\textsuperscript{2+}. The polymeric probe 84 showed high sensitivity than the probes 82 and 83. Similarly, the absorption of 82, 83, and 84 at 520, 545, and 548 nm was blue-shifted and enhanced respectively at 490, 499, and 501 nm.

3.2. Colorimetric Sensors

The colorimetric sensors provide naked-eyes detectable color change for the cost-effective of target analytes (Table 3). The ruthenium derived complex 85 (Figure 28) was reported for the colorimetric detection of Hg\textsuperscript{2+}, where the coordination of mode of Ru\textsuperscript{2+} to the C-atom is changed to S-atom with the addition of Hg\textsuperscript{2+} (Figure 29). The Hg\textsuperscript{2+}-prompted switch in coordination mode in 85 caused a color from dark red to light yellow, and the absorption band at 506 and 730 nm are gradually quenched with the sensitivity limit of 21 nM [110]. With the similar approach, the ruthenium complex based sensor 86 (Figure 28)
was reported for the selective colorimetric detection of Hg\textsuperscript{2+}. The color of 86 turned from red to yellow, due to the formation of a new low energy band at 410 nm, while pre-existed band at 503 declined with the addition of Hg\textsuperscript{2+}. The detection limit of 86 for Hg\textsuperscript{2+} is 0.053 µM. Job’s plot confirmed the binding stoichiometry of sensor 86-Hg\textsuperscript{2+} complex is 1:1 mode. For the practical applicability, the sensor was grafted into a polymer membrane and applied for the colorimetric detection of Hg\textsuperscript{2+} [111].

Table 3. Analytical parameters of the colorimetric sensors 85 to 102.

| Sensors | Medium             | $\lambda_{abs}$ (with/without Hg\textsuperscript{2+})     | LOD        | Applications                                      | Ref.   |
|---------|--------------------|------------------------------------------------------------|------------|---------------------------------------------------|--------|
| 85      | Aqueous media      | Quenching of bands at 506 and 730 nm                        | 21 nM      | -                                                  | [110]  |
| 86      | DMSO:HEPES (5:95, v/v) | Quenching at 503 with the new band formation at 610 nm     | 53 nM      | Polymer coated membrane                            | [111]  |
| 87      | HEPES buffer       | Quenching of bands at 390 and 530 nm                       | 0.27 µM    | Test color strips                                 | [112]  |
| 88      | CH$_3$CN:CH$_2$O (7:3, v/v) | Band at 447 nm shifted to 519 nm                          | 0.473 µM   | Test color strips and real water analysis         | [113]  |
| 89      | DMSO:CH$_2$O (4:1 v/v) | Band at 502 nm shifted to 395 nm                           | 6.1 µM     | Cellulose test strips, Logic Gate Operation       | [114]  |
| 90      | BufferDMF (98:2, v/v) | Quenching of bands at 350 and enhancement at 400 nm        | 0.11 µM    | Real water sample analysis                        | [115]  |
| 91      | Aqueous medium     | Enhancement of band at 540 nm                              | 0.100 and 0.180 µg/L | Real water sample analysis | [116]  |
| 92      | Aqueous medium     | Enhancement of band at 567 nm                              | 0.22 and 0.61 µg/L | Real water sample analysis                        | [117]  |
| 93      | CH$_3$CN:CH$_2$O (1:1, v/v) | Band at 448 nm shifted to 523 nm                          | -          | -                                                 | [118]  |
| 94      | CH$_3$OH:HEPES (7:3, v/v) | Band at 414 nm shifted to 498 nm                          | 220 nM     | Real water analysis, silica coating and test color strips | [119]  |
| 95      | CH$_3$CN:CH$_2$O (1:1, v/v) | Band at 517 nm shifted to 415 nm                          | -          | Real water analysis                               | [120]  |
| 96      | HEPES buffered     | Band at 247 nm shifted to 234 nm                          | 40 nM      | -                                                 | [121]  |
| 97      | CH$_3$OH:CH$_2$O (1:1, v/v) | Band at 465 nm shifted to 485 nm                          | 25.7 nM    | INHIBIT logic gate                                | [122]  |
| 98      | MeCN:CH$_2$O (1:1, v/v) | Formation of new band at 470 and quenching at 380 nm       | 0.95 nM    | Real water sample analysis                        | [123]  |
| 99      | DMSO:CH$_2$O (2:1, v/v) | Enhancement of band at 280 nm                             | 4.89 µM    | Real water sample analysis                        | [124]  |
| 100     | H$_2$O:CH$_3$CN (9:1, v/v) | Quenching of bands at 319 and 380 nm with the formation of new band at 610 nm | 8.5 µM | Test color strips and real water sample analysis | [125]  |
| 101     | Aqueous medium     | Band at 335 nm shifted to 305 nm                          | 1 µM       | Real time application                             | [126]  |
| 102     | DMSO:CH$_2$O (8:2, v/v) | Band at 290 nm shifted and two new bands formed at 255 and 292 nm | 0.10 mM | -                                                 | [127]  |
The \( p \)-toluenesulfonate salt of merocyanine dye 87 (Figure 28) was reported for the colorimetric sensing of \( \text{Hg}^{2+} \) in HEPES buffer. Sensor 87 showed two absorption bands at 390 nm and 530 nm. After complexation with \( \text{Hg}^{2+} \), both the absorption bands showed decrease in the intensity with the color change from pink to colorless easily detected by naked eyes. Sensor showed a micromolar detection limit of 0.27 \( \mu \text{M} \) and was also applied for the visual detection of \( \text{Hg}^{2+} \) by using paper test strip of 87 [112]. The pyrazole-based colorimetric sensor 88 (Figure 28) was developed for the detection of \( \text{Hg}^{2+} \) in semi-aqueous medium. The absorption band of 88 at 447 nm was red-shifted to 519 nm with a noticeable color change from yellow to pink upon complexation with \( \text{Hg}^{2+} \) in 2:1 stoichiometry. Sensor
showed LOD of $4.73 \times 10^{-7}$ M, and was applied successfully to quantify Hg$^{2+}$ in various environmental samples [113]. In another work, the azo dye based chromogenic sensor 89 (Figure 28) was studied for selective detection of Hg$^{2+}$ in DMSO-H$_2$O (4:1 v/v) medium. Sensor 89 showed absorption at 502 nm due to strong intermolecular charge transfer (ICT) transition with the solution color as reddish-pink. Addition of Hg$^{2+}$ decreased the absorbance at 502 nm and blued-shifted to 395 nm. The complex formation between sensor 89 and Hg$^{2+}$ restricted the ICT that caused blue-shift in absorbance, and the solution become colorless. Sensor 89 formed complex with Hg$^{2+}$ in 1:1 stoichiometric through the salicylaldehyde unit and the complexation reversed with the addition of F$^-$ [114].

The conjugated Schiff base receptor 90 (Figure 30) showed colorimetric response for the detection of Hg$^{2+}$ in buffer/DMF (98:2) [115]. Only Hg$^{2+}$ showed spectral and color changes from pale yellow to orange. The absorption band at 400 nm was increased with decreasing of 90 band at 350 nm due to the formation a complex 90-Hg$^{2+}$ in 2:1 stoichiometry. The limit of detection of probe for Hg$^{2+}$ is 0.11 µM, and applied to quantify Hg$^{2+}$ in real water samples.

Thirumalai et al. [116] reported two solid templates, mesoporous silica monoliths (MSMs) and mesoporous polymer monoliths (MPMs), immobilized with the amphiphilic chromo-ionophoric 91 (Figure 30) to develop solid-state sensors for naked-eye colorimetric sensing of Hg$^{2+}$. Upon interaction with Hg$^{2+}$, the solid-state sensors showed color transition from light orange to deep red due to the metal to ligand charge transfer (MLCT). The LOD for 91-MPM and 91-MSM sensors was estimated as 0.100 and 0.180 µg/L, respectively. Both the sensors applied successfully in real sample analysis to quantify Hg$^{2+}$ concentrations. The solid-state sensor 92 (Figure 30) based on rhodamine B hydrazide derivative mobilized in mesoporous silica monolith was developed for the ultra-trace colorimetric detection of Hg$^{2+}$ from aqueous medium [117]. The absorption of 92 was changed at 567 nm after each addition of Hg$^{2+}$, and the light pink color turned to deep violet. Sensor 92 can be used to detect Hg$^{2+}$ down to 0.61 µg/L, and applied successfully for quantifying Hg$^{2+}$ ion in real water samples (ground, lake, and river water).
Hyokyung et al. [118] introduced a Pt complex 93 (Figure 30) coordinated with the ligands 1,2-bis[bis(pentafluorophenyl)phosphino]ethane and 1,3-dithiole-2-thione-4,5-dithiolate for the selective colorimetric detection of Hg²⁺ in CH₃CN/H₂O (1:1, v/v). Complex 93 showed Hg²⁺ selective color change from yellow to vivid red due to the interaction of Hg²⁺ at >C=S, and also the absorbance at 448 nm of 93 was red-shifted to 523 nm. The colorimetric sensor 94 (Figure 30) based on the anthracene moiety was developed for the detection of Hg²⁺ in HEPES buffered CH₃OH·H₂O (7:3) medium. Sensor 94 showed absorbance at 414 nm was disappeared with the formation of a new red-shifted band at 498 nm after the addition of Hg²⁺. The complexation-induced spectral change caused due to ICT also showed naked-eye detectable color change from yellow to pink. With sensor 94, the concentration of Hg²⁺ can be detected down to 220 nM. Sensor 94 was applied for the various practical applications including naked-eyes detection of Hg²⁺ using paper strips and solid silica gel, and also to quantify Hg²⁺ concentration in real water samples [119].

The benzopyran based colorimetric sensor 95 (Figure 31) was reported for the visual detection of Hg²⁺ ion in CH₃CN/H₂O medium (1:1, v/v). In this sensor, the dicyanomethylene-4H-chromene serve as a fluorophore whereas the dithiadioxo-monoaza crown ether as the recognition unit. Sensor 95 absorption at 517 nm showed hypsochromic shift with Hg²⁺ and a new band generated at 415 nm. Color of probe is also changed from rose red to yellow detected by naked eyes. Color and spectral changes mainly attributed by the blocking of ICT process. In fluorescence study, the strong emission peak at 645 nm was significantly quenched with Hg²⁺ due to the complex formation of 95-Hg²⁺ in 1:1 ratio. The detection limit is 0.14 μM. For practical applications, sensor was applied for Hg²⁺ detection in real aqueous samples and live cell imaging [120].

Zhang et al. [121] reported chemosensor 96 (Figure 31) based on azobenzene for the selective detection of Hg²⁺ in HEPES buffered solution. In UV–vis study, the sensor 96 showed characteristic absorbance at 358 and 247 nm respectively due to π-π* and n–π* transitions. After interaction with Hg²⁺, the π-π* band was suppressed and blue-shifted by 13 nm. Absorption changes arising from the pull–push effect between electron-withdrawing and electron-donating groups of the azobenzene chromophore. The nitrobenzoxadiazole-antipyrene conjugate 97 (Figure 31) was studied for the colorimetric sensing of Hg²⁺ and CN⁻ in CH₃OH·H₂O (1:1, v/v). Addition of Hg²⁺ evolved new absorption band at 530 nm and band at 465 was red-shifted to 485 nm leading to the color change from pale yellow to pink due to the formation of a charge-transfer complex between probe and Hg²⁺ in 2:1 stoichiometry. Sensor showed reversibility with Na₂S and the LOD of 2.57 × 10⁻⁸ M Hg²⁺ [122]. The colorimetric sensor 98 (Figure 31) was reported for the detection of Hg²⁺ and Cu²⁺ in MeCN·H₂O (1:1, v/v). The colorless solution of 98 turned brick-red to the naked eye with the addition of Hg²⁺ and Cu²⁺, respectively due to the complexation-induced LMCT. The complexation led to the formation of a new absorption at 470 nm and the quenching of sensor band at 380 nm. The estimated detection limit for Hg²⁺ is 0.95 nM, and the sensor applied for real sample analyses [123].

The azo-phenylthiourea based receptor 99 (Figure 31) was applied as a colorimetric sensor for Hg²⁺ in DMSO/H₂O (2:1, v/v). Sensor 99 showed an absorption band centered at 365 nm, attributed to ICT of azo skeleton. Interaction with Hg²⁺ in 1:1 stoichiometry, sensor 99 generated a new ICT band at 280 nm with the significant hypsochromic shift of 85 nm. Sensor 99 showed the lower detection limit of 4.89 μM for Hg²⁺, and applied for the monitoring of Hg²⁺ in the real samples [124]. In another work, Gargi et al. reported an azo dye based colorimetric sensor 100 (Figure 31) for the detection of Hg²⁺ in 9:1 (v/v) aqueous CH₃CN. After addition of Hg²⁺ to the colorless solution of 100 showed a new absorption band at 610 nm that enhance π-delocalization and reduced the energy of π → π* transition leading to the appearance of greenish-blue color. Job’s plot confirmed 1:1 stoichiometry between 100 and Hg²⁺. The limit of detection was 8.5 μM, and the sensor was applied for the real water analysis [125].
Figure 31. Molecular structures of the sensors 95 to 102.

The ninhydrin–thiosemicarbazone based sensor 101 (Figure 31) was developed for the colorimetric sensing of Hg$^{2+}$ in aqueous medium [126]. After complexing with Hg$^{2+}$ in 1:1 ratio, the absorption band of 101 at 335 nm was shifted to 305 nm without any pH effect, and the solution color turned from yellow to colorless. Addition of strong chelating agent ETDA reversed the color change occurred due to the 101-Hg$^{2+}$ complex formation in solution. The LOD of 101 for Hg$^{2+}$ detection was 1 µM. Gurjaspreet et al. [127] prepared an antipyrine based sensor 102 (Figure 31) for the detection of Hg$^{2+}$ and Fe$^{3+}$ in DMSO/H$_2$O (8/2 v/v) by UV–vis method. After interaction with Hg$^{2+}$, the absorption band of sensor at 290 nm was blue-shifted with the evolution of two bands at 255 nm and 292 nm due to the participation of azomethine linkage in the formation of Hg-N bond. The 102 LOD to detect Hg$^{2+}$ was estimated as 0.10 mM.

4. Conclusions

In this review, we have summarized 102 chromo-fluorogenic chemosensors reported in the year 2020 for the sensing of mercuric ion. Most of the developed sensors are easy-to-prepare, low cost, and showed high selectivity and rapid response. In compared to colorimetric sensors, more focus is given on the development of fluorescent sensors because of the high sensitivity and their utility in monitoring intracellular Hg$^{2+}$ ions in live cells. The majority of the summarized sensors are based on the well-known sensing mechanisms...
like PET, ICT, ESIP, AIE, FRET, and excimer-monomer. The majority of the fluorescent sensors are either turn-off or turn-on, and there is need of more research on the designing of ratiometric sensors for Hg$^{2+}$. Also, there is need of more attention in the designing of sensors applicable in pure aqueous medium over a wide pH range. Despite high sensitivity, the commercialization of sensors for real-world samples detection required great efforts on improving the sensor performance and also on fabrication methods. Therefore, future research may be focused on integrating the fascinating color change shown by the sensors even at low concentration with smartphone and other portable devices for the on-site, real-time and cost-effective detection of Hg$^{2+}$. The paper chips, polymeric or other testing strips of sensors may be developed for the detection of Hg$^{2+}$. The sensing mechanisms should be properly investigated to provide appropriate future directions to optimize the structure and performance for the designing of sensors. The concepts from nano and supramolecular chemistry may also be incorporated in the designing of novel sensors with improve sensing performance and to minimize the interference from other analytes in complex biological samples. We believe this review will provide new directions for designing novel and cost-effective sensors for Hg$^{2+}$ with improved aqueous solubility, selectivity, and sensitivity.

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